

This is the peer reviewed version of the following article:

Comparing Haemophilus influenzae type b Conjugate Vaccine Schedules: Systematic Review and Meta-Analysis of Vaccine Trials / Low, N; Redmond, Sm; Rutjes, A; Martínez-González, Na; Egger, M; Di Nisio, M; Scott, P. - In: THE PEDIATRIC INFECTIOUS DISEASE JOURNAL. - ISSN 0891-3668. - 32:11(2013), pp. 1245-1256. [10.1097/INF.0b013e31829f0a7e]

Terms of use:

The terms and conditions for the reuse of this version of the manuscript are specified in the publishing policy. For all terms of use and more information see the publisher's website.

09/01/2026 11:27

Comparing *Haemophilus influenzae* type b Conjugate Vaccine Schedules: A Systematic Review and Meta-Analysis of Vaccine Trials

Nicola Low*, MD, Shelagh M. Redmond*, PhD, Anne W.S. Rutjes, PhD, Nahara A. Martínez-González, MSc, Matthias Egger, MD, Marcello di Nisio, PhD, and Pippa Scott, PhD

From the Institute of Social and Preventive Medicine University of Bern, Switzerland (NL, SR, AR, NM, ME, PS); Center for Aging Sciences (Ce.S.I.) (AR) and Department of Medical, Oral and Biotechnological Sciences (MdN), University 'G. d'Annunzio', Chieti, Italy; and the Department of Vascular Medicine, Academic Medical Center, Amsterdam, the Netherlands (MdN)

* Authors contributed equally

Corresponding author: Pippa Scott

Tel: +41 31 631 35 55; Fax: +41 31 631 3520

Email: pscott@ispm.unibe.ch

Conflicts of Interest and Source of Funding:

This project received funding from the World Health Organization and from the Swiss National Science Foundation (grant no. 138490). None of the authors has conflicts of interest to declare.

Key words: *Haemophilus influenzae* type B conjugate vaccine, vaccine schedules, systematic review, meta-analysis

Abbreviated title: Meta-Analysis of Hib Vaccine Schedules

Running head: Hib Vaccine Schedules

Abstract

Background: The optimal schedule and the need for a booster dose are unclear for *Haemophilus influenzae* type b (Hib) conjugate vaccines. We systematically reviewed relative effects of Hib vaccine schedules.

Methods: We searched 21 databases to May 2010 or June 2012 and selected randomized controlled trials (RCTs) or quasi-RCTs that compared different Hib schedules (three primary doses with no booster dose [3p+0], 3p+1 and 2p+1) or different intervals in primary schedules and between primary and booster schedules. Outcomes were clinical efficacy, nasopharyngeal carriage and immunological response. Results were combined in random-effects meta-analysis.

Results: Twenty trials from 15 countries were included; 16 used vaccines conjugated to tetanus toxoid (PRP-T). No trials assessed clinical or carriage outcomes. Twenty trials examined immunological outcomes and found few relevant differences. Comparing PRP-T 3p+0 with 2p+0 there was no difference in seropositivity at the 1.0µg/ml threshold by six months after the last primary dose (combined risk difference -0.02, 95%CI -0.10, 0.06). Only small differences were seen between schedules starting at different ages, with different intervals between primary doses, or with different intervals between primary and booster doses. Individuals receiving a booster were more likely to be seropositive than those at the same age who did not.

Conclusions: There is no clear evidence from trials that any 2p+1, 3p+0 or 3p+1 schedule of Hib conjugate vaccine is likely to provide better protection against Hib disease than other schedules. Until more data become available, scheduling is likely to be determined by epidemiological and programmatic considerations in individual settings.

Introduction

Haemophilus influenzae type b (Hib) conjugate vaccines have led to large reductions in the incidence of invasive Hib disease, including meningitis and pneumonia, in countries that include them into their routine immunization schedule.¹ Nevertheless, there are still more than eight million cases of severe Hib disease worldwide annually in children under five years.² Conjugate vaccines in use in 2012 contained Hib capsular polysaccharide (polyribosylribitol phosphate, PRP) conjugated to diphtheria CRM197 protein, an oligosaccharide conjugate (PRP-HbOC), meningococcal outer membrane protein (PRP-OMP) or, most commonly tetanus toxoid (PRP-T).¹

Countries are faced with decisions about optimal schedules for vaccines recommended for infants. The 2006 World Health Organization position paper on Hib conjugate vaccines, states that a three-dose schedule can be used with one to two months between doses, starting as young as six weeks.³ The position paper does not explicitly recommend a booster dose, but states that if given it should be at 12-18 months of age. In 2012, most countries using Hib vaccine used a three-dose primary schedule with no booster dose (3p+0 schedule). Some countries, mainly in Europe and the Americas, added a booster dose to the three-dose primary schedule (3p+1 schedule) while other countries, mainly in Europe, used schedules with two primary doses and a booster (2p+1 schedule).⁴ Variation in Hib vaccination schedules reflects not only differences in the historical scheduling of childhood vaccines, setting-specific epidemiology, existing health service infrastructure and co-administered vaccines, but also uncertainties about the optimal number of primary doses, the interval between doses in the primary schedule, and the need for a booster dose.⁵ Whilst the clinical efficacy of Hib conjugate vaccines has been summarized,⁶⁻⁹

there have been no systematic reviews summarizing immunological, carriage and clinical outcomes from trials making head-to-head comparisons of different Hib vaccine schedules. Here we systematically review the evidence from randomized controlled trials (RCTs) or quasi-randomized trials about the relative effects of 2p+0, 3p+0, 2p+1 and 3p+1 schedules and the effects of different timing of Hib conjugate vaccine doses.

Methods

The review process followed a protocol, which was completed before starting the review (Supplementary text 1). Minor amendments were made after the review started and these are recorded in the protocol document. We report here results for the head-to-head comparisons of Hib conjugate vaccine schedules described in the protocol. Comparisons of Hib schedules to no Hib vaccination will be reported elsewhere.

Study identification

The literature search covered 21 electronic databases from the earliest citation until May 2010. There were five databases of published articles (AIM, Cochrane Library, LILACs, IndMED, Medline), three trial registries, 11 vaccine manufacturer databases and two regulatory authority websites. Search strategies included terms for “Hib” and “conjugate vaccine” adapted for each search engine (Supplementary text 2). In June 2012 the Medline search was updated, using a filter to identify RCTs (2012 search only), and the AIM, CENTRAL, LILACs, and IndMED searches were updated using the 2010 search strategy. Eligible trial registrations found in the 2010 search were also checked for new publications in June 2012.

Study selection

Studies were considered eligible if they were randomized or quasi-randomized (e.g. allocated according to date of birth) and examined children vaccinated with PRP-T, PRP-OMP or PRP-HbOC at less than 6 years of age. Trials were eligible if they assigned participants to the following comparisons: 3p+0 vs. 2p+0; 3p+0 vs. 2p+1; 3p+1 vs. 2p+1; 3p+1 vs. 3p+0. We also included studies that compared different intervals between doses and different ages at the start of the primary schedule. We excluded studies where both the schedule and the PRP-conjugated molecule differed between available comparison groups so that no comparisons within the trial assessed the effect of schedule differences alone.

Outcomes included invasive Hib disease as a combined outcome or separate diagnoses of Hib meningitis; pneumonia due to any cause; Hib pneumonia; epiglottitis; nasopharyngeal carriage of Hib; seropositivity after vaccination or geometric mean concentration (GMC) of PRP antibody. Seropositivity was defined by IgG antibody levels measured by enzyme-linked immunoassay (ELISA) or Farr-type radio-assay at threshold values of 0.15µg/ml and 1.0µg/ml.¹⁰ Only systematically collected clinical outcomes were considered eligible.

Each title and abstract was screened for eligibility by independent two reviewers. The full texts of abstracts assessed by one or both reviewers to be potentially eligible were then screened for eligibility by two reviewers. Data were extracted on to a structured piloted form (available on request). Data were extracted by two independent reviewers and differences were resolved by consensus. Items extracted included trial characteristics, outcomes, potential sources of heterogeneity, and the risk of bias in individual trials.¹¹ The risk of bias was assessed by examining trial features including the adequacy of random sequence generation, adequacy of allocation concealment, the use of outcome assessor blinding, and the type of analysis.^{12, 13}

Analysis types included modified intention to treat (mITT), and per protocol (PP). Modified intention to treat is used to describe analyses that included all randomized (or assigned) participants who had outcome data available with the possible exclusion of those who received no doses of vaccine, and PP is used to describe those that additionally excluded individuals with other protocol violations. We did not contact authors to obtain additional information.

Analysis

We combined data statistically, where appropriate, using DerSimonian and Laird random-effects meta-analysis¹⁴ in STATA version 12 (StataCorp LP, College Station, TX, USA). Between-trial heterogeneity was described using the I^2 statistic, where values below 25% represent low heterogeneity, up to 50% moderate heterogeneity, up to 75% severe heterogeneity and more than 75%, very severe heterogeneity.¹⁵ Where multiple intervention groups (or “trial arms”) were available within a trial to make a comparison of two schedules, we compared the groups which were most similar except for the difference in schedule. The decision about intervention groups to compare was made by two senior reviewers (NL and PS) without reference to trial results. For immunological outcomes, and for both the 1.0ug/ml and 0.15ug/ml thresholds, we calculated the difference between groups in proportions seropositive (and 95% confidence intervals using the normal approximation to the sampling distribution of the difference) and reported the risk difference as a proportion. A risk difference of 0.08 would indicate that an additional 8% of individuals in the first comparison group were seropositive than in the second comparison group (e.g. 88% vs. 80%). Immunogenicity data were stratified according to the conjugated molecule (PRP-HbOC, -OMP or -T). We report 1.0 µg/ml threshold data in figures in preference to 0.15µg/ml threshold data because risk differences were generally larger at the higher threshold. We report GMC data where seropositivity data were not available. We did not assess the

presence of small trials biases using funnel plots or the Egger test because few trials were available for most analyses.

Results

The literature searches yielded a total of 4337 unique items; 4032 items from the 2010 database and 305 from reference lists or repeat database searches. Of these, 4299 items were excluded (Figure 1). The remaining 38 items referred to 20 randomized or quasi-randomized trials reporting on eligible comparisons and outcomes. Included studies are described in Table 1 and Supplementary Table 1.¹⁶⁻³⁴ The 20 trials were conducted in 15 countries in Africa, Asia, Europe, and North and South America. Sixteen trials used PRP-T, three used PRP-OMP, and two used PRP-HbOC. One trial used PRP-T in two trial groups and PRP-HbOC in two other groups (Chile1). Five trials did not report the number of individuals assigned to each intervention group. Where numbers were reported, a total of 6312 infants were assigned to intervention groups analyzed in this review: 661 infants to 2p+0 schedules, 1194 to 3p+0, 300 to 2p+1, and 4157 to 3p+1 schedules. The median number of participants in trials was 283 (range 54 - 1782).

Risk of bias in methods of included studies

Table 2 shows methodological features which could influence the risk of bias for the 20 trials. All trials individually assigned participants to intervention groups, and only one trial was judged to be quasi-randomized (USA3). Allocation concealment was assessed as adequate in two trials and inadequate in one trial. In 17 trials allocation concealment was not well enough described to be assessed. Outcome assessors (laboratory staff) were described as blinded in 11 of the 20 trials. Four trials reported mITT analyses (three of which also conducted PP analyses but only stated that results were similar to mITT results), nine reported PP analyses (two of which also

conducted mITT analyses but only stated that results were similar to PP results) and for seven trials it was not clear which analysis was reported.

Head-to-head comparisons between schedules

There were no eligible clinical or carriage outcome data from trials that compared different schedules of Hib vaccination. Twenty trials examined eligible schedule comparisons and presented seropositivity or GMC data. Nine of these provided data for comparisons of schedules with different numbers of doses in the primary or booster schedules and 14 of these provided data for comparisons of schedules with the same number of doses but different timings.

Supplementary Figures 1 and 2 show seropositivity ($\geq 0.15\mu\text{g/ml}$ and $\geq 1.0\mu\text{g/ml}$) for all trial arms used in eligible comparative analyses.

Number of doses in primary and booster schedules, immunological data

3p+0 vs. 2p+0 schedules

Seven trials provided data for this comparison (Chile1, Chile2, Guatemala, Netherlands, Niger, Sweden, USA2). Six examined PRP-T and two examined PRP-HbOC (one trial examined both). Six trials reported seropositivity (Chile1, Chile2, Guatemala, Netherlands, Niger, Sweden) and all trials reported GMC data.

Figure 2 shows the risk difference ($\geq 1.0\mu\text{g/ml}$) for seropositivity between groups receiving 3p+0 and 2p+0 schedules for trials where the interval between the last dose and blood draw was the same for both arms. In three trials examining PRP-T (Chile1, Niger, Sweden), neither the 2p nor the 3p schedule was consistently favored and heterogeneity was high (I^2 90% at the $1.0\mu\text{g/ml}$ threshold and 67% at the $0.15\mu\text{g/ml}$ threshold, shortly after the last primary dose). By six months after the last primary dose, there was no difference between the schedules at the $1.0\mu\text{g/ml}$ threshold (combined risk difference -0.02, 95%CI -0.10, 0.06) and no heterogeneity (I^2 0%).

Heterogeneity remained high six months after the last primary dose at the 0.15µg/ml threshold (I^2 75%).

One trial (Chile1) examined PRP-HbOC and presented seropositivity data. Point estimates favored the 3p group but the confidence interval included the null effect. The trial which reported only GMC (USA2) examined PRP-HbOC and compared a birth dose plus doses at 2 and 4 months of age to doses at 2 and 4 months of age. Two months after the last dose, the reported GMC in the 3p group (birth-dose group) was 0.93µg/ml (95%CI 0.48, 1.69) and 0.20µg/ml (95%CI 0.10, 0.29) in the 2p group.

3p vs. 2p+1 schedules

One trial (Sweden) using PRP-T provided data for this comparison. At 13 months of age (seven months after the 3p group received their last primary dose and one month after the 2p+1 group received their booster) the risk difference was -0.79 (95%CI -0.87, -0.71) at the 1.0µg/ml threshold, and -0.20 (95%CI -0.27, -0.13) at 0.15µg/ml, favoring the 2p+1 schedule.

