**Abstract**

Introduction of Hib vaccine is known to positively impact on reduction of both morbidity and mortality in children less than 5 years of age. Incorporation of this vaccine into a National EPI, however, does come at a significant cost, which is especially important in non-GAVI funded countries. Compounded reduction in response in certain patient populations and possible indication of booster doses further impacts on cost-benefit analyses. Despite these issues, South Africa has supplied Hib vaccine as part of the National EPI in the form of a combination vaccine, Pentaxim®, which combines Hib with Diphtheria, Tetanus, acellular Pertussis (DTP) and Poliomyelitis since 2009. Prior to this, another combination vaccine was utilized containing Hib and DTP. This has subsequently lead to a significant reduction in invasive Hib disease post-introduction, therefore largely justifying utilization.

**Introduction**

Historically, *Haemophilus influenzae* type b (Hib) was considered the most common severe invasive infection in children younger than 5 years of age[1, 2] in industrialized countries[3], causing in excess of 8 million serious infections worldwide[4]. The peak incidence among unvaccinated individuals varies from 6 to 7 months in developing countries[5], to slightly older in developed countries[6]. Hib-related mortality is attributed to meningitis and pneumonia, but invasive disease may also present as epiglottitis, osteomyelitis, septic arthritis, septicemia, cellulitis and pericarditis[6]. Worldwide studies conducted prior to the introduction of Hib vaccines amongst almost 4000 patients showed that in excess of 90% of patients presented with one of six clinical syndromes. Of these, meningitis accounted for more than half, but other clinical manifestations included bacteremic pneumonia, epiglottitis, septicemia, cellulitis and osteoarticular disease (with septic arthritis more common than osteomyelitis)[7]. Invasive disease represented only part of the clinical implication, as meningitis is often complicated with hearing impairment, seizure disorders, cognitive and developmental delay, and various other permanent neurological sequelae[8]. Introduction of Hib vaccination has had a major impact on invasive disease in both developing[9-12] and industrialized countries[7, 13, 14] despite the fact that disease epidemiology differs in these settings (table 1).
South Africa was the first African country to introduce Hib vaccine as part of the National Expanded Program on Immunization (EPI) in 1999[15]; the estimated coverage in 2004 was 92%[6]. Comparison of pre- and post-vaccination burden of diseases data is not possible as a national laboratory-based surveillance system for invasive Hib disease was established simultaneously with the introduction of Hib vaccine in 1999[15]. However, a study from Cape Town in the pre-immunization era performed at an academic hospital reported an incidence rate of invasive Hib disease of 169 and 47 per 100 000 population for children less than 1 and less than 5 years of age, respectively[17]. Based on the national laboratory-based surveillance (which yields only a fraction of the real burden) reported rates of invasive Hib disease in the first year following vaccination were 6.2 and 1.9 per 100 000 population in less than 1 year and less than 5 years old respectively. Over the period of 2000-2004 rates of invasive Hib disease decreased significantly, by 65% and 71% in less than 1 year old and less than 5 year old[16], indicating the impact of the Hib vaccine introduction in 1999.

Since 2003, the laboratory surveillance system became an active system including enhanced surveillance conducted at sentinel sites in each of the 9 provinces; detection rates of invasive Hib disease remains low, but from 2003 to 2009 the detection rate increased from 0.7 to 1.3 cases per 100 000 population in children less than 5 years old. Most of these cases were in fully vaccinated children (primary series of 3 doses at 6, 10 and 14 weeks of age[18]. These findings supported the decision to add since November 2010 a booster dose of Hib at 18 months of age as part of the a new pentavalent vaccine[18].

The World Health Organization (WHO) Strategic Advisory Group of Experts recommended worldwide implementation of Hib vaccination, in 2006. They further stated exception from this only if “robust epidemiologic evidence exists of low disease burden, lack of benefit, or overwhelming impediments to implementation”[19]. Despite convincing evidence collected over more than twenty years, indicating vaccine efficacy[20, 21], only 42% of children worldwide had received this vaccine by 2010[22]. Two main obstacles have been cited for this; firstly the lack of accurate epidemiological data due to various practical issues surrounding disease identification (discussed in text) and secondly, the high vaccine cost[23].

