The Immunological Basis for Immunization Series

Module 15: Meningococcal Disease
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<th>Full Form</th>
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<tbody>
<tr>
<td>ALL</td>
<td>acute lymphoblastic leukaemia</td>
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<tr>
<td>AML</td>
<td>acute myeloid leukaemia</td>
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<tr>
<td>aP</td>
<td>acellular pertussis</td>
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<tr>
<td>CI</td>
<td>confidence interval</td>
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<tr>
<td>CRM&lt;sub&gt;197&lt;/sub&gt;</td>
<td>non toxigenic natural variant of diphtheria toxin</td>
</tr>
<tr>
<td>DT</td>
<td>diphtheria-tetanus</td>
</tr>
<tr>
<td>DTaP</td>
<td>diphtheria-tetanus-acellular pertussis</td>
</tr>
<tr>
<td>DTwP</td>
<td>diphtheria-tetanus-whole cell pertussis</td>
</tr>
<tr>
<td>ELISA</td>
<td>enzyme-linked immunosorbent assay</td>
</tr>
<tr>
<td>EPI</td>
<td>Expanded Programme on Immunization</td>
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<tr>
<td>EU-IBIS</td>
<td>European Union Invasive Bacterial Infections Surveillance Network</td>
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<tr>
<td>FDA</td>
<td>Food and Drug Administration (USA)</td>
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<tr>
<td>GBS</td>
<td>Guillain-Barré syndrome</td>
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<td>GMT</td>
<td>geometric mean titre</td>
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<tr>
<td>GSK</td>
<td>GlaxoSmithKline Biologicals</td>
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<tr>
<td>Hib</td>
<td><em>Haemophilus influenzae</em> type b</td>
</tr>
<tr>
<td>HBV</td>
<td>hepatitis B virus</td>
</tr>
<tr>
<td>hSBA</td>
<td>serum bactericidal antibody measured with exogenous human complement</td>
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<td>HSCT</td>
<td>hematopoietic stem cell transplant</td>
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<tr>
<td>Ig</td>
<td>immunoglobulin</td>
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<tr>
<td>IPV</td>
<td>inactivated polio vaccine</td>
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<tr>
<td>LOS</td>
<td>lipooligosaccharide</td>
</tr>
<tr>
<td>MCC</td>
<td>meningococcal group C conjugate</td>
</tr>
<tr>
<td>ME</td>
<td>myalgic encephalomyelitis</td>
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<tr>
<td>MHRA</td>
<td>The Medicines and Healthcare Products Regulatory Agency</td>
</tr>
<tr>
<td>MLST</td>
<td>multilocus sequence type</td>
</tr>
<tr>
<td>MMR</td>
<td>measles, mumps and rubella vaccine</td>
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<tr>
<td>MVP</td>
<td>Meningitis Vaccine Project</td>
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<tr>
<td>Acronym</td>
<td>Description</td>
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<tr>
<td>NACI</td>
<td>National Advisory Committee on Immunization</td>
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<tr>
<td>OPA</td>
<td>opsonophagocytosis</td>
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<tr>
<td>OMP</td>
<td>outer membrane protein</td>
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<tr>
<td>OMV</td>
<td>outer membrane vesicle</td>
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<tr>
<td>PATH</td>
<td>Program for Appropriate Technology in Health</td>
</tr>
<tr>
<td>PCR</td>
<td>polymerase chain reaction</td>
</tr>
<tr>
<td>rSBA</td>
<td>serum bactericidal antibody measured with exogenous rabbit complement</td>
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<tr>
<td>SAE</td>
<td>serious adverse event</td>
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<tr>
<td>SBA</td>
<td>serum bactericidal antibody</td>
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<tr>
<td>ST</td>
<td>sequence type</td>
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<tr>
<td>TT</td>
<td>tetanus-toxoid</td>
</tr>
<tr>
<td>Td</td>
<td>diphtheria-toxoid</td>
</tr>
<tr>
<td>VAERS</td>
<td>Vaccine Adverse Event Reporting System</td>
</tr>
<tr>
<td>WHO</td>
<td>World Health Organization</td>
</tr>
<tr>
<td>wP</td>
<td>whole-cell pertussis</td>
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Preface

This module is part of the series The Immunological Basis for Immunization, which was initially developed in 1993 as a set of eight modules focusing on the vaccines included in the Expanded Programme on Immunization (EPI). In addition to a general immunology module, each of the seven other modules covered one of the vaccines recommended as part of the EPI programme — diphtheria, measles, pertussis, polio, tetanus, tuberculosis and yellow fever. The modules have become some of the most widely used documents in the field of immunization.

With the development of the Global Immunization Vision and Strategy (GIVS) (2005–2015) and the expansion of immunization programmes in general, as well as the large accumulation of new knowledge since 1993, the decision was taken to update and extend this series.

The main purpose of the modules — which are published as separate disease/vaccine-specific modules — is to give immunization managers and vaccination professionals a brief and easily-understood overview of the scientific basis of vaccination, and also of the immunological basis for the World Health Organization (WHO) recommendations on vaccine use that, since 1998, have been published in the Vaccine Position Papers.

WHO would like to thank all the people who were involved in the development of the initial Immunological Basis for Immunization series, as well as those involved in its updating, and the development of new modules.

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1 This programme was established in 1974 with the main aim of providing immunization for children in developing countries.
1. Meningococcal disease

1.1 Introduction
Meningococcal disease is caused by the Gram-negative bacterium *Neisseria meningitidis*, also known as the meningococcus. Meningococcal disease remains a significant public-health issue globally, with infections occurring both endemically and epidemically in developed and developing countries (Rosenstein et al., 2001). Humans are the only reservoir for the meningococcus. Normally the bacteria colonize the nasopharynx without any deleterious effects on its host, but can also occasionally cause life-threatening disease. Why a particular individual colonized by the meningococcus develops systemic infection while others, who are equally colonized, develop immunity, is due to complex interactions involving both the host and the bacterium (Stephens, 1999).

1.2 *Neisseria meningitidis*
Meningococci are aerobic, Gram-negative, oxidase positive, encapsulated diplococci (Vedros & Genus, 1984). There are 13 capsular groups that have been identified based on the immunochemistry of the capsular polysaccharide (Branham, 1953; Vedros, 1987). Capsular groups A, B, C, W135 and Y are responsible for the majority of meningococcal disease observed. Further classification of *N. meningitidis* is based on the expression of outer membrane proteins (PorA and PorB) (Tsai et al., 1981; Frasch et al., 1985; Abdillahi & Poolman, 1988a; Abdillahi & Poolman, 1988b; Abdillahi & Poolman, 1988c), lipooligosaccharide (LOS) structure (Scholten et al., 1994) and sequence polymorphisms in housekeeping genes which provides a multilocus sequence type (MLST), often abbreviated to sequence type (ST) (Maiden et al., 1998).