3p+1 vs. 2p+1 schedules

Two trials using PRP-T provided data on seropositivity for this comparison (Netherlands, Sweden). Proportions seropositive one month after the booster vaccinations were high and similar in both groups. The combined risk difference was 0.01 (95%CI -0.03, 0.05) at the 1.0µg/ml threshold and 0.01 (95%CI -0.01, 0.02) at 0.15µg/ml, with moderate (I^2 56%) and low (I^2 24%) heterogeneity, respectively.

3p+1 vs. 3p schedules

Two trials examined PRP-T for this comparison (Canada2, Europe). One reported seropositivity data (Europe) and both reported GMC. At 13 months of age (one month after the 3p+1 group received their booster dose), the 3p+1 schedule resulted in higher seropositivity than the 3p

schedule at both the 1.0µg/ml (risk difference 0.59, 95%CI 0.52, 0.67) and 0.15µg/ml thresholds (risk difference 0.16, 95%CI 0.11, 0.22). One trial reported only GMC (Canada2). Multiple intervention groups in this trial were available for comparison and not all are presented here. At 16 months of age the intervention group which received a 3p schedule with a booster dose at 15 months of age achieved a GMC of 29.2µg/ml (95%CI 24.58, 36.43, Canada2) and a group which had received a 3p schedule with no booster dose by 16 months of age achieved a GMC of 0.32µg/ml (95%CI 0.25, 0.41, Canada2).

Age at start of primary schedule, immunological data

Eight trials compared schedules with the same number of doses, in which the first dose was given earlier or later (Belgium, Chile2, China1, China2, Gambia1, Gambia2, Netherlands, Turkey). Seven examined PRP-T, and one examined PRP-OMP (Gambia1). Seven trials reported seropositivity data and eight reported GMC. Seropositivity results at the 1.0µg/ml threshold are shown in Figure 3. Some schedule comparisons differed in both the age at first dose and in the interval between doses in the primary schedule. There were only small differences in seropositivity between schedules and heterogeneity was low. The combined risk difference one month after the last primary dose was 0.02 (95%CI -0.01, 0.05) at the 1.0µg/ml threshold, based on 3 trials (I^2 1%). It was 0.01 (95%CI 0.00, 0.02) at 0.15µg/ml based on 4 trials (I^2 0%). The trial which reported only GMC (Gambia2) compared PRP-T doses at 2 and 4 months to doses at 1 and 3 months of age. One month after the last dose of vaccine, the GMC was 0.41µg/ml (95%CI 0.28, 0.61) in infants receiving the first dose at 2 months and 0.26µg/ml (95%CI 0.19, 0.35) in the group with the earlier start. One study comparing a birth dose of PRP-HbOC plus doses at 2, 4 and 6 months of age with doses at 2, 4 and 6 months (USA2) concluded that antibody levels were not higher after a birth dose.

Interval between doses, immunological data

Longer vs. shorter interval in primary schedules

Five trials provided immunological data comparing longer and shorter intervals in the primary schedule (Belgium, France, Turkey, USA1, USA3). Four trials compared two-month intervals to one-month intervals (Belgium, France, Turkey, USA3); three used 3p schedules with PRP-T and reported both seropositivity and GMC data (Belgium, France, Turkey) and one used a 2p schedule with PRP-OMP and reported GMC data only (USA3). At the 1.0 µg/ml threshold, neither the two-month nor the one-month interval schedule was consistently favored but results were heterogeneous (Figure 4). At the 0.15 µg/ml threshold, no difference was seen between the schedules and heterogeneity was low: the combined risk difference one month after the last primary dose was 0.00 (95%CI -0.02, 0.02), I^2 0%. The trial using PRP-OMP (USA3) was quasi-randomized, using alternation for assignment of interventions. The mean age at first vaccination was older in the two-month-interval group than in the one-month-interval group (4.1 months and 3.2 months respectively). Age adjusted GMCs one month after the second vaccination were 3.95 µg/ml (95%CI 2.63, 5.92) in the two-month-interval group and 2.32 µg/ml (95%CI 1.48, 3.64) in the one-month-interval group. One trial compared 4-month intervals to two-month intervals using PRP-OMP (USA1), but results were difficult to interpret because the interval between vaccination and blood-sampling differed between the groups being compared.

Longer vs. shorter interval between primary and booster schedules

Seven trials examined PRP-T and provided seropositivity and GMC data (Canada1, Canada2, Canada3, Chile2, China1, Europe, France). There were no differences in seropositivity one month after the booster dose and little between-study heterogeneity. The combined risk difference was 0.00 (95%CI -0.01, 0.01) at the 1.0µg/ml threshold (Figure 5) and 0.00 (95%CI -0.01, 0.01) at 0.15µg/ml, with I^2 14% and I^2 0%, respectively.

Discussion

Immunological data in this systematic review showed few differences that were both consistent and clinically relevant between Hib conjugate vaccine schedules with two or three primary doses or between schedules with different intervals between doses. Participants who had received booster doses were more likely to be seropositive than those of the same age who had not. There is an absence of clinical outcome or nasopharyngeal carriage data in head-to-head comparisons of Hib schedules.

This study is, to our knowledge, the first systematic review to examine the evidence from head-to-head comparisons of different Hib conjugate vaccine schedules. The wide search means that relevant RCTs are unlikely to have been missed. We also attempted a detailed assessment of potential sources of heterogeneity and bias but many trials were not reported completely enough for the risk of bias to be assessed. A limitation identified by this review was the paucity of data on several outcomes and comparisons of interest. We did not include data from observational studies because well-conducted RCTs are at lower risk of bias than observational study designs,^{35, 36} and because observational studies have been summarized elsewhere.^{37, 38} The potential for bias does remain in many of the included trials, with allocation concealment,

blinding of outcome assessors and exclusions after randomization being key trial design features influencing the risk of bias within trials.³⁹ In particular, many trials in this review explicitly excluded some randomized individuals by conducting only a per protocol analysis. For some design features it is difficult to categorize the risk of bias if the design feature is poorly described. For example, an incomplete description of allocation concealment could be compatible with either a high or low risk of bias; if allocation was adequate, the risk of bias is low but if allocation concealment was not well conducted, bias might occur if it can be easily predicted which individuals are more or less likely to seroconvert. Incomplete descriptions for features such as blinding are less important when considering immunological results where outcomes are assessed by laboratory technicians. It is possible and even likely that outcome assessors were blinded, even if this was not reported. Even if the laboratory staff are not blinded, automated procedures are likely to reduce the risk of bias.

The immunological data from available trials do not clearly favor either a two-dose or a three-dose primary schedule. There were also no important differences in seropositivity for PRP-T schedules starting at either 2 vs. 3 months or PRP-OMP schedules starting at 1 vs. 2 months of age. Available clinical data show good protection against invasive Hib disease with 2p+0 schedules using PRP-OMP,⁴⁰ and with 3p+0 schedules using PRP-T or PRP-HbOC,⁴⁰⁻⁴⁴ when compared to no Hib vaccine and these data have been summarized several times.⁶⁻⁹ However, estimates of VE from different trials cannot be compared directly as evidence of equivalence or superiority of one particular schedule and there are too few trials for a network meta-analysis, which would allow such a comparison.^{45, 46}

Two-month intervals between doses in the primary schedule were not shown to be consistently more immunogenic than one month intervals. Meta-analyses either showed marked heterogeneity or showed little heterogeneity and no difference between two and one month intervals. It is challenging to draw conclusions about clinical efficacy based on immunological findings because the clinical relevance of Hib seropositivity levels and GMCs are not well established in general,¹⁰ and also because of differences in the schedules compared within each study other than the difference of interest. Data from an observational review found no strong evidence from cohort or case-control studies that the choice of intended intervals of one or two months between doses affects vaccine effectiveness,³⁸ but differences between the intended and actual schedules and other factors such as herd immunity in the population again add complexity to interpretation.⁵

A booster dose after a primary series of either two or three doses of Hib conjugate vaccine results in high levels of seropositivity. There was no evidence from trials that the age at which the booster dose is given, or the interval between the primary series and the booster dose affect the level of seropositivity. Seropositivity levels in children after a booster dose are much higher than in children who received the same primary schedule without a booster. The interval between the last vaccine dose and blood draw is, however, shorter in children receiving the booster than in those who received only the primary schedule and it is not clear if differences in antibody levels can be interpreted as differences in protection from Hib disease.¹⁰ This review was not designed to collect data about antibody persistence, and therefore caution should be taken when examining such data from this review. However, when data from individual groups in trials eligible for this review are plotted alongside each other (Supplementary figures 1 and 2), it can be seen that the proportion seropositive tends to be higher soon after a booster dose than

soon after the last primary dose, or several months after the last primary dose, particularly at the 1.0µg/ml threshold. Trials that assessed seropositivity more than one month after the last primary dose showed generally lower proportions seropositive than those assessing seropositivity one month after the last primary dose. In the one trial with long follow up after a booster dose, a high proportion of individuals remained seropositive at the 0.15µg/ml threshold years after the booster dose and a much lower proportion at the 1.0µg/ml threshold. These trends are in general agreement with studies which have found sustained antibody persistence after a booster dose.^{47,48} The UK experienced an increase in Hib cases several years after an initial decline in cases subsequent to the introduction of a 3p+0 schedule (2, 3, 4 months) alongside an early catch-up campaign. Cases again declined after two booster campaigns and the introduction of a routine booster dose to the vaccine schedule.⁴⁹ However, the situations in which a booster dose should be used remain unclear, and might relate to local epidemiology, co-administered vaccines, and the potential for natural boosting as well as other factors.^{50,51}

This review did not aim to examine the effects of co-administrated vaccines on Hib conjugate vaccine efficacy, which is best examined in trials comparing groups with different co-administered vaccines but with the same schedule. However, conclusions from our review about the relative effects of different schedules do not change when restricted to trials that co-administered aP or trials that co-administered wP. In analyses that included both trials in which whole cell pertussis vaccine (wP) was co-administered and trials in which acellular pertussis vaccine (aP) was co-administered, the relative effects of different schedules of Hib vaccine did not appear to change substantially between studies. However, owing to the limited availability of data in each analysis, this could not be formally assessed using statistical methods such as meta-regression. The observational review conducted simultaneously with our review found no strong

evidence from cohort studies that co-administration with aP reduced vaccine effectiveness, but two case-control studies conducted in the UK provided some evidence of a reduction.^{38, 51, 52} Further carefully conducted systematic reviews of RCTs, as well as observational data, could provide useful information about this and other questions about Hib vaccine scheduling.

Hib conjugate vaccine 2p+1, 3p+0 and 3p+1 schedules are all likely to provide protection against Hib disease and, until further data about the relative effects of different Hib vaccine schedules are available, the choice of schedule is likely to depend on the setting. For example, in settings where the burden of severe Hib disease lies with children under one year of age it might be more appropriate to provide three doses of Hib vaccine early in life. In settings where the disease burden occurs later, or where a resurgence of Hib cases is seen after the introduction of Hib vaccine, it might be advantageous to use a schedule where the third dose is given as a booster. Programmatic considerations are also likely to influence the choice of Hib vaccine schedule. Costs of vaccine administration are likely to be lower and vaccine coverage higher if vaccine administration is combined with other routine scheduled health visits. Additionally, most Hib vaccines are administered as combined vaccines, which means that the scheduling of the other co-administered vaccines must also be taken into account when choosing a Hib vaccine schedule.

Future decisions relating to Hib vaccination could be informed by well-conducted randomized controlled trials with head-to-head comparisons of schedules that collect data on clinical outcomes. Trials comparing schedules would need to be extremely large to provide sufficient statistical power to show difference between schedules, but trials of this type have been conducted for other vaccines.⁵³

Variation in the burden of disease, health infrastructure and scheduling of other vaccines create complexity in determining optimal vaccination schedules. Thus, information on the benefits of different vaccine schedules is essential if informed decisions are to be made. In this comprehensive systematic review, we highlight the absence of clinical and carriage data from trials comparing Hib vaccine schedules and scarce immunological data from such comparisons. We show there is no clear evidence from vaccine trials that any 2p+1, 3p+0 or 3p+1 schedule of Hib conjugate vaccine is likely to provide better protection against Hib disease than other schedules. Until additional data about the relative effects of different Hib vaccine schedules are available, the choice of Hib vaccination schedule is likely to be determined by the epidemiological and programmatic conditions in individual settings.

Acknowledgements

We thank the WHO secretariat and participants at expert meetings at WHO for their input, discussion and comments on this review. We also thank the World Health Organization and the Swiss National Science Foundation (grant no. 138490) for funding this project.

Disclaimer

The authors alone are responsible for the views expressed in this publication and they do not necessarily represent the decisions, policy or views of the World Health Organization.

References

1. Morris SK, Moss WJ, Halsey N. Haemophilus influenzae type b conjugate vaccine use and effectiveness. *Lancet Infect Dis*. 2008;8(7):435-443.
2. Watt JP, Wolfson LJ, O'Brien KL, et al. Burden of disease caused by Haemophilus influenzae type b in children younger than 5 years: global estimates. *Lancet*. 2009;374(9693):903-911.
3. World Health Organization. WHO position paper on Haemophilus influenzae type b conjugate vaccines. *Wkly Epidemiol Rec*. 2006;81(47):445-452.
4. World Health Organization. WHO Vaccine Preventable Diseases Monitoring System: Immunization schedules by antigen. Available at: http://apps.who.int/immunization_monitoring/en/globalsummary/scheduleselect.cfm. Accessed Jan 24, 2013.
5. Fitzwater SP, Watt JP, Levine OS, Santosham M. Haemophilus influenzae type b conjugate vaccines: considerations for vaccination schedules and implications for developing countries. *Hum Vaccin*. 2010;6(10):810-818.
6. Swingler G, Fransman D, Hussey G. Conjugate vaccines for preventing Haemophilus influenzae type B infections. *Cochrane Database Syst Rev*. 2007(2):CD001729.
7. Obonyo CO, Lau J. Efficacy of Haemophilus influenzae type b vaccination of children: a meta-analysis. *Eur J Clin Microbiol Infect Dis*. 2006;25(2):90-97.
8. Griffiths UK, Clark A, Gessner B, et al. Dose-specific efficacy of Haemophilus influenzae type b conjugate vaccines: a systematic review and meta-analysis of controlled clinical trials. *Epidemiol Infect*. 2012;140(8):1343-1355.