**Development of Hib conjugate vaccines**

Development of the first polysaccharide Hib vaccines started in the 1970’s with the only field studies performed in Finland[24]. This was achieved by utilizing

<table>
<thead>
<tr>
<th>Age of disease</th>
<th>African population</th>
<th>American population</th>
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<tbody>
<tr>
<td>6-7 months</td>
<td>46 (31–52)</td>
<td>1724 (1 574 – 2 817)</td>
</tr>
<tr>
<td>12 months</td>
<td>25 (16–30)</td>
<td>510 (466–834)</td>
</tr>
<tr>
<td>Clinical features (number of cases)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Meningitis</td>
<td></td>
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</tr>
<tr>
<td>Pneumonia</td>
<td></td>
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<tr>
<td>Death rate</td>
<td>60 (40–85)</td>
<td>11 (7–15)</td>
</tr>
</tbody>
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Table 1. Differential epidemiology of Hib disease in Africa versus the Americas described in children younger than 5 years of age and expressed per 100 000 population. (Adapted from reference 4)
the polyribosyrlribitol phosphate (PRP) subunits of the bacterial capsule[25]. This vaccine showed an 90% efficacy (95% confidence interval of 55-98%) specifically in children older than 18 months[24]. Efficacy in younger children is markedly lower due to the T-cell independent nature of the vaccine response. These formulations were only licensed for use in the United States (US)[26], Canada[27] and parts of Saudi Arabia[28], where more than 10 million doses were administered from 1985 to 1989 in the US alone[13]. By the late 1980’s, conjugate vaccines were being developed against Hib disease, and following this, combination formulations were developed containing these Hib conjugate vaccines[29]. These conjugate vaccines were proven to be superior to PRP vaccines as the PRP-only vaccines were poorly immunogenic in children under the age of 18 months[24], lacked a booster response[30] and did not show any reduction in nasal carriage[31]. This was by and large due to the T-cell-independent nature of the immune response to polysaccharides. Based on disease epidemiology where severe infection is typically noted in younger children, an alternative was needed to improve immunogenicity in this target group[6]. The first Hib conjugate vaccine introduced to the market was a diphtheria toxoid conjugate (PRP-D), thereafter altered to the mutant diphtheria toxin conjugate (PRP-HbOC)[7]. Later on, conjugates were developed containing the outer membrane protein of Neisseria meningitides (PRP-OMP) and tetanus toxoid (PRP-T)[32, 33] (table 2). The first vaccines to be commercially produced were formulated as PRP-HbOC, PRP-D or PRP-OMP and effectiveness was established by extensive clinical trials[34]. Subsequently, PRP-T formulations were produced and efficacy and licensing were based on demonstrating equivalent serum antibody levels compared to PRP-OMP and PRP-HbOC. Of note, most formulations currently utilized, conjugate to tetanus toxoid, as the conjugation technology is not protected by patent laws[6]. PRP-D formulations are no longer in clinical use as these vaccines have been shown to have inferior effectiveness, especially in high prevalence disease populations[35].

Table 2. Conjugate vaccines developed to improve immunogenicity of Hib vaccines.

<table>
<thead>
<tr>
<th>Subunit utilized</th>
<th>Licensing</th>
</tr>
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<tbody>
<tr>
<td><strong>Corynebacterium diphtheriae</strong></td>
<td>Modified non-toxic fragment of diphtheria toxin (PRP-HbOC)</td>
</tr>
<tr>
<td></td>
<td>Diphtheria toxoid (PRP-D)</td>
</tr>
<tr>
<td><strong>Neisseria meningitides</strong></td>
<td>Outer membrane protein (PRP-OMP)</td>
</tr>
<tr>
<td><strong>Clostridium tetani\textsuperscript{24}</strong></td>
<td>Tetanus toxoid (PRP-T)</td>
</tr>
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</table>

In December 2007, a voluntary recall of specific Hib conjugate vaccine lots (PRP-OMP Pedvax Hib\textsuperscript{®} and Combax\textsuperscript{®}) by Merck & Co., Inc. (West Point, USA) indirectly lead to generalized reduction in vaccine coverage. The recall was purely precautionary following identification of Bacillus cereus in vaccine manufacturing equipment[36], and subsequent surveillance did not reveal any contaminated vaccine lots[37] or clinical cases of vaccine-associated B. cereus
infection to recipients[36]. Subsequent recommendations were to simply omit use of the booster dose, but to continue vaccination otherwise. Despite this, a generalized reduction in vaccine coverage was noted. This finding highlights the importance of clearly communicated guidelines once a change in national policy is necessary[38].