1.3 Epidemiology
Epidemic meningitis is defined by WHO for sub-Saharan Africa (weekly attack rates allow detection of epidemic and alert thresholds) (WHO, 2000a). The area designated the “Meningitis Belt”, in sub-Saharan Africa, stretching from the Atlantic in the west to the African Horn in the east, still experiences epidemics of group A disease regularly, with thousands of cases and deaths occurring during these widespread epidemics. A characteristic feature is that they always start around the middle of the dry season, rapidly building up to a peak and then subsiding abruptly with the coming of the rains (Greenwood, 2006). In 2007, an epidemic occurred in Burkina Faso with approximately 25 000 cases of capsular group A and 1700 deaths reported to the Ministry of Health.
by May (WHO, 2007d). Although most epidemics in sub-Saharan Africa have been caused by group A meningococci, group C and also group W135 and X outbreaks have been described (Greenwood, 2007). Epidemics of group A disease have also been reported in Asia (Hu et al., 1991; WHO, 1995; Hu, 2001; Sachdeva et al., 2005). Other regions have experienced epidemics of group B disease, including Norway (Bjune et al., 1991) where disease was due to ST32 clonal complex, phenotype B:15P1.7, 16 and Cuba (Sierra et al., 1991) where the clonal complex was also ST32 but phenotype B:4:P1.19,15. This Cuban strain spread to the São Paulo region of Brazil in the late 1980s (Cruz et al., 1990; de Moraes et al., 1992). Since 1991, New Zealand has experienced an epidemic caused by a strain identified as B:4P1.7–2,4, ST-41/44 clonal complex (Martin et al., 1998). Endemic disease (groups B and C) due to heterogenous meningococcal clonal complexes is of major concern in North and South America, Australia and Europe.

Capsular group W135 was a relatively uncommon cause of invasive disease worldwide until outbreaks of group W135 disease during the Hajj pilgrimage were reported in Saudi Arabia (Popovic et al., 2000; Taha et al., 2000; Finn et al., 2001; Taha et al., 2004). The outbreak was caused by a group W135 clone that belonged to the ST-11 complex, and this clone was shown to spread from pilgrims to contacts in their home countries (Wilder-Smith et al., 2003). Group W135 isolates belonging to the ST-11 complex were also isolated from patients with meningococcal disease in Mali in 1994 (Kwara et al., 1998), in the Gambia (Kwara et al., 1998), Cameroon in 1995 and Chad in 1996 (Guibourdenche et al., 1996), and in Rio Grande do Sul, Southern Brazil in 2003 (Weidlich et al., 2008). In Burkina Faso in 2002, more than 10 000 cases were caused by group W135 (Decosas & Koama, 2002).

Meningococcal infections due to capsular group X are relatively uncommon but have been reported from Niger in 2006 (Boisier et al., 2007). From January to June 2006, a total of 4185 cases of meningitis were reported, with 2905 cerebrospinal fluid samples tested. Group X represented 51% of 1139 confirmed cases of meningococcal disease, but in southwestern Niger, it represented 90%. In the agglomeration of Niamey, the reported cumulative incidence of confirmed group X meningitis was 27.5 cases per 100 000 population (74.6 cases per 100 000 population in children aged 5–9 years). The group X isolates had the same phenotype (X:NT:P1.5) and sequence type (ST-181) as the group X isolates that were circulating in small outbreaks in Niamey in the 1990s (Campagne et al., 1999; Djibo et al., 2003). Small outbreaks of group X disease were also reported from northern Ghana (Gagneux et al., 2002) and from Uganda (Caugant, 2006, personal communication).

Meningococcal infections with group Y are more likely to be pneumonia than disease caused by other capsular groups (Koppes et al., 1977). However, in the United States of America, the proportion of disease caused by group Y strains rose from 2% of disease in 1989–1991 to 32.6% in 1996 (Rosenstein et al., 1999). Although carriage of group Y is common in many other parts of the world, disease remains relatively rare.
1.4 Risk factors

The meningococcus only colonizes the nasopharynx of humans and has no other known environmental niche. It is transmitted from person-to-person by aerosol droplets or by contact with respiratory secretions. Respiratory infections, such as influenza A (Cartwright et al., 1991), influenza B (Harrison et al., 1991) and Mycoplasma (Moore et al., 1990), exposure to tobacco smoke (Fischer et al., 1997; MacLennan et al., 2006) or indoor firewood stoves (Hodgson et al., 2001), bar and disco patronage (Cookson et al., 1998; Honish et al., 2008), binge drinking (Finn et al., 2001) and intimate kissing (MacLennan et al., 2006) have all been associated with increased rates of meningococcal carriage or disease.

1.5 Clinical presentation

The classical presentation of meningococcal disease is fever, rash and meningitis with symptoms including abrupt onset of fever, headache, photophobia, myalgias, malaise and altered consciousness, but this can be indistinguishable from other bacterial or viral illnesses (Steven & Wood, 1995). In infants, there may be a more gradual onset of fever, with poor appetite and lethargy often symptoms, plus bulging fontanel indicating involvement of the central nervous system. Dissemination into the bloodstream results in rapid progression of severe septicemia with petechial or purpuric rash and hypotension, and can lead to multiple organ failure. Meningococcal disease can also present as pneumonia which, in the absence of a rash, may go unrecognized. Permanent sequelae can result from meningococcal disease and may include loss of limbs (peripheral gangrene), hearing loss and neurological complications.

1.6 Confirmation of meningococcal disease

Confirmation of meningococcal disease is traditionally made by culture of meningococci from a sterile site such as blood or cerebrospinal fluid. Gram staining for demonstration of the characteristic Gram-negative diplococcus remains an important confirmatory tool. Serodiagnosis can be performed, either by latex agglutination, which is a rapid test, or by polymerase chain reaction (PCR) measurement of antibody levels which is retrospective by nature. For rapid capsular grouping, particularly within developing countries, a gold particle immunochromatography methodology for detection of soluble polysaccharide has been developed (Chanteau et al., 2006). As few reference laboratories exist in sub-Saharan Africa, a simple, rapid test will be of great use in informing on vaccination strategies. PCR assays have been developed, and initial screening utilizes PCR assays based on conserved regions of either the ctrA gene (a capsular transport gene) or the crgA gene (Taha et al., 2005). Confirmation of capsular group-specific sequences within the sialyltransferase (siaD) gene is then used to discriminate between capsular groups B, C, Y or W135 meningococci (Guiver & Borrow, 2001; Mothershed et al., 2004; Taha et al., 2005). For confirmation of group A disease, PCR amplification targets orf2 sequences within the gene cassette required for the synthesis of the capsule for group A (Orvelid et al., 1999) which can provide an initial screen to confirm if the sample is positive for meningococci, and can confirm the capsular group (Orvelid et al., 1999; Mothershed et al., 2004; Taha et al., 2005). The bacterial load, as measured by real time-PCR, has been shown to be unaffected by the administration of antibiotics prior to the patient’s hospital admission (Darton et al., 2008).
1.7 Treatment

There are several antimicrobial agents available for the treatment of meningococcal disease, although penicillin remains the therapy of choice. In resource-poor countries with limited health facilities, such as those in the meningitis belt, the treatment of meningococcal meningitis during epidemics is based on short-course, long-acting oily chloramphenicol. Its administration, by the intramuscular route, is straightforward, and a single dose has an efficacy greater than 90%. However, it is contraindicated in pregnant women and in children less than one year old. The continued production of this drug, which is used exclusively for treatment of epidemic meningitis in Africa, is uncertain. Following a study conducted in peripheral health structures in an epidemic context in Niger in 2003, an alternative treatment was suggested that demonstrated that a single-dose of ceftriaxone is equally effective, and can be used in pregnant women and infants (Nathan et al., 2005). Use of a single-dose of ceftriaxone was subsequently recommended by the Essential Drug Expert Committee for the treatment of epidemic meningococcal meningitis; (http://www.who.int/csr/resources/publications/meningitis/WHO_CDS_EPR_2007_3/en/index.html).