9. Theodoratou E, Johnson S, Jhass A, et al. The effect of Haemophilus influenzae type b and pneumococcal conjugate vaccines on childhood pneumonia incidence, severe morbidity and mortality. *Int J Epidemiol*. 2010;39 Suppl 1:i172-185.
10. Goldblatt D, Assari T. Immunological basis for immunization series. Module 9: Haemophilus influenzae type b. 2007. Available at: http://whqlibdoc.who.int/publications/2007/9789241596138_eng.pdf. Accessed Jan 24, 2013.
11. Juni P, Altman DG, Egger M. Systematic reviews in health care - Assessing the quality of controlled clinical trials. *BMJ*. 2001;323(7303):42-46.
12. Nuesch E, Trelle S, Reichenbach S, et al. The effects of excluding patients from the analysis in randomised controlled trials: meta-epidemiological study. *BMJ*. 2009;339:b3244.
13. Nuesch E, Reichenbach S, Trelle S, et al. The importance of allocation concealment and patient blinding in osteoarthritis trials: a meta-epidemiologic study. *Arthritis Rheum*. 2009;61(12):1633-1641.
14. DerSimonian R, Laird N. Meta-analysis in clinical trials. *Control Clin Trials*. 1986;7(3):177-188.
15. Higgins JP, Thompson SG. Quantifying heterogeneity in a meta-analysis. *Stat Med*. 2002;21(11):1539-1558.
16. Hoppenbrouwers K, Kanra G, Roelants M, et al. Priming effect, immunogenicity and safety of an Haemophilus influenzae type b-tetanus toxoid conjugate (PRP-T) and diphtheria-tetanus-acellular pertussis (DTaP) combination vaccine administered to infants in Belgium and Turkey. *Vaccine*. 1999;17(7-8):875-886.

17. Scheifele DW, Guasparini R, Lavigne P. A comparative study of PENTA vaccine booster doses given at 12, 15, or 18 months of age. *Vaccine*. 1999;17(6):543-550.
18. Scheifele DW, Halperin SA, Rubin E, et al. Safety and immunogenicity of a pentavalent combination vaccine (diphtheria, tetanus, acellular pertussis, polio, and haemophilus influenzae type B conjugate) when administered as a fourth dose at 15 to 18 months of age. *Hum Vaccin*. 2005;1(5):180-186.
19. Scheifele DW, Halperin SA, Ochnio JJ, Mozel M, Duarte-Monteiro D, Wortzman D. Immunologic considerations for the timing of the booster dose of 7-valent pneumococcal conjugate vaccine in young children. *Pediatr Infect Dis J*. 2007;26(5):387-392.
20. Lagos R, Valenzuela MT, Levine OS, et al. Economisation of vaccination against Haemophilus influenzae type b: a randomised trial of immunogenicity of fractional-dose and two-dose regimens. *Lancet*. 1998;351(9114):1472-1476.
21. Lagos R, Kotloff K, Hoffenbach A, et al. Clinical acceptability and immunogenicity of a pentavalent parenteral combination vaccine containing diphtheria, tetanus, acellular pertussis, inactivated poliomyelitis and Haemophilus influenzae type b conjugate antigens in two-, four- and six-month-old Chilean infants. *Pediatr Infect Dis J*. 1998;17(4):294-304.
22. Li RC, Li FX, Li YP, et al. Antibody persistence at 18-20 months of age and safety and immunogenicity of a booster dose of a combined DTaP-IPV//PRP approximately T vaccine compared to separate vaccines (DTaP, PRP approximately T and IPV) following primary vaccination of healthy infants in the People's Republic of China. *Vaccine*. 2011;29(50):9337-9344.

23. GlaxoSmithKline. Immunogenicity and safety of GlaxoSmithKline Biologicals' DTPa-IPV/Hib (Infanrix-IPV+Hib™) in infants. Results summary for study ID 112584. Available at: <http://www.gsk-clinicalstudyregister.com>. Accessed Jan 24, 2013.
24. Knuf M, Pantazi-Chatzikonstantinou A, Pfletschinger U, et al. An investigational tetravalent meningococcal serogroups A, C, W-135 and Y-tetanus toxoid conjugate vaccine co-administered with Infanrix hexa is immunogenic, with an acceptable safety profile in 12-23-month-old children. *Vaccine*. 2011;29(25):4264-4273.
25. European Medicines Agency (EMA). Hexavac, scientific discussion. Available at: http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Scientific_Discussion/human/000298/WC500074582.pdf. Accessed Jan 24, 2013.
26. Campbell H, Byass P, Ahonkhai VI, Vella PP, Greenwood BM. Serologic responses to an Haemophilus influenzae type b polysaccharide-Neisseria meningitidis outer membrane protein conjugate vaccine in very young Gambian infants. *Pediatrics*. 1990;86(1):102-107.
27. Mulholland EK, Byass P, Campbell H, et al. The immunogenicity and safety of Haemophilus influenzae type b-tetanus toxoid conjugate vaccine in Gambian infants. *Ann Trop Paediatr*. 1994;14(3):183-188.
28. Asturias EJ, Mayorga C, Caffaro C, et al. Differences in the immune response to hepatitis B and Haemophilus influenzae type b vaccines in Guatemalan infants by ethnic group and nutritional status. *Vaccine*. 2009;27(27):3650-3654.

29. Labadie J, Sundermann L, Rumke H, The DPT-IPV Hib vaccine study group. Multi-center study on the simultaneous administration of DPT-IPV and Hib PRP-T vaccines. Rijksinstituut voor Volksgezondheid en Milieu RIVM. 1996. Available at: <http://www.rivm.nl/bibliotheek/rapporten/124001003.html>. Accessed Jan 24, 2013.
30. Campagne G, Garba A, Schuchat A, et al. Response to conjugate *Haemophilus influenzae* B vaccine among infants in Niamey, Niger. *Am J Trop Med Hyg*. 1998;59(5):837-842.
31. Carlsson RM, Claesson BA, Selstam U, et al. Safety and immunogenicity of a combined diphtheria-tetanus-acellular pertussis-inactivated polio vaccine-*Haemophilus influenzae* type b vaccine administered at 2-4-6-13 or 3-5-12 months of age. *Pediatr Infect Dis J*. 1998;17(11):1026-1033.
32. Anderson EL, Decker MD, Englund JA, et al. Interchangeability of conjugated *Haemophilus influenzae* type b vaccines in infants. *JAMA*. 1995;273(11):849-853.
33. Lieberman JM, Greenberg DP, Wong VK, et al. Effect of neonatal immunization with diphtheria and tetanus toxoids on antibody responses to *Haemophilus influenzae* type b conjugate vaccines. *J Pediatr*. 1995;126(2):198-205.
34. Lenoir AA, Granoff PD, Granoff DM. Immunogenicity of *Haemophilus influenzae* type b polysaccharide-*Neisseria meningitidis* outer membrane protein conjugate vaccine in 2- to 6-month-old infants. *Pediatrics*. 1987;80(2):283-287.
35. Schulz KF, Altman DG, Moher D. CONSORT 2010 statement: updated guidelines for reporting parallel group randomised trials. *BMJ*. 2010;340:c332.
36. Sibbald B, Roland M. Understanding controlled trials. Why are randomised controlled trials important? *BMJ*. 1998;316(7126):201.

37. O'Loughlin RE, Edmond K, Mangtani P, et al. Methodology and measurement of the effectiveness of Haemophilus influenzae type b vaccine: systematic review. *Vaccine*. 2010;28(38):6128-6136.
38. Jackson C, Mann A, Mangtani P, Fine P. Effectiveness of Haemophilus influenzae type b (Hib) vaccines administered according to different schedules: systematic review and meta-analysis of observational data. Unpublished report; 2013.
39. Schulz KF, Chalmers I, Hayes RJ, Altman DG. Empirical evidence of bias. Dimensions of methodological quality associated with estimates of treatment effects in controlled trials. *JAMA*. 1995;273(5):408-412.
40. Santosham M, Wolff M, Reid R, et al. The efficacy in Navajo infants of a conjugate vaccine consisting of Haemophilus influenzae type b polysaccharide and Neisseria meningitidis outer-membrane protein complex. *N Engl J Med*. 1991;324(25):1767-1772.
41. Lagos R, Horwitz I, Toro J, et al. Large scale, postlicensure, selective vaccination of Chilean infants with PRP-T conjugate vaccine: practicality and effectiveness in preventing invasive Haemophilus influenzae type b infections. *Pediatr Infect Dis J*. 1996;15(3):216-222.
42. Mulholland K, Hilton S, Adegbola R, et al. Randomised trial of Haemophilus influenzae type-b tetanus protein conjugate vaccine [corrected] for prevention of pneumonia and meningitis in Gambian infants. *Lancet*. 1997;349(9060):1191-1197.
43. 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(9453):43-52.

44. Black SB, Shinefield HR, Fireman B, Hiatt R, Polen M, Vittinghoff E. Efficacy in infancy of oligosaccharide conjugate *Haemophilus influenzae* type b (HbOC) vaccine in a United States population of 61,080 children. The Northern California Kaiser Permanente Vaccine Study Center Pediatrics Group. *Pediatr Infect Dis J*. 1991;10(2):97-104.
45. Higgins JP, Whitehead A. Borrowing strength from external trials in a meta-analysis. *Stat Med*. 1996;15(24):2733-2749.
46. Lu G, Ades AE. Combination of direct and indirect evidence in mixed treatment comparisons. *Stat Med*. 2004;23(20):3105-3124.
47. Borrow R, Andrews N, Findlow H, et al. Kinetics of antibody persistence following administration of a combination meningococcal serogroup C and *haemophilus influenzae* type b conjugate vaccine in healthy infants in the United Kingdom primed with a monovalent meningococcal serogroup C vaccine. *Clin Vaccine Immunol*. 2010;17(1):154-159.
48. Southern J, McVernon J, Gelb D, et al. Immunogenicity of a fourth dose of *Haemophilus influenzae* type b (Hib) conjugate vaccine and antibody persistence in young children from the United Kingdom who were primed with acellular or whole-cell pertussis component-containing Hib combinations in infancy. *Clin Vaccine Immunol*. 2007;14(10):1328-1333.
49. Ladhani S, Slack MP, Heys M, White J, Ramsay ME. Fall in *Haemophilus influenzae* serotype b (Hib) disease following implementation of a booster campaign. *Arch Dis Child*. 2008;93(8):665-669.

50. Slack MP, Azzopardi HJ, Hargreaves RM, Ramsay ME. Enhanced surveillance of invasive *Haemophilus influenzae* disease in England, 1990 to 1996: impact of conjugate vaccines. *Pediatr Infect Dis J*. 1998;17(9 Suppl):S204-207.
51. McVernon J, Andrews N, Slack MP, Ramsay ME. Risk of vaccine failure after *Haemophilus influenzae* type b (Hib) combination vaccines with acellular pertussis. *Lancet*. 2003;361(9368):1521-1523.
52. McVernon J, Andrews N, Slack M, Moxon R, Ramsay M. Host and environmental factors associated with Hib in England, 1998-2002. *Arch Dis Child*. 2008;93(8):670-675.
53. Palmu AA, Jokinen J, Borys D, et al. Effectiveness of the ten-valent pneumococcal *Haemophilus influenzae* protein D conjugate vaccine (PHiD-CV10) against invasive pneumococcal disease: a cluster randomised trial. *Lancet*. 2013;381(9862):214-222.

Figure legends

Figure 1: Flow diagram of studies

Legend:

The 4032 items found in initial database searches include duplicates that were retrieved in 2 or more databases.

1 All 6 items relate to one trial where the only eligible outcomes were pneumonia and death and children were randomized to either Hib and pneumococcal conjugate vaccine or to a malaria vaccine. Difference between groups could be due to Hib or pneumococcal vaccines.

Figure 2: Comparison of seropositivity after 3 or 2 primary doses of Hib conjugate vaccine, 1.0µg/ml

Additional data for this comparison are not shown on this plot because the interval between the last dose of vaccine and blood sampling differed between the groups being compared within each study, making the comparison unfair. These data came from Chile² (at one or two months after the primary dose), Guatemala (at three or six months after the primary dose) and Netherlands (four or six months after the primary dose).

Legend:

Combined - Hib vaccine administered in the same syringe as pertussis containing vaccine;
separate - Hib vaccine administered by itself, either at the same time as or at different time from other vaccines; aP - acellular pertussis vaccine ; wP - whole-cell pertussis vaccine

* not specified as whole cell pertussis vaccine but assumed to be whole cell due to the year in which the trial was conducted

Figure 3: Comparison of seropositivity after late or early start of primary course of Hib conjugate vaccine, 1.0µg/ml

Additional data for this comparison are not shown on this plot because the interval between the last dose of vaccine and blood sampling differed between the groups being compared within each study, making the comparison unfair. These data came from China¹ (at 13 or 14 months after the primary dose), Gambia¹ (at 14 or 15 months after the primary dose) and Netherlands (four or six months after the primary dose).