**Cost, Distribution and Delivery**

Hib vaccine is more expensive than most of the other EPI vaccines. Costs were estimated to be as much as seven times that of measles, polio, Bacillus Calmette-Guérin (BCG), diphtheria, tetanus and pertussis vaccine in 2005[23] but current prices are 3 to 9 times the cost (S. Phoshoko, Personal communication). By the end of 2004, the WHO reported that only ten countries in Africa included Hib conjugate vaccine as part of their EPI. These countries are Burundi, The Gambia, Ghana, Kenya, Madagascar, Malawi, Rwanda, South Africa, Uganda and Zambia[39]. However, the current state of Hib vaccine use in Africa seems promising as only Equatorial Guinea, Nigeria, Tunisia, Botswana and Somalia are not including Hib in their routine EPI[40]. In January 2000, the Global Alliance for Vaccines and Immunization (GAVI) was launched, with the mission statement, to provide access to vaccines to the 70 poorest countries in the world[41]. Subsequently, this was expanded to the poorest 76 countries[23, 42]. The strategy aims to provide these vaccines through collaboration between the WHO, UNICEF, the World Bank, the Bill & Melinda Gates Foundation, donor governments, international development and finance organizations, the pharmaceutical industry, as well as from the developing countries themselves[41]. By the end of 2010, an additional 91 million children had received a full course of Hib vaccines, they would otherwise not have had access to[43]. One of the requirements for GAVI support is proof of burden of disease[44]. This has been an issue in the past in the Indian subcontinent, where inadequate surveillance data existed to motivate for provision of vaccines[45]. Fortunately, this has fueled research in this field, confirming mortality due to Hib meningitis to be as high as 11% with 30% of survivors suffering subsequent major neurological sequelae[45].

Despite formal inclusion in the respective EPI’s, vaccine coverage varies significantly from as low as 50% (Madagascar)[39] and more recently, Central African Republic (58%), to 99% in Burkina Faso[46]. Evaluation of rates of invasive Hib disease is dependent on National Surveillance systems. Several countries do not make use of this, and those who do, report significantly variable rates. These vary from no reported cases (Congo, Gambia, Guinea-Bissau, Lesotho, Rwanda, Suriname and Zambia) in excess of 6 000 Hib meningitis cases (Burkina-Faso)[46].

**Mechanism of Vaccine Protection**

Initial efforts to develop a vaccine utilized polysaccharide antigens. This elicits a T-cell independent response, classically characterized by lower antibody titers, low affinity antibodies with the absence of immune memory. Subsequent efforts to conjugate vaccines to proteins, significantly improved the protective response[47]. Clinically, the improved immunogenicity is of particular importance in children under the age of 18 months[24].
It has been established through PRP vaccine studies\cite{48}, that antibody levels in excess of 0.15 μg/mL and 1μg/mL in serum is indicative of short- and long-term protection, respectively\cite{49}. This is due to both the opsonic activity of antibodies\cite{50-52}, as well as complement-mediated bactericidal activity\cite{51, 53}. In addition, the vaccine offered indirect protection by delaying nasopharyngeal carriage and asymptomatic colonization amongst vaccinated infants\cite{54-57}. This provides an additional level of herd immunity\cite{58, 59}. For this reason, Hib vaccine is considered to have effectiveness greater than reported efficacy\cite{6}. Various studies have described community settings where unvaccinated children show significant disease reduction, as a byproduct of vaccination of their peers\cite{9, 60-63}. Intermittent colonization, however, is associated with development of natural immunity\cite{64, 65} and is likely the cause of the natural decline in incidence of invasive disease with age amongst unvaccinated children\cite{66}.

HIV-1 positive patients generally do not launch the same degree of immune response to vaccines as their HIV-negative counterparts in terms of antibody titer production\cite{49}. In addition, presence of anti-PRP antibodies may not confer protection within this population group, as in some instances, antibodies have been shown to be functionally impaired\cite{67}. This quantitative and qualitative impairment seems to correlate with degree of HIV-1 disease progression\cite{67,68}.