Early initiation of antibiotic therapy and presentation to hospital have been associated with reduced mortality and morbidity (Jolly & Stewart, 2001; Public Health Laboratory Service et al., 2002). One study explored the impact on mortality and morbidity of parenteral penicillin given to children before admission to hospital with suspected meningococcal disease. This study demonstrated that children who were given parenteral penicillin by a general practitioner had more severe disease on reaching hospital than those who were not given penicillin before admission. The association with poor outcome may be because children who are more severely ill are being given penicillin before admission (Harnden et al., 2006). For the treatment of non-confirmed cases of meningococcal disease, antibiotics with a broader spectrum of action should be considered, such as cefotaxine or ceftriaxone (Rosenstein et al., 2001). Antibiotic resistance in meningococci remains at a low level, although group A strains resistant to penicillin and non-susceptible to cefotaxine and ceftriaxone have been reported from Delhi, India (Manchanda & Bhalla, 2006). The sulfonamides are the only group of antimicrobial agents to be no longer of use in treating meningococcal infection (Block & Vazquez, 2006). Low-level resistance to other antimicrobials of choice has been reported (Jackson et al., 1994), but meningococci, unlike other bacterial organisms, seem to be particularly inefficient at developing resistance to antimicrobial agents. Between January 2007 and January 2008, three cases of fluoroquinolone-resistant group B meningococci have been described among residents of the North Dakota-Minnesota border (CDC, 2008). Isolated cases of ciprofloxacin-resistant strains have been reported in Argentina, Australia, China, France, India and Spain (Jorgensen et al., 2005; Chu et al., 2007; Singhal et al., 2007). Ciprofloxacin is utilized for the elimination of carriage; however its effectiveness against fluoroquinolone-resistant strains is unknown.
Immunity to meningococcal disease is known to develop with age. Initially, transfer of maternal antibodies protects infants (Goldschneider et al., 1969a) and this is known to persist for several months. After this, acquisition of serum bactericidal antibodies (SBA) is inversely related to age (Figure 1). In young children aged less than two years, low levels of bactericidal antibodies are observed that correspond with the highest risk of infection. There is a gradual increase in the acquisition of SBA from two years onwards coinciding with a decrease in disease incidence. The acquisition of protective SBA occurs due to the asymptomatic carriage of *N. meningitidis*, whether pathogenic or non-pathogenic. It has also been hypothesized that carriage of *N. lactamica* (Gold et al., 1978) and other species with cross-reacting antigens (Robbins et al., 1972; Guirguis et al., 1985) can also contribute to the acquisition of protective SBA. However, the role of these organisms inducing SBA activity to meningococci has been questioned (Trotter et al., 2007).

Figure 1: Age-related incidence of meningococcal disease in the USA and prevalence of serum bactericidal activity against three pathogenic strains of *N. meningitidis*
The role of SBA in protective immunity to meningococcal disease was confirmed by studies performed by Goldschneider and colleagues in the 1960s (Goldschneider et al., 1969a). This study involved bleeding American army recruits at the start of their basic training, and related the occurrence of meningococcal disease due to capsular group C to the presence or absence of SBA at recruitment. Disease occurred in only 1% of individuals who had SBA at start of the training but in 22% of those who were SBA negative. This was further confirmed by the population study performed by the same authors who demonstrated the inverse relationship between disease incidence and the presence of SBA (Goldschneider et al., 1969a) (Figure 1).

The activation of the complement cascade by antibody is one of the critical mechanisms of immunity to meningococci. Evidence of this is provided by studies of individuals who have a deficiency in the complement cascade and are more susceptible to meningococcal disease (Nicholson & Lepow, 1979). Individuals with late complement component deficiency (C5–C9) are at a greater risk of infection and also recurrent attacks (Ross & Densen, 1984; D’Amelio et al., 1992). Infections in such individuals differ from those in immunocompetent individuals as they usually occur at an older age and may involve a rare capsular group (Petersen et al., 1979; Ross & Densen, 1984; Fijen et al., 1989; D’Amelio et al., 1992). Those individuals with deficiency in the properdin or factor D, both components of the alternative complement pathway, have a much higher case-fatality rate (Ross & Densen, 1984; Densen, 1991), but there is rarely a recurrence of infection. Hence the alternative complement pathway also has a role to play in the immunity to meningococci.

Studies have illustrated that the prevalence of SBA specific for group C is lower than was reported by Goldschneider and colleagues in the 1960s, with 10% of adolescents in British Columbia with SBA titres ≥ 4 (Mitchell et al., 1996) and 10%–30% of non-immunized adults in the United Kingdom (Jones et al., 2000; Trotter et al., 2003). In the study of military recruits (Goldschneider et al., 1969a), more than half of the recruits were negative for SBA and were colonized with the circulating epidemic strain but did not develop disease. This suggests that other mechanisms can contribute to immunity to meningococci, such as opsonophagocytosis and antibody-dependent cellular cytotoxicity (Lowell et al., 1980; Halstensen et al., 1989).

Although meningococci colonize the nasopharynx, our understanding of mucosal immunity is still limited. The process of colonization involves a complex interaction between the meningococci and host factors (Bourdoulous & Nassif, 2006). This results in many different factors with the potential to be involved in host defence mechanisms. Secretory IgA and other antibodies are present in nasopharyngeal secretions (Brandtzaeg, 1992) and are likely to contribute to the prevention of carriage. Meningococcal reactive T-cells have been identified in the mucosa, suggesting cellular immunity at this localized site (Davenport et al., 2003). Since carriage of meningococci is hypothesized to induce the development of natural immunity, the interaction at the mucosa will contribute to the generation of both localized and systemic immunity.
3. Immunological assays

3.1 Serum bactericidal antibody assay

Following the studies of Goldschneider and colleagues in the 1960s (Goldschneider et al., 1969a; Goldschneider et al., 1969b), the measurement of SBA in vitro became the gold standard assay for the assessment of immunity to meningococci. The SBA assay allows for the determination of antibody-mediated complement lysis of meningococcal cells, and is therefore a measurement of functional antibodies. The original SBA assays used human complement lacking anti-meningococcal antibodies as the source of exogenous complement. Obtaining suitable human complement involves screening many donors or sources, and quite often is not suitable for universal use against a panel of meningococcal strains. It is also difficult to standardize, and therefore is a factor to be considered when comparing data generated from different laboratories.