Legend:

Combined - Hib vaccine administered in the same syringe as pertussis containing vaccine;
separate - Hib vaccine administered by itself, either at the same time as or at different time from other vaccines; aP - acellular pertussis vaccine ; wP - whole-cell pertussis vaccine

* not specified as whole cell pertussis vaccine but assumed to be whole cell due to the year in which the trial was conducted

Figure 4: Comparison of seropositivity after 2 or 1 month intervals between doses in the primary course of Hib conjugate vaccine, 1.0µg/ml

Additional data for this comparison are not shown on this plot because the interval between the last dose of vaccine and blood sampling differed between the groups being compared within each study, making the comparison unfair. These data came from France (at nine or 11 months after the primary dose)

Legend:

Combined - Hib vaccine administered in the same syringe as pertussis containing vaccine;
separate - Hib vaccine administered by itself, either at the same time as or at different time from other vaccines; aP - acellular pertussis vaccine ; wP - whole-cell pertussis vaccine

* Data for this trial reported unclearly at this time point and for this definition of seropositivity

Figure 5: Comparison of seropositivity after long or short intervals between primary and booster doses of Hib conjugate vaccine, 1.0µg/ml

Legend:

Combined - Hib vaccine administered in the same syringe as pertussis containing vaccine;

separate - Hib vaccine administered by itself, either at the same time as or at different times from other vaccines; aP - acellular pertussis vaccine ; wP - whole-cell pertussis vaccine

* not specified as whole cell pertussis vaccine but assumed to be whole cell due to the year in which the trial was conducted

Legend of Supplemental Digital Content

Supplementary Text 1: Study protocol

Supplementary Text 2: Search strategy

Supplementary Table1: Trials included in Hib conjugate vaccine review, detailed information

Supplementary Figure 1: Seropositivity after 2p, 3p, 2p+1 and 3p+1 schedules, 0.15µg/ml

Supplementary Figure 2: Seropositivity after 2p, 3p, 2p+1 and 3p+1 schedules, 1.0µg/ml

ACCEPTED

Table 1: Summary of included studies

Trial name and location	Conjugate vaccine	Allocation level	Schedules, age at administration in months		Number of participants randomized	Immunological outcomes reported
			Intended	Actual, mean (SD)		
Belgium ¹⁶	PRP-T	Individual	3, 4, 5*	3.0 (0.1)	49 [†]	Seropositivity
				4.0 (0.1)		GMC
				5.0 (0.2)		
			2, 4, 6*	2.1 (0.2)	54 [†]	
				4.0 (0.2)		
				5.9 (0.2)		
Canada ¹⁷	PRP-T	Individual	2, 4, 6 + b18	NR [‡]	82	Seropositivity
			2, 4, 6 + b15		85	GMC
			2, 4, 6 + b12		86	
Canada ² ¹⁸	PRP-T	Individual	3p+ b18	18.3 (0.3)	438	Seropositivity
			3p+ b17	17.4 (0.3)	450	GMC
			3p+ b16	16.4 (0.3)	449	
			3p+ b15	15.4 (0.3)	445	
				Primary: NR		
Canada ³ ¹⁹	PRP-T	Individual	2, 4, 6 +b18	18.3 (0.3)	167	Seropositivity
			2, 4, 6 +b15	15.3 (0.3)	168	GMC
				Primary: NR		
Chile ¹ ²⁰	PRP-T	Individual	2, 4, 6	NR	78	Seropositivity
			4, 6		79	GMC
	PRP-HBOC		2, 4, 6		78	
			4, 6		78	

Trial name and location	Conjugate vaccine	Allocation level	Schedules, age at administration in months		Number of participants randomized	Immunological outcomes reported
			Intended	Actual, mean (SD)		
Chile ²¹	PRP-T	Individual	3, 5, 7 + b12 [§] 2, 4, 6 + b12 [§]	NR	710 [¶]	Seropositivity GMC
China ¹²²	PRP-T	Individual	3, 4, 5 +b18-20 2, 3, 4 +b18-20	NR	264 264	Seropositivity GMC
China ²³	PRP-T	Individual	3, 4, 5** 2, 3, 4**	3.3 (0.3) 2.3 (0.3) dose 2-3:NR	324 330	Seropositivity GMC
Europe ²⁴ (Austria, Germany, Greece)	PRP-T (booster) ^{††}	Individual	3p +b13 ^{††} 3p +b12 ^{††}	NR 14.9 (3.2) primary NR	220 224	Seropositivity GMC
France ²⁵	PRP-T	Individual	2, 4, 6 + b15-17 2, 3, 4 + b15-17	NR	258 258	Seropositivity GMC
Gambia ¹²⁶	PRP-OMP	Individual	2, 4 1, 3	NR ^{§§}	95 99	Seropositivity GMC
Gambia ²⁷	PRP-T	Individual	2, 4 1, 3	NR	43 45	GMC
Guatemala ²⁸	PRP-T	Individual	2, 4, 6 7, 9	NR	325 106	Seropositivity GMC
Netherlands ²⁹	PRP-T	Individual	3, 4, 5 + b11 ^{¶¶} 6, 7 + b13 ^{¶¶}	NR	181 182	Seropositivity GMC

Trial name and location	Conjugate vaccine	Allocation level	Schedules, age at administration in months		Number of participants randomized	Immunological outcomes reported
			Intended	Actual, mean (SD)		
Niger ³⁰	PRP-T	Individual	1.5, 2.5, 3.5	Over all groups,	59	Seropositivity
			2.5, 3.5	mean (range):	62	GMC
				1.9 (0.9-2.8)		
				3.0 (2.1-5.1)		
				4.2 (3.0-6.8)		
Sweden ³¹	PRP-T	Individual	2, 4, 6 +b13	NR	118	Seropositivity
			3, 5 +b12		118	GMC
Turkey ¹⁶	PRP-T	Individual	3, 4, 5*	3.0 (0.1)	78 [†]	Seropositivity
				4.0 (0.2)		GMC
				5.1 (0.3)		
			2, 4, 6*	2.1 (0.2)	81 [†]	
				4.0 (0.3)		
			5.9 (0.3)			
USA1 ³²	PRP-OMP	Individual	2, 6	NR	36***	Seropositivity
			2, 4		39***	GMC
USA2 ³³	PRP-HbOC	Individual	2, 4, 6	NR ^{†††}	150 ^{†††}	GMC
			0, 2, 4, 6			
USA3 ³⁴	PRP-OMP	Individual	2-6, 4-8	4.1 (1.6)	27	GMC (adjusted)
				6.1 (1.6)		
			2-6, 3-7	3.2 (1.3)	27	
				4.2 (1.3)		

Legend

All times are in months of age unless otherwise stated. One reference is supplied for each trial in this table. A complete list of references for each trial can be found in Supplementary Table 1.

3p – 3-dose primary schedule where intended ages at vaccination not specified; +b – booster dose given at number of months indicated

DTaP - diphtheria, tetanus, acellular pertussis vaccine; DTwP - diphtheria, tetanus, whole cell pertussis vaccine; Hib – Haemophilus influenzae type b vaccine; IPV - inactivated polio vaccine; IQR - inter-quartile range; Men A and C vaccines - conjugate or polysaccharide meningococcal A and C vaccines; NR not reported; OPV - oral polio vaccine; p - primary course; PRP - polyribosylribitol phosphate; PRP-HbOC - polyribosylribitol phosphate conjugated to diphtheria toxin CRM 197; PRP-OMP - polyribosylribitol phosphate conjugated to outer membrane protein of Neisseria meningitidis; PRP-T - polyribosylribitol phosphate conjugated to tetanus toxoid; SD - standard deviation.

* Multiple groups provide this comparison for this trial. Results presented compare a group receiving PRP-T and DTaP in separate syringes at 3, 4, 5m to a group receiving PRP-T and DTaP in separate syringes at 2, 4, 6m. Another group receiving PRP-T at 3, 4, 5m in the same syringe as DTaP.

† Number receiving vaccine; number randomized not reported

‡ Ages not stated but the following information is given for the booster doses: “The intended schedule of immunization was met for each child with single exceptions at 15 months (one week late) and 18 months (2 weeks late)”

§ Multiple groups provide this comparison for this trial. Results presented compare a group receiving PRP-T at 3, 5, 7m and DTaP combined with IPV at 2, 4, 6m to a group receiving PRP-T at 2, 4, 6m and DTaP combined with IPV at 2, 4, 6m in another limb. Other groups receiving PRP-T at 3, 5, 7m either received OPV instead of IPV, or had DTaP and IPV given as separate injections. The other group receiving PRP-T at 2, 4, 6m received PRP-T in the same syringe as DTaP and IPV

¶ Number randomized to each group not reported. 710 infants randomized to five groups (not all included here)

|| Multiple groups provide this comparison for this trial. Results presented compare a group receiving PRP-T, IPV and DTaP in the same syringe at 3, 4, 5m to a group receiving PRP-T, IPV and DTaP in the same syringe at 2, 3, 4m. Another group receiving PRP-T at 3, 4, 5m received DTaP and IPV separately at the same time (i.e. 3 separate syringes).

** Multiple groups provide this comparison for this trial. Results presented compare a group receiving PRP-T, IPV and DTaP in the same syringes at 3, 4, 5m to a group receiving PRP-T, IPV and DTaP in the same syringes at 2, 3, 4m. Another group receiving PRP-T at 2, 3, 4m received DTaP in the same syringe and IPV at the same time but in a separate syringe.

†† Type of conjugate vaccines for the primary series was not specified in this trial.

‡‡ Multiple groups exist for the 3p + b12 schedule in this trial. Results presented compare a group receiving 3p then Meningococcal ACWY conjugate vaccine at 12m and PRP-T at 13m to a group receiving 3p then PRP-T at 12 months.

§§ Ages not stated but the following information is given: “Full compliance with the vaccination schedule and blood sampling was achieved by 85 infants in group A (immunized with two doses of vaccine at 1 and 3 months) and by 56 in group B (immunized at 2 and 4 months).”

¶¶ Multiple groups provide this comparison for this trial. Results presented compare a group receiving PRP-T at 3, 4, 5 + b11m and DTwP combined with IPV as a separate injection from PRP-T at 3, 4, 5 + b11m to a group receiving PRP-T at 6, 7 + b13m and DTwP combined with IPV (not with PRP-T) at 3, 4, 5 + b11m. The other group receiving PRP-T at 3, 4, 5 + b11m received PRP-T in the same syringe as DTwP and IPV

|| Ages not stated but most doses were given on time: "805 injections were administered. Seven injections were given 1 to 6 days out of time range, 2 injections were given >1 month out of time range"

*** Number analyzed; number of randomized or immunized children not reported

††† The group receiving 2, 4, 6 PRP-HbOC received the 3rd dose at a mean age of 6.7 months. Other groups and doses not reported.

‡‡‡ Total recruited, randomized and immunized; numbers per group not reported

ACCEPTED

Table 2: Methodological features of trials

Study, vaccine (manufacturer)	Adequate randomization sequence generation	Adequate randomization allocation concealment	Blinding of patient or parent to exposure status	Blinding of outcome assessors (immunological outcomes)	Blinding of other persons	Modified Intention to treat or per protocol analyses, immunological outcomes
Belgium ¹⁶	Unclear, randomization list but generation not reported	Unclear, not reported. Allocated “according to a randomization list and following chronological order of enrolment in the trial”	No, not possible due to schedule differences	Yes	Unclear, not reported	mITT (PP performed and “similar”)
Canada ¹⁷	Yes, computer-generated list of random numbers	Unclear, sealed, serially- numbered envelopes that were opened in sequence, but not stated if opaque	No, not possible due to schedule differences	Unclear, authors refer to “code-numbered samples”, but no explicit description of blinding	Not reported	mITT
Canada ²¹⁸	Unclear, not reported	Unclear, not reported	No, not possible due to schedule differences	Unclear, trial described as open-label	Unclear, trial described as open-label	PP (mITT performed and “similar”)

Study, vaccine (manufacturer)	Adequate randomization sequence generation	Adequate randomization allocation concealment	Blinding of patient or parent to exposure status	Blinding of outcome assessors (immunological outcomes)	Blinding of other persons	Modified Intention to treat or per protocol analyses, immunological outcomes
Canada ¹⁹	Unclear, not reported	Unclear, not reported	Parents partially blinded. Not blinded to age at vaccination	Unclear, not reported	Unclear, not reported	Unclear
Chile ¹²⁰	Unclear not reported how "list of correlative numbers" generated	Unclear, not well reported	No, not possible due to schedule differences	Yes	Vaccinators not blinded	Unclear
Chile ²²¹	Unclear, does not report how "list of ... study numbers, in blocks of 10" generated	Unclear, not reported	No, not possible due to schedule differences	Yes	Unclear, trial reported to be "open"	mITT (PP analysis conducted with "identical results")
China ¹²²	Unclear, not reported	Unclear, not reported	No, not possible due to schedule differences	Yes	Unclear, trial reported to be "open"	Unclear

Study, vaccine (manufacturer)	Adequate randomization sequence generation	Adequate randomization allocation concealment	Blinding of patient or parent to exposure status	Blinding of outcome assessors (immunological outcomes)	Blinding of other persons	Modified Intention to treat or per protocol analyses, immunological outcomes
China ²³	Unclear, not reported	Unclear, not reported	No, not possible due to schedule differences	Unclear, trial reported to be "open"	Unclear, trial reported to be "open"	PP
Europe ²⁴ (Austria, Germany, Greece)	Unclear, not reported	Unclear, not reported	No, not possible due to schedule differences	Unclear, trial reported to be "open"	Unclear, trial reported to be "open"	PP
France ²⁵	Unclear, not reported	Unclear, not reported	Unclear, but unlikely due to schedule differences	Unclear, trial reported to be "open"	Unclear, trial reported to be "open"	PP (mITT performed and reported to be 'consistent with PP)
Gambia ¹²⁶	Unclear, "using a system of random numbers"	Yes, on site computer system, with automated and consecutive allocation of vaccination codes corresponding to coded vials.	No, not possible due to schedule differences	Yes	Field workers not blinded	PP

Study, vaccine (manufacturer)	Adequate randomization sequence generation	Adequate randomization allocation concealment	Blinding of patient or parent to exposure status	Blinding of outcome assessors (immunological outcomes)	Blinding of other persons	Modified Intention to treat or per protocol analyses, immunological outcomes
Gambia ²⁷	Unclear, "system of random numbers incorporated into a computerized call program"	Yes, on site computer system, with automated and consecutive allocation of vaccination codes corresponding to coded vials.	No, not possible due to schedule differences	Yes, laboratory staff blinded	Unclear, not reported	Unclear
Guatemala ²⁸	Yes, computer generated random numbers	Unclear, sequentially numbered sealed envelopes. Not stated if opaque or if linked to individuals before opening	Unclear, trial reported to be "open"	Unclear, trial reported to be "open"	Described as "open study"	Unclear
Netherlands ²⁹	Yes, computer generated list	Unclear, not reported	Unclear, not reported	Yes	Unclear, not reported	PP
Niger ³⁰	Unclear, not reported	Unclear, not reported	Unclear, not reported	Unclear, "assays were performed on coded specimens" but no additional description given.	Those who assess adverse events were blinded	Unclear