**Vaccine efficacy and effectiveness**

Epidemiologically, Hib vaccine effectiveness is considered as a measure of reduction in invasive disease. Invasive disease is confirmed by culture of *Haemophilus influenzae* type b from blood or cerebrospinal fluid (CSF). However, invasive disease is generally considered as a poor indicator of overall Hib disease. In addition, there a relatively small fraction of children with other invasive disease forms like Hib pneumonia will be bacteremic. Furthermore, false laboratory negative results may occur due to inherent difficulties in bacterial culture. Hib is a fastidious organism and is highly sensitive to variables pertaining to sampling after initiation of antibiotic therapy, transport delay and incubation conditions\cite{7}. For these reasons using this case definition, the number of clinical cases, may be significantly underestimated\cite{6}. Various authors have suggested alternative diagnostic and epidemiological means, to overcome this problem, including antigen detection methods\cite{69,70} as well as vaccine-probe study designs\cite{71}. Laboratory parameters focusing on both quantitative and qualitative antibody characteristics have been suggested as markers of protection against disease\cite{72-76}. This is in light of the fact that immunoglobulin quantity\cite{77}, subtype\cite{73,76} and avidity\cite{72-75} all contribute to the protective effect.

The WHO advocates that all countries should measure the impact of Hib vaccination in their setting, if practically permitted\cite{78}. The aim with this is not only to determine the vaccine performance under field conditions in general, but specifically within certain population groups. Of particular interest is HIV-1 positive individuals, as vaccine efficacy may be reduced\cite{79}. Proof of effectiveness within a specific population aids in justifying use of these vaccines.
as part of national guidelines, seeing as this vaccine is in general more costly than most other vaccines included in the EPI. To determine effectiveness of Hib vaccine, a high-quality population-based surveillance system is required to be in place, to collect data from both pre-vaccine and post-vaccine periods[78]. The South African National Surveillance system was established the same year as introduction of Hib vaccine into the EPI in 1999[18]. Therefore, pre-vaccination data is restricted to studies not performed on national level[17]. Of note, all countries that have included Hib vaccines as part of the national vaccination schedule have shown a significant reduction in invasive disease[14, 21, 80]. As part of an extensive meta-analysis performed by O’Loughlin et al in 2010, vaccine efficacy against invasive disease was found to be 95% (95% CI 82-99)[78]. This corresponds well with a Cochrane review on this matter[81], as well as various geographically diverse studies[9, 82-87].

Hib vaccination and HIV-1 populations
At present, there are an estimated 3.4 million HIV-infected children under the age of 15 years, with 390 000 children newly infected per annum[88]. Of these, less than 10% receive the ART they require[89]. Amongst these patients, one in three children will demise before one year of age, from various causes, including serious bacterial infection[90]. HIV-infected children are at a significantly higher risk of developing bacteremic pneumonia than their HIV-negative counterparts[91]. In the absence of ART, use of co-trimoxazole prophylaxis has been shown to reduce mortality by as much as 43%[92]. It is postulated that is due to prevention of invasive bacterial infection[93]. This practice, however, requires daily administration of antibiotics for prolonged periods of time, and therefore preventative vaccines may have a major impact in reducing mortality in this respect[93]. The WHO recommends use of all routine vaccines in HIV-infected patients with some notable alterations[94]; firstly evaluation of risk for disseminated BCG[95], and secondly earlier administration of measles vaccine owing to reduced maternal antibody levels for placental transfer[96]. Additionally, due to the inherent immunosuppression noted in HIV-infected individuals, effectiveness of vaccines, in general, has been described to be reduced[79], often necessitating booster doses[93].

Very limited data is available on Hib vaccine effectiveness within settings with a high HIV-1 seroprevalence. Attempts to study vaccine performance have been published from Malawi[84] and Kenya[97], showing either very low patient numbers or low HIV-1 seroprevalence, respectively. Most of our currently understanding of Hib vaccine in this population, comes from a study performed in South Africa. In this study, it is evident that vaccine effectiveness is reduced amongst HIV-infected patients, although still showing moderate activity (55% disease reduction versus 91% amongst HIV-1 negative patients). The HIV-1 rate used in this study was obtained from antenatal clinic attendee data, and therefore, vaccine efficacy may have been underestimated[93]. Furthermore, the risk of vaccine failure was estimated to be 35 fold higher (95% CI 14.6 – 84.6) for HIV-infected patients[67]. The reason for this seems to be due to a reduction in both quantity and quality of Hib antibodies[67]. In the South African study, HIV-infected children not only had a statistically significantly lower geometric mean antibody concentration 1-month post-immunization, but also lower rates of
HibPS antibody concentrations to enable a 50% serum bactericidal activity[67]. Notably, booster doses of Hib vaccine have shown a subsequent improvement of antibody titers to protective levels[98-99], and some authors advocate this practice, although optimal timing of vaccination has not been established. However, the use of antibody level as a marker of protection within this patient population has been questioned by some authors[93] and more extensive studies are therefore warranted[93].