In an effort to standardize the assay, a recommended protocol using baby rabbit complement as the exogenous source was published (Maslanka et al., 1997) and adopted by the World Health Organization (WHO) as the recommended assay for the assessment of SBA titres following immunization with meningococcal polysaccharide vaccines (WHO, 1976). Meningococci are known to be more susceptible to complement-mediated lysis in the presence of exogenous rabbit complement compared to human complement (Griffiss & Goroff, 1983; Zollinger & Mandrell, 1983). Studies have shown a positive correlation between titres obtained from an SBA assay using either human or baby rabbit complement (Borrow et al., 2001a; Trotter et al., 2003). Group C conjugate vaccines were licensed in the United Kingdom partly on the basis of immunogenicity data generated using an SBA assay with baby rabbit complement (Miller et al., 2001). For the assessment of future vaccines containing a group C conjugate component or group A conjugate vaccines, assessment of immunogenicity by SBA assay using either human or baby rabbit complement is recommended (WHO, 2004; WHO, 2006).

Human complement is still the preferred choice of exogenous complement for the determination of SBA against group B because the use of baby rabbit complement has been associated with elevated SBA titres due to the presence of low-avidity anti-group B capsular polysaccharide antibody in test sera (Zollinger & Mandrell, 1983; Mandrell et al., 1995). Efforts have been made to investigate the use of non-human complement sources for group B SBA assays but, to date, a viable alternative has not been found (Zollinger & Mandrell, 1983; Zollinger et al., 1997; Findlow et al., 2007b). The difficulties of trying to standardize SBA assays for group B have been highlighted (Borrow et al., 2005a) and efforts to recommend a standardized protocol are underway (Borrow et al., 2006a).
It has become more apparent that the selection of target strain in the SBA assay is of critical importance. Strains for capsular groups A and C have been recommended for use in a standardized SBA assay (Maslanka et al., 1997), but for groups B, W135 and Y there is no consensus on strains. For group B, the diverse epidemiology of prevalent strains and the fact that the antigens inducing protective antibodies are subcapsular, make it necessary to analyse multiple strains in any evaluation of vaccine candidates. The choice of target strain is therefore of importance in that its subcapsular makeup is representative of that of the circulating strain, which is especially relevant for studies of natural immunity. This was illustrated by a study in Niger where the proportion of individuals protected against the outbreak of W135 strain bearing a PorA subtype P1.5,2 increased from 26% to 42% within 10 months, whereas against a reference strain (M.01, 0240070) with a PorA subtype P1.18-1,3, the proportion of individuals protected did not increase significantly (34% to 37%) (Boisier et al., 2006). Studies have highlighted that strains considered to be phenotypically or genotypically similar can result in variable SBA titres from the same serum samples (Vermont et al., 2003; Findlow et al., 2006; Martin et al., 2006; Findlow et al., 2007a). Differences in SBA titres have been observed in different laboratories using the same group B strain (Borrow et al., 2005a). This was attributed to the different methods used to prepare stocks of the strain for use in the SBA assay which resulted in different expression of minor outer membrane proteins. This is a particularly important point because there are group B candidate vaccines currently being evaluated that include outer membrane proteins, and hence to correctly assess the immunogenicity of these products, strain selection is critical.

### 3.2 Measurement of capsular group-specific immunoglobulin

The determination of capsular group-specific immunoglobulin (Ig) provides a measurement of antibody that recognizes the individual capsular groups but does not necessarily reflect the level of functional (protective) antibody. Original assays were performed as radioimmunoassay or precipitation assays and tended to measure total Ig. The only correlate of protection suggested to date that is based on an antibody-binding assay comes from the Finnish efficacy trials of a meningococcal polysaccharide group A vaccine and was determined to be 2 µg/mL Ig by radioimmunoassay (Makela et al., 1975; Peltola et al., 1977). Serological evaluation of vaccines normally includes measurement of capsular group-specific IgG. A standardized enzyme-linked immunosorbent assay (ELISA) for the determination of groups A and C-specific IgG have been recommended (Carlone et al., 1992; Ghodesling et al., 1994). Protocols for measuring capsular group A, W135 and Y-specific IgG have been developed, although there is no consensus on a standardized assay (Élie et al., 2002; Giardina et al., 2003; Joseph et al., 2004; Giardina et al., 2005). A multiplex assay for the measurement of IgG to groups A, C, W135 and Y using a bead-based assay was developed (Lal et al., 2004) which has the advantage of using very small volumes of sera to determine the concentrations of IgG to four groups in one assay and is also more sensitive than a traditional ELISA due to its greater dynamic range. The multiplex assay can also be adapted to include measurement of antibodies to group B or different Ig classes (Laher et al., 2006; de Voer et al., 2008). The measurement of group-specific IgG subclasses has also been described, and used in evaluating response to immunization (Giardina et al., 2003).
The correlation between capsular group-specific IgG and SBA titres is not sufficiently strong to allow measurement only of IgG (Granoff et al., 1998b). It has been suggested that a better correlation may be obtained by using a modified ELISA that is designed to detect only high avidity antibody, which is more likely to be functional and also a measurement of immunological memory (Granoff et al., 1998b). However, as described later (see Section 7 — Effectiveness), the view of the role of immunological memory has changed, and it is widely accepted that measurement of SBA is regarded as the ‘gold standard’ for meningococcal vaccine serological evaluation.

3.3 Opsonophagocytic assays

Phagocytosis of meningococci has been demonstrated (Roberts, 1967; Roberts, 1970; Halstensen et al., 1989) and it is thought that this may contribute to protective mechanisms, especially for group B. The original method of measuring opsonophagocytosis (OPA) was by fluorescence microscopy and visual inspection; however this was very labour intensive and alternative methods have been developed. Chemiluminescence (Sjursen et al., 1992) and flow cytometry (Halstensen et al., 1989; Aase et al., 1995; Aase et al., 1998) protocols have been developed with various end-points using cell lines or freshly isolated peripheral blood mononuclear cells. Different targets can be used in these assays, with either killed meningococci (Aase et al., 1995; Findlow et al., 2006), viable meningococci (Aase et al., 1998) or latex particles coated with capsular polysaccharide or outer membrane proteins (Lehmann et al., 1999). A multiplex flow cytometric assay has been developed that allows determination of the OPA activity to groups A, C, W135 and Y (Martinez et al., 2002). Cell surface labelling assays have also been developed and may be an alternative to the OPA assays (Findlow et al., 2006).

The importance of this mechanism of immunity to protection against meningococcal infection is still unclear. The production of SBA is established as the correlate of protection for group C and is likely to have a similar role for groups A, W135 and Y where the capsular polysaccharide is immunogenic. However, for group B, OPA has been suggested to contribute to protection and consequently has been used in the assessment of immune responses to candidate group B vaccines (Lehmann et al., 1997; Naess et al., 1999; Findlow et al., 2006; Wedege et al., 2007). There is currently no standardized protocol for OPA assays, which is an issue if this type of measurement is to be used in the evaluation of candidate group B vaccines.
3.4 Other assays