Study, vaccine (manufacturer)	Adequate randomization sequence generation	Adequate randomization allocation concealment	Blinding of patient or parent to exposure status	Blinding of outcome assessors (immunological outcomes)	Blinding of other persons	Modified Intention to treat or per protocol analyses, immunological outcomes
Sweden ³¹	Unclear, "randomly assigned, in blocks of 10", but sequence generation not reported	Unclear, not reported	No, not possible due to schedule differences	Yes	Unclear, trial reported as "open"	PP
Turkey ¹⁶	Unclear, randomization list but generation not reported	Unclear, not reported. Allocated "according to a randomization list and following chronological order of enrolment in the trial"	No, not possible due to schedule differences	Yes	Unclear, not reported	mITT (PP performed and "similar")
USA1 ³²	Unclear, site-specific randomization lists but generation not reported	Unclear. Vials supplied only with a code number but not reported if vials were identical in appearance. Unclear who randomized the infants.	Yes, placebo used	Yes	"Investigators who enrolled, interviewed, or evaluated subjects or parents were blinded to study group assignment"	PP

Study, vaccine (manufacturer)	Adequate randomization sequence generation	Adequate randomization allocation concealment	Blinding of patient or parent to exposure status	Blinding of outcome assessors (immunological outcomes)	Blinding of other persons	Modified Intention to treat or per protocol analyses, immunological outcomes
USA2 ³³	Unclear, not reported	Unclear, not reported	Yes	Yes	Vaccinators not blinded. Those assessing safety were blinded.	Unclear
USA3 ³⁴	No, alternation	No, alternation	No, not possible due to schedule differences	Unclear, not reported	Unclear, not reported	PP

Legend:

ITT - intention-to-treat analysis - analysis where no randomized individuals are excluded; mITT- modified intention-to- treat analysis - similar to an ITT analysis but with some modifications to inclusion criteria such as excluding those who did not receive a first dose of vaccine; NA - not applicable because eligible outcomes not reported in this trial; PP - per protocol analysis, analysis where individuals with protocol violations (such as not receiving the intended vaccination schedule) are excluded

All assessments based on information contained in published articles or pre-publication manuscripts. Authors of individual trials were not contacted for information on methodological features. One reference is supplied for each trial in this table. A complete list of references for each trial can be found in Supplementary Table 1.

ACCEPTED

Supplementary Table 1: Trials included in Hib conjugate vaccine review, detailed information

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunological
Belgium ¹							
Location: Belgium Recruitment dates: October 1994 to March 1995 Hib vaccine: PRP-T, Act-HIB, Pasteur Mérieux Connaught Pertussis vaccine: aP (2 component), brand name not stated, Pasteur Mérieux, Connaught Funding: Pasteur Mérieux Connaught	Inclusion criteria: healthy infants, Belgian, aged 2 months (22 weeks) with informed written consent from the parents or legal guardian Exclusion criteria: none reported	A: 3, 4, 5 +b12-14 B: 3, 4, 5 +b12-14 C: 2, 4, 6 Additional information: A: DTaP at 3, 4, 5, 12-14 combined B: DTaP at 3, 4, 5, 12- 14m, separate C: DTaP at 2, 4, 6, separate	N= 54* Mean age at randomization (SD): 2 (0.5) Mean age at vaccination (SD): 1 st dose: 3.0 (0.1) 2 nd dose: 4.0 (0.1) 3 rd dose: 5.0 (0.2) Booster:14.0 (0.7) Gender (M/F): 32/22 (59% M)	N= 49* Median age at randomization (SD): 2 (0.5) Mean age at vaccination (SD): 1 st dose: 3.0 (0.1) 2 nd dose: 4.0 (0.1) 3 rd dose: 5.0 (0.2) Booster: 13.8 (0.6) Gender (M/F): 27/25 (50% M)	N= 54* Mean age at randomization (SD): 2 (0.5) Mean age at vaccination (SD): 1 st dose: 2.1 (0.2) 2 nd dose: 4.0 (0.2) 3 rd dose: 5.9 (0.2) No booster Gender (M/F): 22/32 (41% M)		✓
Canada ¹²							
Location: Canada Recruitment dates: Not stated Hib vaccine (booster): PRP-T, PENTA (combined DPT- IPV/PRP-T), Pasteur Mérieux Connaught Pertussis vaccine: Not stated if wP or aP, assume wP given trial date, PENTA, Pasteur Mérieux Connaught Funding: Pasteur Mérieux Connaught	Inclusion criteria: healthy children, written consent from a parent or legal guardian, completed a study of primary immunization with a DPT- IPV/PRP-T combination vaccine Exclusion criteria: any contraindication to receipt of PENTA or MMR vaccines, impairment of immune responsiveness, prior infection with any of the agents targeted by PENTA or MMR vaccines; receipt of any other DPT, polio or Hib vaccine apart from in the earlier study; receipt of blood products within 3 months, receipt of any other vaccine within 2 weeks	A: 2, 4, 6 + b18 B: 2, 4, 6 + b15 C: 2, 4, 6 + b12 Additional information: All children had previously received 3 doses of PENTA (combined DPT- IPV/PRP-T) at 2, 4, 6 months and received a PENTA booster in this study. All received MMR vaccine at 12 months.	N= 82 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR [†] Gender (M/F): NR	N= 85 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR [†] Gender (M/F): NR	N= 86 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR [†] Gender (M/F): NR		✓

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunologica
Canada2 ³⁻⁷							
Location: Canada Recruitment dates: Study performed in 2000 to 2001 Hib vaccine: PRP-T, Act-HIB, Sanofi Pasteur Pertussis vaccine: aP (5 component) Quadracel, Sanofi Pasteur. Funding: Sanofi Pasteur	Inclusion criteria: healthy toddlers, 12 months of age, who had completed a routine three-dose primary series with DTaP-IPV//PRP-T combination vaccine (Pentacel) by eight months of age Exclusion criteria: history of neurologic disorder, confirmed pertussis, chronic underlying disorder; known or suspected hypersensitivity to any component of the study vaccine; impaired immunologic function or receipt of immunosuppressive therapy or immunoglobulins; and prior immunization with a fourth dose of diphtheria, tetanus, pertussis, H. influenza type b conjugate, or poliovirus vaccine)	A: 3p +b18 B: 3p +b17 C: 3p +b16 D: 3p +b15 Additional information: Primary and booster doses were combined DTaP-IPV and PRP-T vaccines. Varicella and MMR vaccines offered upon study entry at 12 months of age to those who had not received them.	N= 438 Mean age at randomization (SD): NR Mean age at vaccination (SD): Primary: NR Booster: 18.3 (0.3) Gender (M/F): 213/225 (47% M) Schedule D: N= 445 Mean age at randomization (SD): NR Mean age at vaccination (SD): Booster: 15.4 (0.3) Gender (M/F): 215/230 (48% M)	N= 450 Mean age at randomization (SD): NR Mean age at vaccination (SD): Primary: NR Booster: 17.4 (0.3) Gender (M/F): 222/228 (49% M)	N= 449 Mean age at randomization (SD): NR Mean age at vaccination (SD): Primary: NR Booster: 16.4 (0.3) Gender (M/F): 211/238 (47% M)	✓	
Canada3 ⁸							
Location: Canada Recruitment dates: 2003 Hib vaccine: PRP-T, Pentacel, Sanofi Pasteur Pertussis vaccine: aP (5 component) Pentacel, Sanofi Pasteur Funding: Wyeth Pharmaceuticals	Inclusion criteria: healthy children who had completed a study of 3-dose primary PCV7 vaccination, with a final blood sample for serology obtained at 7–8 months of age, informed consent from parents Exclusion criteria: none stated.	A: 2, 4, 6 +b18 B: 2, 4, 6 +b15 Additional information: All received DTaP-IPV combined with Hib and offered routine MMR at 12 months. A and B: primary PCV doses either 2, 4, 6 or 3, 5, 7. Booster doses of PCV given at the same time but separately from Hib.	N= 167 Mean age at randomization based on time beyond birthday (SD): 6.3 (0.3) Mean age at vaccination (SD): Primary: NR Booster: 18.3(0.3) Gender (M/F): 98/69 (59% M)	N= 168 Median age at randomization based on time beyond birthday (SD): 3.3 (0.3) Mean age at vaccination (SD): Primary: NR Booster: 15.3 (0.3) Gender (M/F): 100/68 (59.5% M)		✓	

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunological
Chile1 ^{9, 10}							
Location: Chile Recruitment dates: October to December, 1995 Hib vaccine: PRP-T, ActHib, Pasteur Mérieux Connaught PRP-HbOC, HibTiter, Wyeth-Lederle Pertussis vaccine: Funding: Children's Vaccine Initiative (WHO, Geneva, Switzerland), National Institute of Allergy and Infectious Disease	Inclusion criteria: healthy infants born at full term with a birth weight of 2500 g or more, written, informed consent from parent or guardian Exclusion criteria: contraindication to receiving DTP vaccine, major chronic or congenital diseases, or known immunological disorders	A: 2, 4, 6 (PRP-T) C: 4, 6 (PRP-T) B: 2, 4, 6 (PRP-HbOC) D: 4, 6 (PRP-HbOC) Additional information: PRP given to all at 12 months of age (results after PRP not eligible for this review. Fractional dose groups also not eligible	N= 78 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Gender (M/F): NR Schedule D: N= 78 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Gender (M/F): NR	N= 79 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Gender (M/F): NR	N= 78 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Gender (M/F): NR		✓
Chile2 ¹¹							
Location: Chile Recruitment dates: December 20, 1995 to April 2, 1996 Hib vaccine: PRP-T, ActHIB, Pasteur Mérieux Connaught Pertussis vaccine: aP (2 component), brand name not stated, Pasteur Mérieux Connaught Funding: Pasteur Mérieux Connaught	Inclusion criteria: healthy 2 month-old infants (±4 weeks) planning to receive primary care at the selected health centres for the complete study period, informed consent from parents or guardian Exclusion criteria: known or suspected disease; previous vaccination against diphtheria, tetanus, pertussis, Hib or polio; <37 weeks of gestation; birth weight <2500g; known contraindication to receiving DTP, PRP-T or IPV vaccines	A: 3, 5, 7 +b12 B: 3, 5, 7 +b12 C: 3, 5, 7 +b12 D: 2, 4, 6 +b12 (separate) E: 2, 4, 6 +b12 (combined) Additional information: All children received MMR and DTaP combined with Hib vaccine at 12 months. A, B, C, D, E: received DTaP at 2, 4, 6 B, C, D, E: received IPV at 2, 4, 6 (B separate, others combined with DTaP), OPV at 7, 13 A: OPV at 2, 4, 6, 13	N= NR(710 total in study)* Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Gender (M/F): NR. Schedule D: N= NR(710 total in study)* Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Gender (M/F): NR.	N= NR(710 total in study)* Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Gender (M/F): NR. Schedule E: N= NR(710 total in study)* Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Gender (M/F): NR.	N= NR(710 total in study)* Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Gender (M/F): NR.	✓	✓

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunologica
China ¹²⁻¹⁴							
Location: China Recruitment dates: NR Hib vaccine: PRP-T, Pentacel, Sanofi Pasteur Pertussis vaccine: aP (2 component) in combined schedules) Pentaxim, Sanofi Pasteur aP (1 component) in separate schedule, brand name not stated, Wuhan Institute of Biological Products Funding: Sanofi Pasteur	Inclusion criteria: children who had completed the primary vaccination study and had informed consent from parents or legal representatives Exclusion criteria: participation in another clinical trial in the 4 weeks preceding the trial inclusion, immunodeficiency, immunosuppressive therapy, hypersensitivity to vaccine components, chronic illness; receipt of blood products	A: 3, 4, 5 +b18-20 (combined) B: 3, 4, 5 +b18-20 (separate) C: 2, 3, 4 +b18-20 (combined) Additional information: A and C: DTaP-IPV combined with Hib B: DTaP, Hib, IPV separately 3, 4, 5, 18-20	N= 264 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall gender based on N=792 (M/F): 393-444/348- 399 (49.6–56% M).	N= 264 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall gender based on N=792 (M/F): 393-444/348- 399 (49.6–56% M).	N= 264 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall gender based on N=792 (M/F): 393-444/348- 399 (49.6–56% M).	✓	✓
China ^{15, 16}							
Location: China Recruitment dates: Study period: March 24 to November 19, 2010 Hib vaccine: PRP-T, Infanrix-Hib or Infanrix- IPV+Hib, GlaxoSmithKline Pertussis vaccine: aP (3 component), Infanrix-Hib or Infanrix-IPV+Hib, GlaxoSmithKline Funding: GlaxoSmithKline	Inclusion criteria: healthy infants 60-90 days old, born after a gestation period of 36 to 42 weeks, written informed consent from the parents Exclusion criteria: previous or intercurrent diphtheria, tetanus, pertussis, poliomyelitis and/or Hib disease or vaccination, current febrile illness or axillary temperature > 37.0°C or other moderate to severe illness within 24 hours of study vaccine administration	A: 3, 4, 5 (DTaP-IPV combined) B: 2, 3, 4 (DTaP-IPV combined) C: 2, 3, 4 (DTaP combined, IPV separate) Additional information:	N= 324 Mean age at randomization (SD): NR Mean age vaccination (SD): 3.3 (0.3) Gender (M/F): 147/177 (45.4% M).	N= 330 Mean age at randomization (SD): NR Mean age at vaccination (SD): 2.3 (0.3) Gender (M/F): 155/175 (47% M).	N= 330 Mean age at randomization (SD): NR Mean age at vaccination (SD): 2.3 (0.3) Gender (M/F): 141/189 (43% M).	✓	✓