**Hib Vaccines and South Africa**

South Africa is not included as one of the GAVI funded nations[43]. Despite this, it was the first African country to self-finance inclusion of Hib conjugate vaccines as part of the EPI since 1999[16]. Furthermore, the most recent EPI schedule, as implemented in 2009, also includes a booster dose at 18 months[100] as part of the pentavalent vaccine (Pentaxim®)[101].

From introduction to the South African EPI in 1999, Hib vaccine was administered as part of a combination formulation with Diphtheria, Tetanus and whole cell Pertussis (DTP). Although monovalent formulations of the Hib vaccine are available, various combination vaccines offer the advantage of fewer injections without compromising on the number of antigens administered[102]. The Centers for Disease Control and Prevention recommends two (PRP-OMP) to three (PRP-T) primary doses, in monovalent or combination form, with a booster dose at 12 to 15 months. Licensed vaccines may be used interchangeably, in order to complete the vaccination series. Of note, the Hibercin® formulation (GlaxoSmithKline, Bryanston, South Africa) is only registered for use as a booster, and not for primary vaccination[102]. The EPI currently in effect, as stipulated in April 2009 utilizes the Sanofi Pasteur vaccine, Pentaxim®, which is a combination vaccine for DTP (acellular formulation), Hib and Poliomyelitis. Since first being marketed in 1997, more than 100 million doses have been administered in over 100 countries, of which 23 have included it as part of the local EPI[103]. GAVI-supported countries use the pentavalent formulation, which contains Diphtheria, Tetanus, whole-cell Pertussis, *Haemophilus influenzae* type b and Hepatitis B virus. This formulation is less expensive but does have a worse side-effect profile owing to use of whole-cell as opposed toacellular Pertussis[42].

There has been a significant reduction in cases of invasive Hib disease since the introduction of the Hib vaccine. This trend is most obvious within the below one year of age population group. Risk factors for developing invasive Hib disease were identified as HIV-1 infection and incomplete vaccination. Despite the clear correlation between HIV-infection and risk of vaccine failure[67], vaccine failures have not been described in other high HIV-1 seroprevalence settings[104]. The changes made within the national vaccination recommendations, are also supported by an improved surveillance system. This laboratory-based surveillance system identified that the incidence of non-typable *Haemophilus influenzae* was higher amongst HIV-1 infected patients, and laboratory tested antimicrobial resistance was becoming increasingly important[16]. It did not allude to the bimodal vaccine failure pattern described
previously in South Africa amongst HIV-infected patients[91,105]. The introduction of the booster dose was prompted by resurgence of invasive Hib disease, which was ascribed to vaccine failure[18]. It will, however be interesting to observe the impact of the booster dose, as introduced in 2009, on the epidemiology of invasive Hib disease amongst HIV-1 infected and uninfected children.

Conclusion
The global acceptance of Hib vaccines has led to major reduction in global disease burden, directly impacting on morbidity and mortality[7]. As the relatively high cost of incorporating this vaccine into any national immunization schedule invariably becomes an important determinant for use, external funding to poorer countries is essential to ensure adequate vaccine coverage to effect the same disease reduction to all.

The high sero-prevalence of HIV-1 in South Africa compounds issues surrounding Hib disease prevention, as HIV-infected children seem to show a more rapid waning immunity as compared to their HIV negative counterparts. This, combined with a higher risk of invasive Hib disease in HIV-infected individuals, is cause for concern. The impact of the booster dose of Hib vaccine administered at 18 months remains to be seen as the hope is that this will provide benefit not only to partially-vaccinated patients, but also unvaccinated individuals by providing an additional vaccination opportunity in the second year of life[18].

References


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