Several other immunological assays have been utilized in the assessment of immunological responses to meningococcal infection or vaccination. The whole-blood assay analyses the bactericidal killing observed in an individual’s whole blood rather than just the antibody-mediated killing in serum (Ison et al., 1999). This assay has been used in the study of outer membrane vesicle (OMV) vaccine responses (Morley et al., 2001) and is reported to be more sensitive than the SBA assay (Ison et al., 1999; Morley et al., 2001); however, due to the labour-intensive nature of the assay, the requirement for fresh blood to be analysed and the difficulty in standardizing such an assay, it is more suitable for research purposes or small-scale studies, not evaluation of candidate vaccines. Animal models for the study of meningococcal pathogenesis have been developed and used in the assessment of protection against meningococci. However, as humans are the only natural hosts for *N. meningitidis*, the results obtained using animal models may not be entirely relevant to human disease. Models using mice have been developed (Miller, 1933) and widely used to analyse active immunization or the role of various host factors in pathogenesis, but tend to be sensitive to the level of challenge bacteria and require iron supplementation (Holbein, 1980; Gray-Owen & Schryvers, 1996). Infant rat models have been developed (Saukkonen, 1988) which allow the demonstration of passive protection and can be used to evaluate human sera for protective immunity to group B meningococci (Toropainen et al., 1999). Other assays, such as immunoblots and T-cell assays, are useful research tools but are not suitable for large-scale studies evaluating meningococcal vaccines.
4. Correlates of protection

Laboratory markers of immunity can be used to determine the protection provided by vaccines without the need to perform large-scale efficacy trials. For meningococcal vaccines, the low incidence of meningococcal disease would require tens of thousands of participants to establish vaccine efficacy. These surrogates of protection are derived from evidence demonstrating that their presence reliably predicts clinical protection, and is actually responsible for mediating the protection observed. A surrogate of protection can be derived from studies of natural immunity, demonstration of passive protection from one individual to another, or from phase III efficacy trials. The surrogates of protection for meningococci have been extensively reviewed (Balmer & Borrow, 2004a; Borrow et al., 2005b; Borrow & Miller, 2006b) where more complete details can be found.

The surrogate of protection for group C meningococci was established in the studies of Goldschneider and colleagues (Goldschneider et al., 1969a). These studies demonstrated that the presence or absence of naturally-occurring SBA (a titre of ≥ 4 using human complement) to group C in military recruits, predicted the risk of subsequent group C disease in an individual. Confirmation that the SBA was mediating the protection was demonstrated by removal of group C-specific SBA from sera, which resulted in a loss of bactericidal activity in vitro (Goldschneider et al., 1969b). As discussed (section 3.1), this surrogate was established using human complement in the SBA assay. Studies have been performed to establish an equivalent surrogate of protection using rabbit complement in the SBA (Borrow et al., 2001a; Andrews et al., 2003). Assessment of the level of natural immunity in the United Kingdom prior to the introduction of the group C conjugate vaccine showed a correlation between protection from disease and a rabbit complement SBA (rSBA) titre ≥ 8 (Trotter et al., 2003). A titre of less than eight was correlated with susceptibility and a titre of greater than 128 correlated with protection. For titres between 8 and 64, additional information, such as an hSBA titre ≥ 4 or evidence of antibody avidity maturation, the last being an indicator of successful priming, was needed.

An hSBA titre ≥ 4 is an individual-based surrogate of protection, but to establish this for an rSBA titre ≥ 8 would require a large cohort to be analysed pre- and post-vaccination and then followed up prospectively for occurrence of disease, as in the original Goldschneider study (Goldschneider et al., 1969a). Post-licensure surveillance following the introduction of the group C conjugate vaccine in the United Kingdom has allowed age-specific efficacy estimates to be calculated. An rSBA titre ≥ 8 was observed to correlate with the vaccine efficacy estimate in the United Kingdom for young children (Andrews et al., 2003).
Both an hSBA titre $\geq 4$ and a rSBA titre $\geq 8$ are surrogates of short-term protection. It has been shown in the British experience that rSBA titres wane rapidly in young children following three doses in infancy (Richmond et al., 1999; MacLennan et al., 2000; Borrow et al., 2002). The assumption was that immunological memory may be a predictor of long-term protection (Richmond et al., 1999; Borrow et al., 2002), but data have now indicated that this may not be the case and that persistence of SBA is critical (Trotter et al., 2004; Auckland et al., 2006).

For capsular groups A, B, W135 and Y there is not an established surrogate of protection, although there is some evidence indicating that SBA is a relevant laboratory marker. In the studies of Goldsneider and colleagues (Goldsneider et al., 1969a), an hSBA titre $\geq 4$ to group A and B was observed to predict protection against group C disease. These data suggest that the protection observed in this study incorporated both capsular polysaccharide and subcapsular antigen-specific SBA activity. Further data from studies of naturally-acquired immunity to group A indicate that carriage of group A meningococci induces hSBA activity to both capsular and subcapsular antigens, but in the absence of carriage of group A, any hSBA activity observed naturally is directed to subcapsular antigens (Amir et al., 2005). These data suggest that an hSBA titre $\geq 4$ could be a generic surrogate of protection against meningococcal disease (Borrow et al., 2006b). For group B, hSBA activity has been correlated to clinical efficacy observed with OMV vaccines (Sierra et al., 1991; Holst et al., 2003; Milagres et al., 2003) and has been recommended as a suitable surrogate of protection for predicting the effectiveness of a meningococcal group B vaccine (Borrow et al., 2006a).

Determination of capsular polysaccharide-specific IgG for groups A, C, Y and W135, or antibodies specific to OMV preparations or proteins for group B, are often performed in conjunction with SBA titre measurement. The correlation between non-functional (SBA was not measured) antibody levels and protection was described in Finnish efficacy studies of a group A polysaccharide vaccine where a level of 2 $\mu$g/mL of total Ig determined by radioimmunoassay was the mean level in unimmunized adults (Makela et al., 1975; Peltola et al., 1977). There are no established correlates of protection associated with non-functional antibody levels for the other groups. However, the measurement of specific antibodies (for capsular polysaccharide, OMV or proteins) can be useful in assessing immune responses to immunization for new candidate vaccines.

The laboratory correlate for the induction of immunological memory following meningococcal group C conjugate (MCC) vaccination used to be an SBA titre greater than or equal to that of the primary response one month following a 10 $\mu$g dose of plain polysaccharide administered at least six months after the primary series of immunization (WHO, 2004). However, as depletion of immunological memory and antibody hyporesponsiveness have been observed after a dose of unconjugated meningococcal group C vaccine, particularly in young children, this is no longer recommended (Gold et al., 1979; MacLennan et al., 2000; WHO, 2006). Recommendations for assessing immunological memory now include demonstration of an anamnestic response to a booster dose of conjugate vaccine when administered at least six months after completion of the primary series or changes in the avidity (Goldblatt, 1997) of group-specific IgG from pre- to post-primary series, and before and after a booster dose of conjugate vaccine (WHO, 2006).
5. Available vaccines

Vaccines against meningococcal disease were first developed over 30 years ago, but there is still no vaccine available that provides broad protection against all groups. The plain polysaccharide, conjugate and OMV vaccines available have been implemented in various strategies globally in response to outbreaks or epidemics, or introduced into the routine immunization schedules of selected countries. Plain polysaccharide(s) vaccines are not licensed for use in children under the age of two years.