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunological
Europe ¹⁷⁻²²							
Location: Austria, Germany, Greece Recruitment dates: August 2007 to October 2008 Hib vaccine: Booster: PRP-T, Infanrix-hexa; GlaxoSmithKline Pertussis vaccine: aP (3 component), Infanrix-hexa, GlaxoSmithKline Funding: GlaxoSmithKline	Inclusion criteria: healthy children between 12 and 23 months, documented evidence of 3-dose primary vaccination with DTaP, hepatitis B, IPV and Hib vaccines completed at least 180 days previously Exclusion criteria: immunosuppression, previous receipt of any meningococcal vaccine or booster vaccination against diphtheria, tetanus, pertussis, hepatitis B, poliomyelitis or Hib, a past history of disease due to meningococcus, or receipt of blood products	A: 3p ⁺ +b13 B: 3p ⁺ +b12 C: 3p ⁺ +b12 (MenACWY-TT, separate at 12) D: 3p ⁺ Additional information: A: MenACWY-TT at 12 months. DTaP combined with Hib at 13 months B: MenACWY-TT at 13 months. DTaP combined with Hib at 12 months C: MenACWY-TT, separate at 12 months, DTaP combined with Hib at 12 months D: MenC conjugate at 12 months	N= 220 Mean age at randomization (SD): NR Mean age at vaccination (SD): Booster dose: 15(3.3) Gender (M/F): 114/106 (51.8% M) Schedule D: N= 127 Median age at randomization: NR Mean age at vaccination (SD): Booster dose: 14.6(3.0) Gender (M/F): 66/61 (52% M)	N= 224 Median age at randomization (SD): NR Mean age at vaccination (SD): Booster dose: 14.9(3.17) Gender (M/F): 105/119 (46.9% M)	N= 224 Median age at randomization (SD): NR Mean age at vaccination (SD): Booster dose: 14.6(3.01) Gender (M/F): 113/109 (50.9% M)	✓	✓

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunologica
France ^{23, 24}							
Location: France	Inclusion criteria: healthy Infants already enrolled in the trial initiated for the investigational vaccine and who had received primary immunisation under schedules 2, 4, 6 and 2, 3, 4 in the study Exclusion criteria: none stated	A: 2, 4, 6 + b15-17	N= 258	N= 258		✓	
Recruitment dates: 1995 to 1996		B: 2, 3, 4 + b15-17	Mean age at randomization (SD): NR	Median age at randomization (SD): NR			
Hib vaccine: PRP-T, Hexavac, Aventis Pasteur		Additional information: DTaP-HepB-IPV combined with Hib at each dose	Mean age at vaccination (SD): NR	Mean age at vaccination (SD): NR			
Pertussis vaccine: aP (2 component), Hexavac, Aventis Pasteur.			Gender (M/F): NR	Gender (M/F): NR			
Funding: Not stated, likely Aventis Pasteur							
Gambia ^{1 25-28}							
Location: The Gambia	Inclusion criteria: children living in the area of the health center, informed consent from mothers Exclusion criteria: none stated	A: 2, 4	N= 95	N= 99	N= 90	✓	
Recruitment dates: January 1 to December 31, 1985		B: 1, 3	Mean age at randomization (SD): NR	Mean age at randomization (SD): NR	Mean age at randomization (SD): NR		
Hib vaccine: PRP-OMP, PedvaxHib, Merck Sharp & Dohme		C: No doses	Mean age at vaccination (SD): NR	Mean age at vaccination (SD): NR	Mean age at vaccination (SD): no Hib		
Pertussis vaccine: Not given as part of trial. Not stated if wP or aP, assume wP given trial date. No brand name or manufacturer stated		Additional information: Other routine EPI vaccinations received but not as part of study.BCG and oral polio vaccines at 1 month of age and DTP and oral polio vaccines at 2, 3, and 4 months. Assume DTP given separately from Hib	Gender (M/F): NR	Gender (M/F): NR	Gender (M/F): NR		
Funding: Merck Sharp & Dohme		C: No control vaccine					

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunological
Gambia ²⁹							
Location: The Gambia	Inclusion criteria: Not stated	A: 2, 4	N= 43	N= 45	N= 40		✓
Recruitment dates: 1990	Exclusion criteria: none stated	B: 1, 3	Mean age at randomization (SD): NR	Mean age at randomization (SD): NR	Mean age at randomization (SD): NR		
Hib vaccine: PRP-T, ActHib, Pasteur Mérieux		C: No doses	Mean age at vaccination (SD): NR	Mean age at vaccination (SD): NR	Mean age at vaccination (SD): NR		
Pertussis vaccine: Not given as part of trial. Not stated if wP or aP, assume wP given trial date. No brand name or manufacturer stated		Additional information: All children had EPI routine vaccination (not specified). Assume DTP separate from Hib	Gender (M/F): NR	Gender (M/F): NR	Gender (M/F): NR		
Funding: Pasteur Mérieux							
Guatemala ³⁰							
Location: Guatemala	Inclusion criteria: healthy infants ≥6 weeks of age	A: 2, 4, 6	N= 325 [§]	N= 106 [§]			✓
Recruitment dates: March 1998 to August 1999	Exclusion criteria: known allergic reaction to any of the vaccine components, immunodeficiency, major congenital defects, serious illness, seizure disorders, history of blood product transfusions, or previous immunizations (except oral polio or Bacillus Calmette-Guerin vaccine)	B: 7, 9 (+b12)	Mean age at randomization (SD): NR	Median age at randomization (SD): NR			
Hib vaccine: PRP-T, Hiberix, GlaxoSmithKline		Additional information: All children had OPV at 2, 4, 6 and MMR at 9-12.	Mean age at vaccination (SD): NR	Mean age at vaccination (SD): NR			
Pertussis vaccine: wP (combined schedule), Tritanrix, GlaxoSmithKline wP (separate schedule), Brand name and manufacturer not clearly stated		A: Hib combined with DTwP and HepB B: DTwP at 2, 4, 6months. HepB given separately from Hib at 7, 9 months. Also received Hib and HepB vaccines at 12 months but no data provided after 12 month dose	Gender (M/F): 238/176 (57.5% M)	Gender (M/F): 56/50 (53% M)			
Funding: GlaxoSmithKline							

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunological
Netherlands ³¹							
Location: The Netherlands	Inclusion criteria: children born in February and March 1993, living in the Rotterdam cluster or in Apeldoorn, written informed consent by the parents Exclusion criteria: None stated	A: 3, 4, 5 +b11 (DTwP-IPV combined)	N=180	N=181	N=182		✓
Recruitment dates: March 1993 to September 2, 1994		B: 3, 4, 5 +b11 (DTwP-IPV separate)	Mean age at randomization (SD): NR	Mean age at randomization (SD): NR	Mean age at randomization (SD): NR		
Hib vaccine: PRP-T, brand name not stated, Pasteur Mérieux		C: 6, 7+b13	Mean age at vaccination (SD): NR	Mean age at vaccination (SD): NR	Mean age at vaccination (SD): NR		
Pertussis vaccine: wP, brand name not stated, Pasteur Mérieux		Additional information: All children had MMR at 14 months. A: DTwP-IPV at 3, 4, 5, 11 in a combined injection.	Gender (M/F): 94/86 (52% M)	Gender (M/F): 102/79 (56% M)	Gender (M/F): 104/78 (57% M)		
Funding: Chief Inspectorate of Health Care, Netherlands		B, C: DTwP-IPV at 3, 4, 5, 11 as a separate injection.					
Niger ³²							
Location: Niger	Inclusion criteria: children between the ages of four and twelve weeks, informed consent from the parents Exclusion criteria: none stated	A: 1.5, 2.5, 3.5	N= 59	N= 62	N= 59		✓
Recruitment dates: January to November 1995		B: 2.5, 3.5	Mean age at randomization: NR	Mean age at randomization: NR	Mean age at randomization: NR		
Hib vaccine: PRP-T, brand name not stated, Pasteur Mérieux		C: No doses	Overall mean age at vaccination (range): 1 st visit: 1.9(0.9-2.8) 2 nd visit: 3.0(2.1-5.1) 3 rd visit: 4.2(3.0-6.8)	Overall mean age at vaccination (range): 11 st visit: 1.9(0.9-2.8) 2 nd visit: 3.0(2.1-5.1) 3 rd visit: 4.2(3.0-6.8)	Overall mean age at vaccination (range): No Hib		
Pertussis vaccine: Not stated if wP or aP, assume wP given trial date. Brand name not stated, Pasteur Mérieux		Additional information: All children had BCG and OPV at birth, DTP (combined with Hib when Hib given) and OPV at 1.5, 2.5, 3.5; measles and yellow fever at 9 months.	Overall gender (M/F): 93/87 (52% M).	Overall gender (M/F): 93/87 (52% M).	Overall gender (M/F): 93/87 (52% M).		
Funding: Supported by the French Ministry of Cooperation and the WHO Global Program on Vaccines		C: Men A/C polysaccharide vaccine at 1.5, 3.5 months					

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunologica
Sweden ^{33, 34}							
Location: Sweden	Inclusion criteria: healthy term infants, with a birth weight of at least 2500 g, who were recruited with written informed consent of parents at the age of 2m +/-2 weeks at routine visits to Child Health Centers (CHC)	A: 2, 4, 6 +b13 B: 3, 5 +b12 Additional information: Both groups received DTaP-IPV in combination with Act-HIB in one injection.	N=118 Median age at randomization (SD): NR Mean age at vaccination (SD): NR but 98.8% of doses given within range stipulated in protocol Gender (M/F): NR	N=118 Median age at randomization (SD): NR Mean age at vaccination (SD): NR but 98.8% of doses given within range stipulated in protocol Gender (M/F): NR		✓	✓
Recruitment dates: November 19, 1994 to April, 1995							
Hib vaccine: PRP-T, ActHIB, Pasteur Mérieux Connaught	Exclusion criteria: none stated.						
Pertussis vaccine: aP (2 component), brand name not stated, Pasteur Mérieux Connaught							
Funding: Pasteur Mérieux Connaught, Göteborg Medical Society, the Medical Faculty of Göteborg University; the County Hospital of Norra Älvsborg							
Turkey ¹							
Location: Turkey	Inclusion criteria: healthy infants, Belgian, aged 2 months with informed written consent was obtained from the parents or legal guardian of each child	A: 3, 4, 5 +b12-14 (DTaP combined) B: 3, 4, 5 +b12-14 (DTaP separate) C: 2, 4, 6 (DTaP separate) Additional information: A: DTaP at 3, 4, 5, 12-14,combined B: DTaP at 3, 4, 5, 12-14, separate syringe. C: DTaP at 2, 4, 6 in a separate syringe.	N= 74* Mean age at randomization: 2 (0.5) Mean age at vaccination (SD): 1 st dose: 3.0 (0.2) 2 nd dose: 4.1 (0.3) 3 rd dose: 5.1 (0.3) Booster: 13.4 (1.1) Gender (M/F): 50/34 (60% M)	N= 78* Median age at randomization: 2 (0.5) Mean age at vaccination (SD): 1 st dose: 3.0 (0.1) 2 nd dose: 4.0 (0.2) 3 rd dose: 5.1 (0.4) Booster: 13.5 (1.1) Gender (M/F): 41/42 (49% M)	N= 81* Median age at randomization: 2 (0.5) Mean age at vaccination (SD): 1 st dose: 2.1 (0.2) 2 nd dose: 4.0 (0.3) 3 rd dose: 5.9 (0.3) No booster Gender (M/F): 51/32 (61% M)		✓
Recruitment dates: October 1994 to March 1995	Exclusion criteria: none reported						
Hib vaccine: PRP-T, Act-HIB, Pasteur Mérieux Connaught.							
Pertussis vaccine: aP, brand name not stated, Pasteur Mérieux, Connaught							
Funding: Pasteur Mérieux Connaught							

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunologica
USA1 ³⁵							
Location: USA Recruitment dates: August 8, 1991 to June 19, 1992 Hib vaccine: PRP-OMP, VaxHib, Merck & Co. PRP-HbOC, HibTiter, Praxis Biologics Pertussis vaccine: Not stated if wP or aP, assume wP given trial date. Brand name and manufacturer not stated Funding: National Institute of Allergy and Infectious Diseases	Inclusion criteria: healthy two month old infants with informed consent of parent or guardian and scheduled to receive routine immunization Exclusion criteria: none stated	A: 2 (PRP-OMP), 4, 6 (HbOC) B: 2 (HbOC), 4, 6 (PRP-OMP) C: 2, 4, 6 (HbOC) D: 2, 6 (PRP-OMP) E: 2, 4 (PRP-OMP) Additional information: DTP, OPV and MMR given to all groups “according to published guidelines”. All children received unconjugated PRP vaccine at 15m. D: Placebo at 4m E: Placebo at 6m	N=36[¶] Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall gender (M/F): 140/117 (55% M) Schedule D: N=36 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall gender (M/F): 140/117 (55% M)	N=35[¶] Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall gender (M/F): 140/117 (55% M) Schedule E: N=39 Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall gender (M/F): 140/117 (55% M)	N=96[¶] Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall gender (M/F): 140/117 (55% M)		✓
USA2 ³⁶							
Location: USA Recruitment dates: NR Hib vaccine: PRP-T, ActHib, Pasteur Merieux HbOC, HibTiter, Lederle-Praxis Biologics Pertussis vaccine: Not stated if wP or aP, assume wP given trial date. Brand name and manufacturer not stated Funding: National Institutes of Health and Connaught Laboratories	Inclusion criteria: healthy infants, 0 months of age with signed informed consent from a parent Exclusion criteria: infants of a gestational age of less than 37 weeks, receipt of any blood product, known or suspected impairment of neurologic function, acute febrile illness, severe congenital defect or major organ dysfunction, known maternal immunodeficiency or human immunodeficiency virus infection	A: 2, 4, 6 (PRP-T) B: 2, 4, 6 (HbOC) C: 0, 2, 4, 6 (HbOC) Additional information: All children received regularly scheduled childhood immunizations including HepB, DTP, and OPV concurrently as separate injections at 2, 4, 6. A and B: DT at birth	N=NR (total in all groups 150)* Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall, gender (M/F): 49% M	N=NR (total in all groups 150)* Mean age at randomization (SD): NR Mean age at vaccination (SD): 3 rd :6.7 Other doses NR Overall, gender (M/F): overall 49% M	N=NR (total in all groups 150)* Mean age at randomization (SD): NR Mean age at vaccination (SD): NR Overall, gender (M/F): overall 49% M		✓