Details on the licensed vaccines, and vaccines nearing licensure including composition and recommendations for use, are given in Table 1. Polysaccharide vaccines tend to be used in older at-risk groups in bivalent (groups A and C) or quadrivalent (groups A, C, W135 and Y) forms, or as part of the response to epidemics in the meningitis belt as a public-health intervention once the epidemic threshold has been reached (refer to section 1.3 and WHO, 2000b). Bivalent A and C polysaccharide vaccines are stockpiled for emergency use in the meningitis belt. The cost of the polysaccharide conjugate vaccines, monovalent or quadrivalent, restricts the use of these vaccines to developed countries. However, the monovalent group A conjugate vaccine being developed by the WHO/PATH collaboration, aims to provide a product at a cost affordable for the countries of the sub-Saharan region.
Table 1: Currently licensed meningococcal polysaccharide and conjugate vaccines, including those in clinical trial

<table>
<thead>
<tr>
<th>Manufacturer</th>
<th>Vaccine</th>
<th>Active constituents per dose</th>
<th>Adjuvant</th>
<th>Other excipients</th>
<th>Presentation</th>
<th>Licensure</th>
<th>Recommendations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sanofi Pasteur</td>
<td>MengivacTM</td>
<td>50 µg Group A, C polysaccharide</td>
<td>Lactose (2 mg)</td>
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<tr>
<td></td>
<td>MenomuneTM</td>
<td>50 µg Group A, C, W135, Y polysaccharide</td>
<td>Lactose (2.5–5.0 mg); Sodium chloride (4.25–4.75 mg)</td>
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<tr>
<td>GSK Biologicals</td>
<td>AC VaxTM</td>
<td>50 µg Group A, C polysaccharide</td>
<td>Lactose (12.6 mg)</td>
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<tr>
<td></td>
<td>ACWY VaxTM</td>
<td>50 µg Group A, C, W135, Y polysaccharide</td>
<td>Lactose (12.6 mg)</td>
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<tr>
<td></td>
<td>ACW</td>
<td>50 µg Group A, C, W135 polysaccharide</td>
<td>WHO</td>
<td>WHO stockpiled for use in epidemic situation in Meningitis Belt</td>
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<tr>
<td>Finlay Institute</td>
<td></td>
<td>50 µg Group A, C, W135, Y polysaccharide</td>
<td>Freeze dried, 1, 2, 5 or 10 dose/vials</td>
<td>China</td>
<td></td>
<td>Two doses at 6–18 months with 3 months interval; 2 further doses at 3 and 6 years, respectively</td>
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<tr>
<td>Lanzhou Institute;</td>
<td></td>
<td>50 µg Group A polysaccharide</td>
<td>Freeze dried, 1 dose/vial</td>
<td>China</td>
<td></td>
<td>Used for 2 doses at 3 and 6 years in selected provinces</td>
<td></td>
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<tr>
<td>Beijing IBP; Shanghai IBP; Wuhan IBP</td>
<td></td>
<td>50 µg Group A + C polysaccharide</td>
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<tr>
<td>Manufacturer</td>
<td>Vaccine</td>
<td>Active constituents per dose</td>
<td>Adjuvant</td>
<td>Other excipients</td>
<td>Presentation</td>
<td>Licensure</td>
<td>Recommendations</td>
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<tr>
<td>Wyeth Vaccines</td>
<td>Meningite™</td>
<td>10 µg O-acetylated group C oligosaccharide conjugated to 11–25 mg CRM197</td>
<td>AlPO4</td>
<td>Sodium chloride</td>
<td>Single dose, liquid suspension vial</td>
<td>*MRP Participants Australia Brazil Canada New Zealand Singapore Switzerland</td>
<td>Routine: 2 doses in infancy, booster in 2nd year of life or 1 dose at ≥12 months</td>
</tr>
<tr>
<td>Novartis Vaccines</td>
<td>Menjugate™</td>
<td>10 µg O-acetylated group C oligosaccharide conjugated to 11–25 mg CRM197</td>
<td>Al(OH)₃</td>
<td>Mannitol, sodium phosphate buffer</td>
<td>Single dose, freeze-dried, vial reconstituted with diluent</td>
<td>MRP Participants Argentina Brazil Romania Australia Canada Chile Cyprus Dominican Republic Hungary Mexico New Zealand</td>
<td>2 doses in infancy 2 months apart, booster in 2nd year of life or 1 dose at ≥12 months</td>
</tr>
<tr>
<td></td>
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<td></td>
<td></td>
<td>Infants &lt; 12 months, 3 doses from 2 months onwards, interval of at least 1 month Children ≥ 12 months, adolescents and adults, one dose</td>
<td></td>
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<tr>
<td>Manufacturer</td>
<td>Vaccine</td>
<td>Active constituents per dose</td>
<td>Adjuvant</td>
<td>Other excipients</td>
<td>Presentation</td>
<td>Licensure</td>
<td>Recommendations</td>
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<tr>
<td>Baxter Bioscience</td>
<td>NeisVac-C™</td>
<td>10 µg de-O-acetylated group C oligosaccharide conjugated to 10–20 µg tetanus toxoid</td>
<td>Al(OH)₃</td>
<td>Sodium chloride</td>
<td>Single dose, prefilled syringe</td>
<td>MRP Participants</td>
<td>Routine: 2 doses in infancy 2 months apart, booster in 2nd year of life or 1 dose at ≥12 months</td>
</tr>
<tr>
<td>Sanofi Pasteur</td>
<td>Menactra™</td>
<td>4 µg each of groups A, C, Y and W-135 polysaccharide conjugated to ~48 µg of diphtheria toxoid</td>
<td>None</td>
<td>Sodium chloride</td>
<td>Single dose, prefilled syringe</td>
<td>USA</td>
<td>Routine: adolescents 11–12 yrs, 1 dose At-risk groups: 11–55 yrs, 1 dose Canada Routine: not recommended (unless epidemiology warrants use in 11–24 yrs) At-risk: 2–55 yrs, 1 dose; ≥56 yrs 1 dose of Menactra™ or Menomune™</td>
</tr>
<tr>
<td>GlaxoSmithKline</td>
<td>Menitorix™</td>
<td>5 µg Hib polysaccharide and 5 µg group C polysaccharide each conjugated to ~17.5 µg of tetanus toxoid</td>
<td>None</td>
<td>Tris, Sucrose, Sodium chloride</td>
<td>Single dose, freeze-dried vial reconstituted with prefilled syringe</td>
<td>United Kingdom</td>
<td>Routine: booster dose at 12 months</td>
</tr>
<tr>
<td>Manufacturer</td>
<td>Vaccine</td>
<td>Active constituents per dose</td>
<td>Adjuvant</td>
<td>Other excipients</td>
<td>Presentation</td>
<td>Licensure</td>
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<tr>
<td>Serum Institute of India</td>
<td>PsA-TT</td>
<td>10 µg group A polysaccharide conjugate to 10–20 µg tetanus toxoid</td>
<td>AlPO4</td>
<td></td>
<td>0.5 mL</td>
<td>Phase II trials</td>
<td>Target population: 1–29 year olds in sub-Saharan Africa; 1 dose</td>
</tr>
<tr>
<td>GlaxoSmithKline</td>
<td>MenACWY-TT</td>
<td>5 µg of group A, C, W135 and Y polysaccharides conjugated directly to tetanus toxoid (W135 &amp; Y) or via a spacer (A &amp; C)</td>
<td></td>
<td></td>
<td></td>
<td>Phase II trials</td>
<td></td>
</tr>
<tr>
<td>Novartis Vaccines</td>
<td>Men ACWY</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Phase II trials</td>
<td></td>
</tr>
</tbody>
</table>

* Mutual Recognition Procedure (MRP) participating countries; Austria, Belgium, Cyprus, Czech Republic, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Iceland, Ireland, Italy, Latvia, Lichtenstein, Lithuania, Luxembourg, Malta, Norway, Poland, Portugal, Slovak Republic, Slovenia, Spain, Sweden, Netherlands, United Kingdom.
6. Response to immunization

6.1 Meningococcal polysaccharide vaccines

Polysaccharide vaccines are regarded as T-cell independent antigens and elicit SBA responses in the absence of T-cell involvement. Hence these vaccines tend to be immunogenic in older children and adults, but fail to be as immunogenic in young children. This age-related response is likely to reflect intrinsic B-cell maturation, possibly due to natural priming because the antibodies elicited in adults are isotype-switched which is normally representative of a secondary antibody response (Barington et al., 1996; Baxendale et al., 2000; Zhou et al., 2002). Immunization of infants with polysaccharide vaccine can induce anticapsular antibody, but often this is not bactericidal (Lieberman et al., 1996; Campagne et al., 2000), and SBA titres elicited by polysaccharide immunization in older children and adults tend to be greater than those observed in young children (Maslanka et al., 1998; Campagne et al., 2000; Harris et al., 2003). Age-dependent responses to immunization with a quadrivalent ACYW135 polysaccharide vaccine were observed in children and adults in Saudi Arabia (Jokhdar et al., 2004; Al-Mazrou et al., 2005; Khalil et al., 2005). Children aged ≤ 2 years received two doses two months apart, and children aged ≥ 3 years received one dose, with a significant increase in the proportions with a rSBA titre ≥ 8 for each serogroup pre- to post-immunization only observed in those aged ≥ 3 years (Figure 2). Age-dependent responses were also observed in Finnish infants and older children aged three months to 19 years receiving a group A polysaccharide vaccine (Kayhty et al., 1980) (Figure 3). Children aged less than 18 months received two doses at a two month interval, with those aged ≥ 18 months receiving one dose. The proportion of children aged 3–5 months, 6–11 months and 12–17 months achieving antibody concentrations > 2 µg/mL, a proposed correlate of protection for group A (Lepow et al., 1977; Makela et al., 1977) following one dose of group A polysaccharide, was 8%, 41% and 64%, respectively. This increased following a second dose to 36%, 71% and 82% for the same age groups. A greater proportion > 2 µg/mL was observed in children aged ≥ 18 months, with ≥ 87% exceeding this concentration.
Children aged 6–18 months received two doses of ACYW135 polysaccharide vaccine two months apart; those aged 24 months to 48 months received one dose of ACYW135 polysaccharide vaccine; those aged 10–29 years received one dose of AC polysaccharide vaccine.

*Adapted, by permission of the publishers, from Al-Mazrou et al., 2005, and Khalil et al., 2005.*
The antibody response following polysaccharide immunization is usually present after ten days and is traditionally assessed by two to four weeks post-immunization (Zangwill et al., 1994; Borrow et al., 2001b). In adults, the SBA declines over the next two years, but remains above baseline for approximately ten years (Zangwill et al., 1994). The decline is more rapid in children with group C SBA titres, returning to baseline by one year in 18 month to five year olds (Espin Rios et al., 2000). Gambian children given two doses at three and six months had SBA titres similar to an unimmunized cohort at 18–23 months of age and also three years later following a further dose at 18–23 months (Leach et al., 1997; MacLennan et al., 1999). A fractional dose of quadrivalent ACYW135 polysaccharide vaccine in individuals aged two to 19 years was shown to be immunogenic for groups A and W135 (Guerin et al., 2008). The public-health relevance of this measure was assessed by a WHO advisory group of experts. The group concluded that, in the context of a vaccine shortage, the available evidence strongly suggested that the use of fractional doses of meningococcal polysaccharide vaccines may be a more effective strategy at the population level than the use of full doses for the control of epidemic meningococcal disease (WHO, 2007a). The effectiveness of such a strategy could be evaluated should such a measure become necessary (WHO, 2007c).
Response to re-immunization with meningococcal polysaccharide vaccines is dependent upon the individual polysaccharides. An initial dose of group A polysaccharide primes for a booster response to a second dose of group A polysaccharide, even in young children (Gold et al., 1979; Leach et al., 1997; Jokhdar et al., 2004). However, group A polysaccharide does seem to be different to the group C, W135 and Y polysaccharides in that it is not strictly acting in the traditional T-cell independent manner associated with polysaccharide antigens. There are extensive data published on antibody hyporesponsiveness to group C polysaccharide immunization in all age groups (Gold et al., 1975; Leach et al., 1997; MacDonald et al., 1998; Granoff et al., 1998a; Richmond et al., 2000; Borrow et al., 2000b; Jokhdar et al., 2004). Hyporesponsiveness is when the immune response is of lower magnitude following a second or more dose of vaccine as opposed to the initial dose. As mentioned above, one dose of group C polysaccharide vaccine is immunogenic in older children and adults but it has been demonstrated that it can also impair the subsequent immune response to a dose of conjugate vaccine (Southern et al., 2004; Keyserling et al., 2005; Vu et al., 2006a). The disparity in the responses to repeated doses of the individual polysaccharides reflects potential differences in the respective mechanism(s) of immunogenicity which currently remain poorly understood. The clinical relevance of antibody hyporesponsiveness is unknown because polysaccharide vaccines have been extensively used with no reported increase in risk of acquiring meningococcal disease (Jackson et al., 1995; Rosenstein et al., 1998; De Wals et al., 2001).

6.2 Meningococcal polysaccharide-protein conjugate vaccines

To overcome the problem of the T-cell independent nature of meningococcal polysaccharide vaccines, and the lack of immunogenicity in infants and young children, the successful approach of conjugating the polysaccharide to a protein carrier applied to Haemophilus influenzae type b (Hib) vaccines was taken. These conjugate vaccines stimulate T-cells to provide the necessary co-stimulation for B-cells, improving the immunogenicity of the polysaccharide even in infants who are unresponsive to unconjugated polysaccharide. Conjugate vaccines also stimulate the induction of immunologic memory through the generation of long-lasting memory B-cells (Goldblatt, 1998; Joseph et al., 2001; Richmond et al., 2001a; Kelly et al., 2005; Kelly et al., 2006) which, upon challenge with meningococcal polysaccharide, produce a rapid antibody response at a greater magnitude than that of the primary response, and the antibody tends to be of increased avidity. Commonly-used carrier proteins include tetanus and non toxigenic natural variant of diphtheria toxin (CRM197).