Study details	Participant characteristics	Schedule A / schedule B / schedule C	Schedule A population characteristics	Schedule B population characteristics	Schedule C population characteristics	Outcomes	
						Mortality	Immunological
USA ^{37, 38}							
Location: USA	Inclusion criteria: healthy children from paediatric clinics in Missouri and Illinois with informed parental consent and with a physical examination performed prior to each immunization Exclusion criteria: history of a serious reaction to any previous vaccination, suspicion of underlying immunodeficiency. Vaccination deferred if history of fever within the previous 72 hours vaccination within the previous week	A: 2-6, 4-8	N= 27	N= 27		✓	
Recruitment dates: NR		B: 2-6, 3-7	Mean age at randomization (SD):	Median age at randomization (SD):			
Hib vaccine:		Additional information:	1st dose: 4.1 (1.6)	1st dose: 3.2 (1.3)			
PRP-OMP, PedvaxHIB, Merck Sharp & Dohme		No other vaccines described.	2nd dose: 6.1 (1.6)	2nd dose: 4.2 (1.3)			
Pertussis vaccine:			Overall mean age at vaccination (SD): 3.6(1.5)	Overall mean age at vaccination (SD): 5.1(1.8)			
Not described			Overall gender at randomization (M/F): 33/21 (61% M)	Overall gender at randomization (M/F): 33/21 (61% M)			
Funding:							
Supported, in part, by National Institute of Allergy and Infectious Diseases, National Institutes of Health, Connaught Laboratories, Inc. and Merck Sharp & Dohme							

Legend:

aP - acellular pertussis vaccine; BCG - Calmette-Guérin Bacillus; combined – Hib vaccine mixed in same syringe as other vaccines; DTP - diphtheria, tetanus, pertussis vaccine; DTaP - diphtheria, tetanus, acellular pertussis vaccine; DTwP - diphtheria, tetanus, whole cell pertussis vaccine; EPI: Expanded Program on Immunization; FHA - filamentous hemagglutinin; FIM - fimbriae; Hib – Haemophilus influenzae type b vaccine; m - months; MenACWY-PsACWY - quadrivalent meningococcal polysaccharide (groups A, C, Y, and W135) conjugate vaccine; MenA-TT-PsA-TT - MenA meningococcal conjugate vaccine; MMR - measles, mumps, rubella vaccine MMRV - measles, mumps, rubella, varicella vaccine; NR - Not reported; OPV - oral polio vaccine; p - primary course; PCV5: 5 valent pneumococcal conjugate vaccine; PCV7: 7-valent pneumococcal conjugate vaccine; PRP - polyribosylribitol phosphate; PRP-HbOC - PRP conjugated to diphtheria toxin CRM 197; PRP-OMP - PRP conjugated to outer membrane protein of Neisseria meningitidis; PRP-T - PRP conjugated to tetanus toxoid; wP - whole cell pertussis vaccine; separate – Hib vaccine not given in same syringe as other vaccines (other vaccines given at same or different time from Hib vaccine).

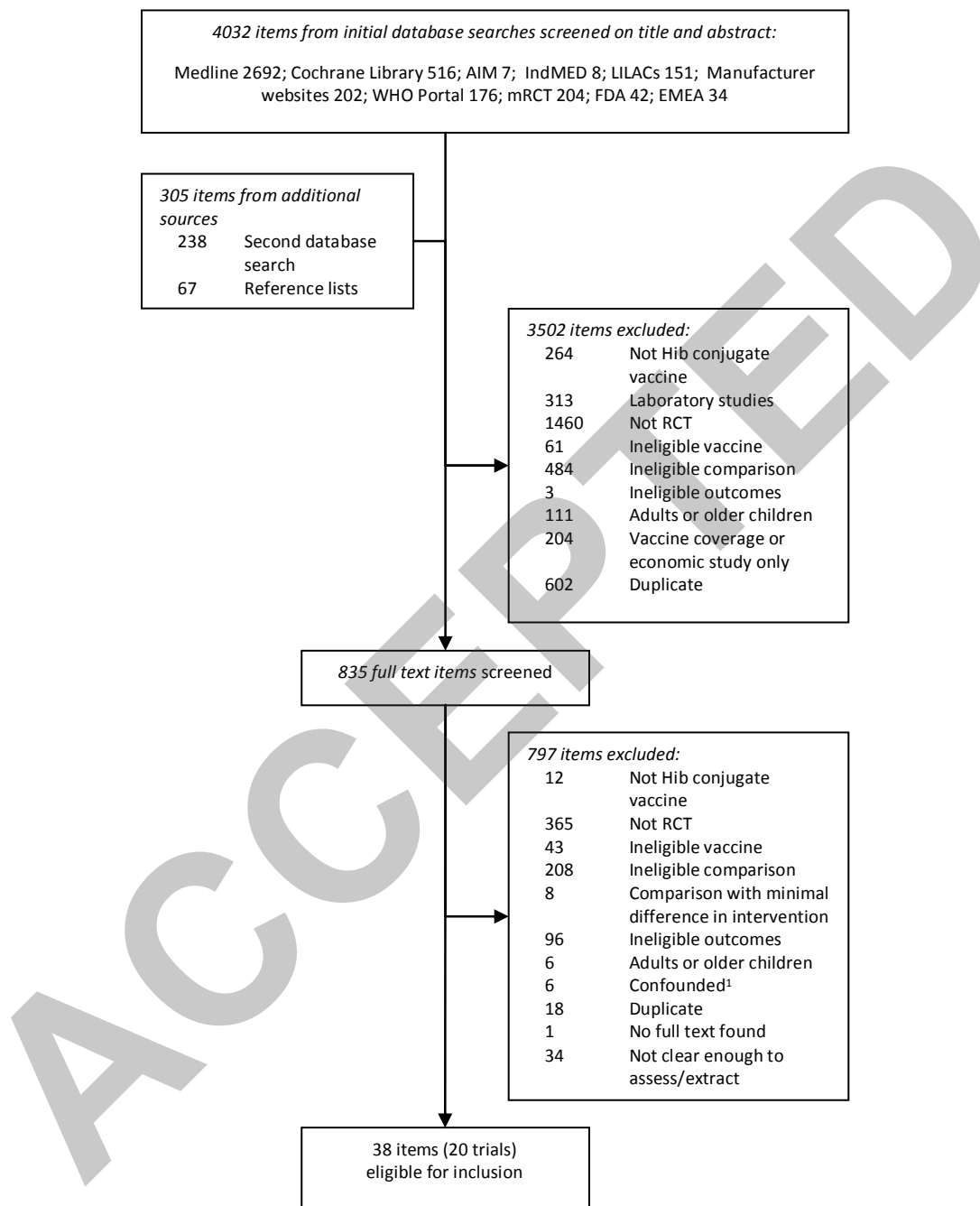
* Number of children vaccinated. Number of randomized children not reported.

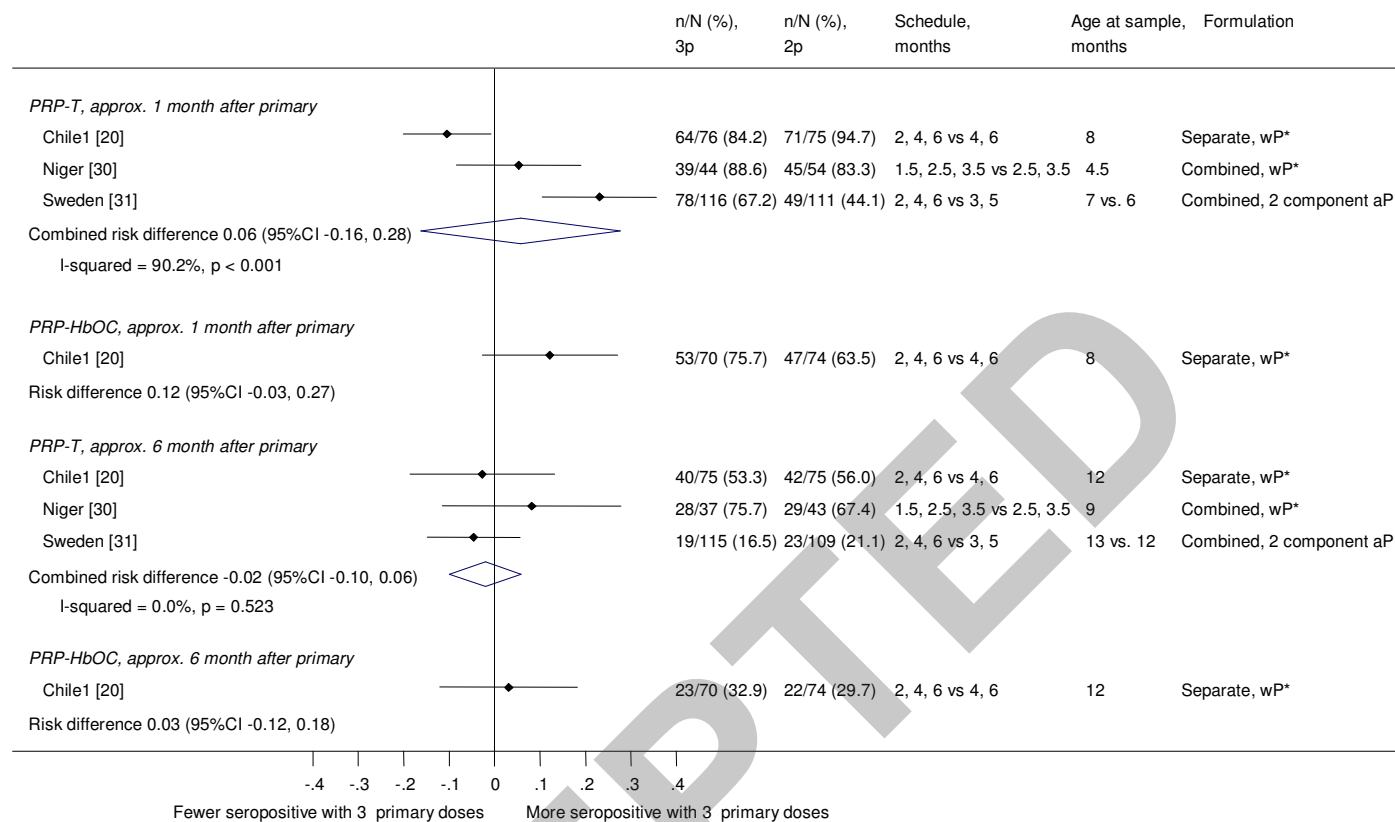
† Authors state the intended schedule immunization was met for each child with only 2 single exceptions

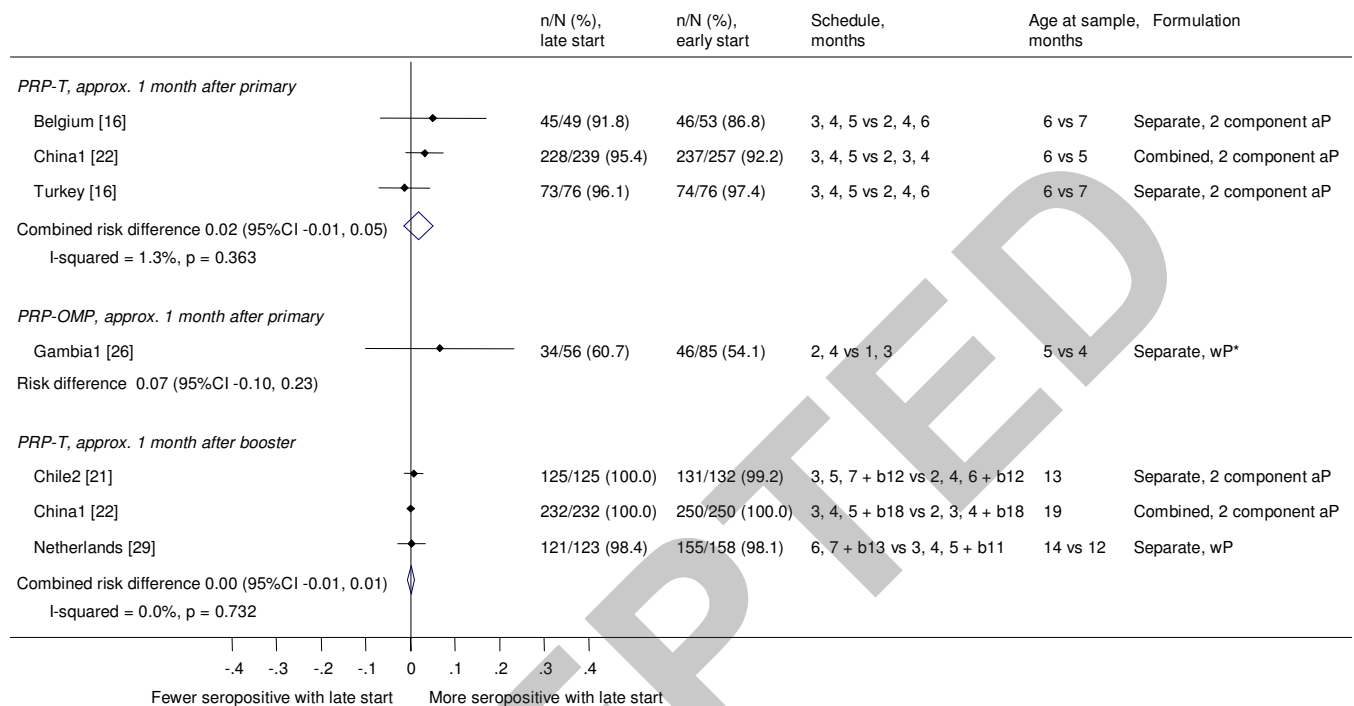
‡Type of conjugate vaccine in primary schedule (3p) not specified.

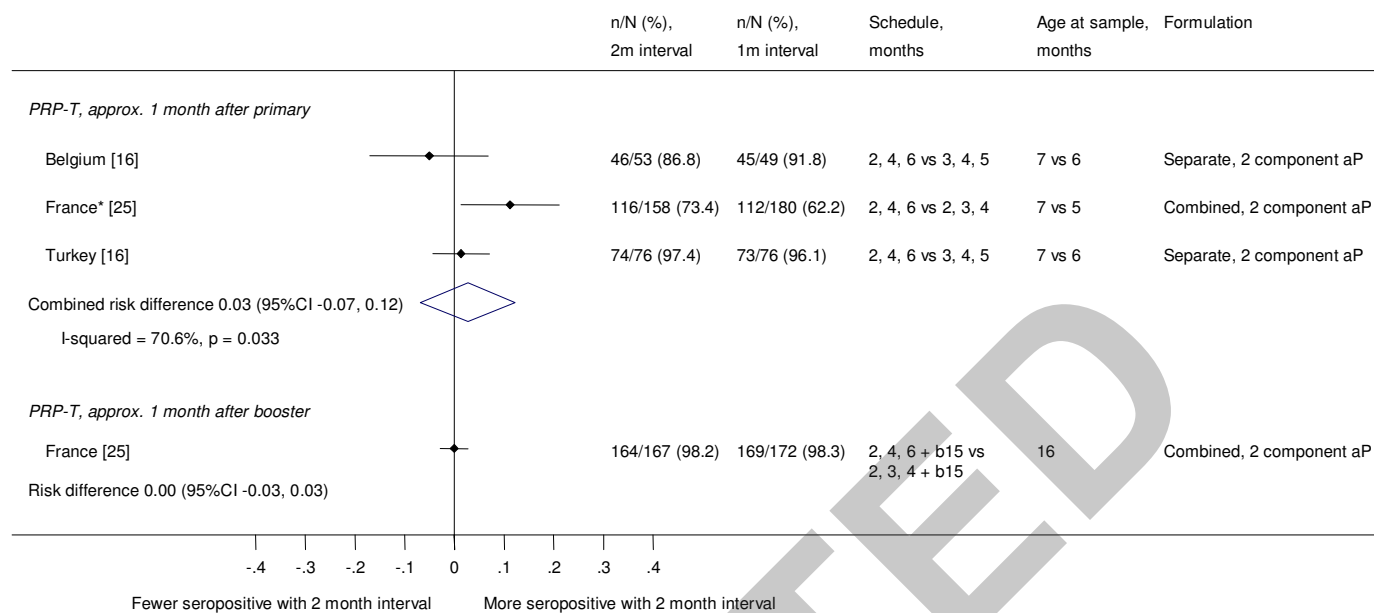
§ Group A includes 164 Ladino and 161 Native Indian participants; Group B includes 47 Ladino and 59 Native Indian participants.

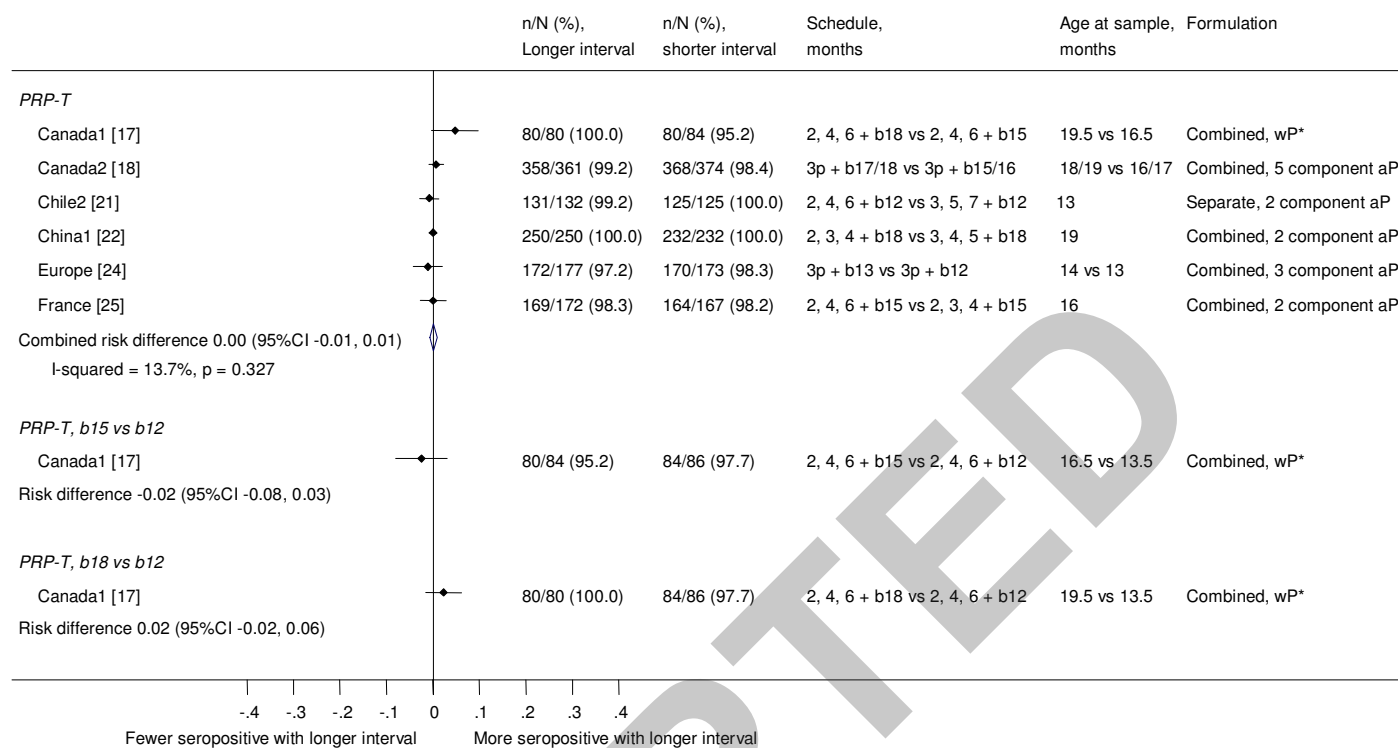
¶ Number of children followed-up. Numbers randomized to each group not reported. Total number randomized 497



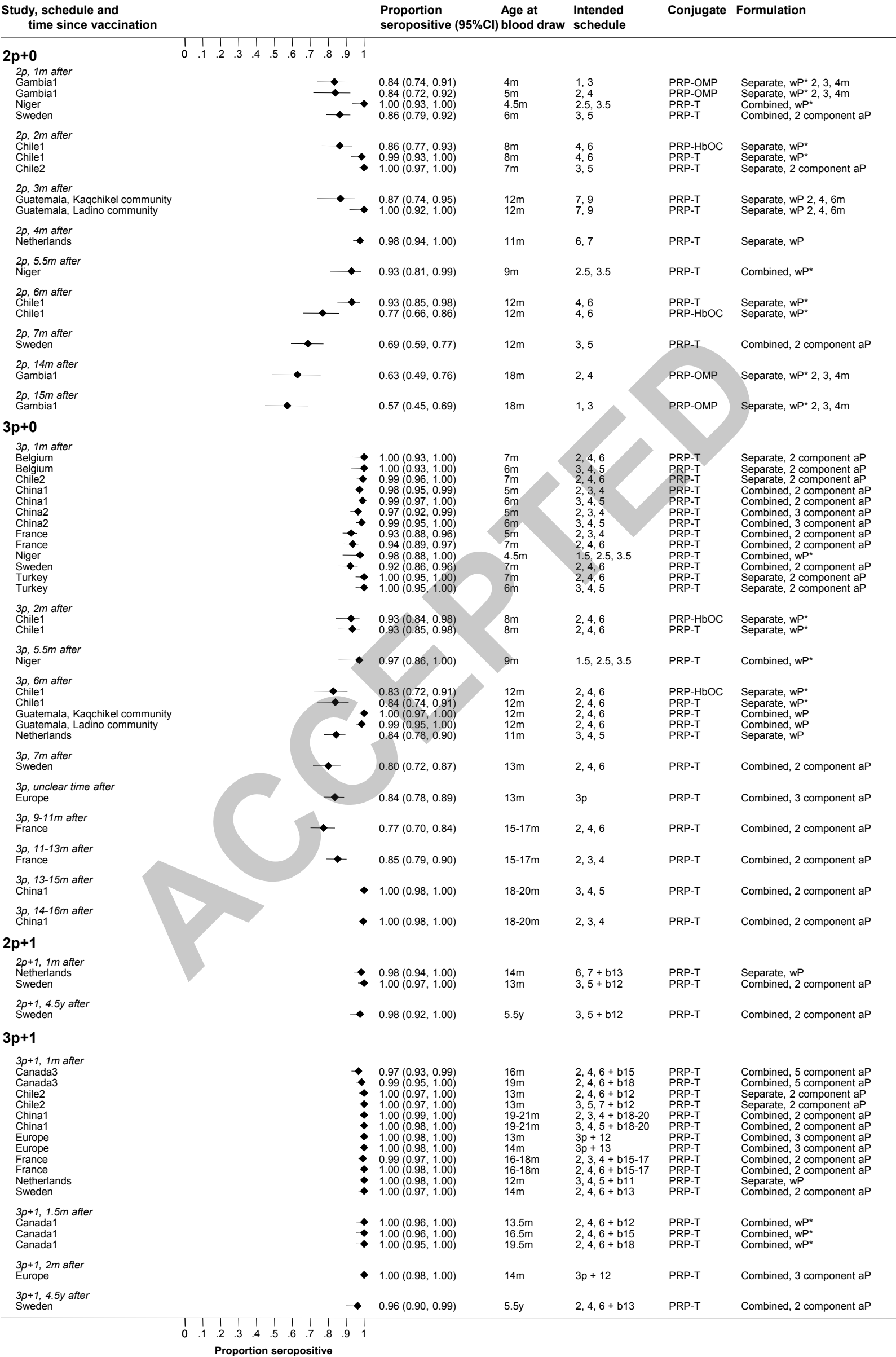






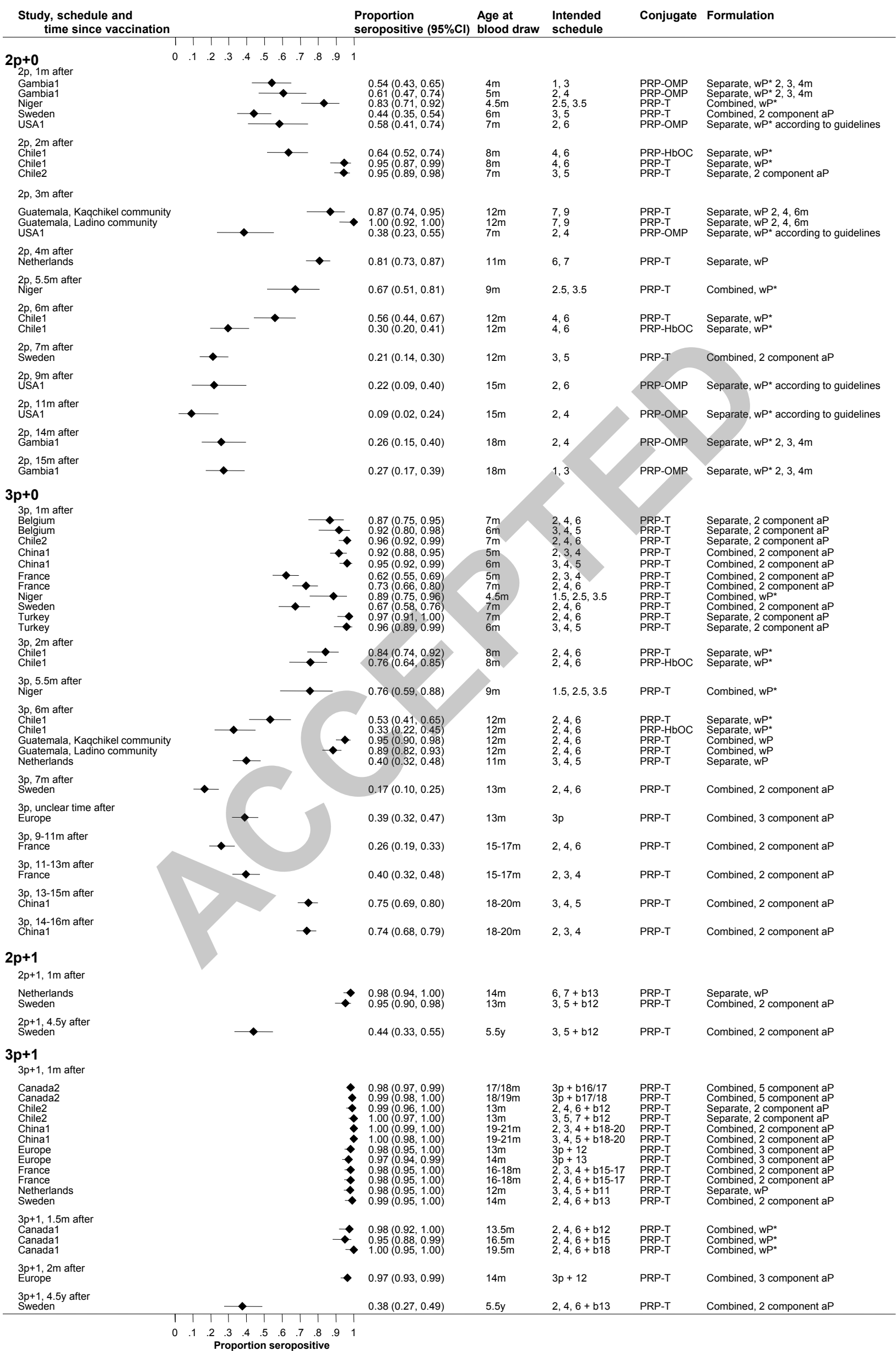


Supplementary figure 1: Seropositivity after 2p, 3p, 2p+1 and 3p+1 schedules, 0.15µg/ml



Combined - Hib vaccine administered in the same syringe as pertussis containing vaccine; separate - Hib vaccine administered by itself, either at the same time as or at different time from other vaccines; aP - acellular pertussis vaccine ; wP - whole-cell pertussis vaccine
* not specified as whole cell pertussis vaccine but assumed to be whole cell due to year trial conducted

Supplementary figure 2: Seropositivity after 2p, 3p, 2p+1 and 3p+1 schedules, 1.0µg/ml



Combined - Hib vaccine administered in the same syringe as pertussis containing vaccine; separate - Hib vaccine administered by itself, either at the same time as or at different time from other vaccines; aP - acellular pertussis vaccine ; wP - whole-cell pertussis vaccine

* not specified as whole cell pertussis vaccine but assumed to be whole cell due to year trial conducted