6.2.1 Monovalent group C conjugate vaccines

In 1999, the United Kingdom became the first country to introduce meningococcal group C conjugate vaccine into the childhood immunization schedule, together with a catch-up campaign up to the age of 24 years (Miller et al., 2001; Chief Medical Officer et al., 2002). Extensive pre-licensure studies had demonstrated the safety and immunogenicity of this vaccine in infants (Richmond et al., 1999; MacLennan et al., 2000; Richmond et al., 2001a) and toddlers (Richmond et al., 2001b). No large-scale efficacy trials were performed, and the vaccines were granted licensure by the demonstration of immunogenicity relative to the accepted surrogate of protection (Goldschneider et al., 1969a; Goldschneider et al., 1969b) and the known efficacy of the polysaccharide vaccine in children aged > 2 years.
The group C conjugate vaccines were observed to be highly immunogenic in infants, stimulating high rSBA titres after two doses, as shown in Figure 4. The proportion of infants with rSBA titres $\geq 8$ was $\geq 98\%$ after two doses, irrespective of the vaccine formulation (Fairley et al., 1996; Richmond et al., 1999; Borrow et al., 2000a; Richmond et al., 2001a). The SBA geometric mean titres (GMTs) observed in infants were similar to those observed in adults receiving one dose of polysaccharide vaccine (Richmond et al., 2000). A trial conjugate vaccine containing a group A and C component was observed to be immunogenic in infants from Niger following three doses at 6, 10 and 14 weeks (Campagne et al., 2000). Upon challenge with plain polysaccharide, a rapid secondary response was observed, which is indicative of the development of immunological memory. Toddlers receiving one dose of MCC had high rSBA titres following vaccination, with $\geq 91\%$ achieving an rSBA titre $\geq 8$ (Richmond et al., 2001b). Immunization of pre-school children (3.5–6 years) and school leavers (13–18 years) in the United Kingdom with one dose of group C conjugate vaccine, resulted in high GMTs and virtually all subjects achieved protective rSBA titres $\geq 8$ (Burrage et al., 2002). Group C conjugate vaccines are highly immunogenic in adults, with significant increases in GMT and fourfold increases observed before, and one month following, vaccination (Richmond et al., 2000; Borrow et al., 2001b; Goldblatt et al., 2002).

**Figure 4: Serogroup C rSBA GMT (95% Confidence Intervals) after MCC vaccines at 2, 3 and 4 months of age and after a polysaccharide challenge in United Kingdom infants**

![Figure 4: Serogroup C rSBA GMT (95% Confidence Intervals) after MCC vaccines at 2, 3 and 4 months of age and after a polysaccharide challenge in United Kingdom infants](image)

Adapted, by permission of the publishers, from Borrow et al., 2000a, Fairley et al., 1996, Richmond et al., 2001a, and Richmond et al. 1999.
6.2.1.1 Persistence of antibody following MCC vaccination

Infants immunized at 2, 3 and 4 months have high SBA titres one month after the third dose but this declines during the first year of life (Richmond et al., 1999) with between 47% to 70% of infants still having protective rSBA titres ≥ 8 at 12–13 months of age (Richmond et al., 2001a; Borrow et al., 2003). In children aged 3.3 years and four years of age who had received three doses of MCC in infancy, 61% had hSBA titres ≥ 4 (Vu et al., 2006b) and 12% had rSBA titres ≥ 8 respectively (Borrow et al., 2002). Analysis of longer-term persistence of rSBA, up to 4–5 years, in children immunized at 2, 3 and 4 months, has revealed a rapid decline during the first 12 months but the rate of decline is not as great over the next three to four years with approximately 33% of children with rSBA titres ≥ 8 (Figure 5).

A decline in SBA titres has been observed in toddlers following a single dose of group C conjugate vaccine, with 75% and 37% protected (rSBA titres ≥ 8) at six months and 1.2 to 2.7 years post-vaccination, respectively (Richmond et al., 2001b; Snape et al., 2005). Twenty-five percent of children either 1.6 to three years following one dose of MCC vaccine had hSBA titres ≥ 4 (Vu et al., 2006b).
In adolescents, there is only a slight decline in SBA GMT during the initial 12-month period following a single dose of group C conjugate vaccine (Choo et al., 2000). Ninety-six percent of adolescents vaccinated at 11–15 years with one dose of MCC vaccine have rSBA titres ≥ 8 at one to two years post-immunization (Borrow et al., 2007). In a study of healthy adolescents aged 11 to 20 years, previously immunized with MCC vaccine five years previously at age 6 to 15 years, 84.1% (95% CI 81.6% to 86.3%) of 987 participants had a rSBA of ≥ 8. The rSBA GMTs were significantly lower in 11 to 13 year olds, 147 (95% CI 115 to 188) as opposed to 14 to 16 year olds, 300 (95% CI 237 to 380) and 17 to 20 year olds, 360 (95% CI 252 to 515) (Snape et al., 2008a). Thus SBA GMTs were higher in those immunized at aged 10 years or above as opposed to those immunized before the age of 10 years. Hence, persistence of antibody appears to be age-dependent and is critical for the long-term persistence of SBA effectiveness of the group C conjugate vaccines (refer to section 6.2.5 “Immunologic memory” and section 7.2.1 “Group C conjugate vaccine efficacy”).

6.2.2 Quadrivalent meningococcal polysaccharide-protein conjugate vaccines

A quadrivalent meningococcal conjugate vaccine, including capsular groups A, C, W135 and Y, was licensed for use in 2005 by the Food and Drug Administration (FDA) in the USA (Bilukha & Rosenstein, 2005). The vaccine contains 4 µg of each of the capsular polysaccharides conjugated to a total of approximately 48 µg of diphtheria toxoid protein. In 2007, licensure was approved in Canada for those aged 2–55 years (National Advisory Committee on Immunization (NACI), 2007). The meningococcal quadrivalent conjugate vaccine has been shown to be highly immunogenic in adolescents and adults non-inferior to immunization with the quadrivalent polysaccharide vaccine (Campbell et al., 2002; Bilukha & Rosenstein, 2005; Keyserling et al., 2005; National Advisory Committee on Immunization (NACI), 2007).

There are limited immunogenicity data currently available in children aged less than two years. In one study, infants received three doses of quadrivalent conjugate vaccine at 2, 4 and 6 months of age with between 54% (group C) and 92% (group A) achieving an rSBA titre ≥ 8 (Rennels et al., 2004). In a similar study in toddlers, 78% to 100% achieved a rSBA titre ≥ 8 to each of the four capsular groups following two doses, 6–12 weeks apart (Rennels et al., 2002). One dose of quadrivalent meningococcal conjugate in children aged 2–10 years was observed to induce greater rSBA titres for all four capsular groups compared to those children receiving one dose of quadrivalent polysaccharide vaccine (Pichichero et al., 2005). The number of subjects with a ≥ 4-fold rise in baseline rSBA titres (from < 8) was also greater in children receiving one dose of quadrivalent conjugate vaccine. In a study of children aged 2 to 10 years of age in Chile, quadrivalent conjugate vaccine was found to be non-inferior to one dose of quadrivalent polysaccharide vaccine with between 66% (group Y) and 92% (group W135) of children receiving either vaccine achieving a ≥ 4-fold rise in rSBA one month following vaccination (Lagos et al., 2005).
SBA persists in children aged two to 10 years of age six months after one dose of quadrivalent conjugate vaccine at a greater level than those who had received one dose of quadrivalent polysaccharide (Pichichero et al., 2005). However, waning of SBA levels at six months following vaccination was evident in both groups. Higher rSBA titres have been observed in children (24–36 months of age in the initial study) two years following a dose of quadrivalent conjugate vaccine, compared to age-matched vaccine naive children. Persistence of antibody in 11–17 year olds three years after initial vaccination was reported to be greater in those receiving meningococcal quadrivalent conjugate vaccine compared to a quadrivalent polysaccharide vaccine (Keyserling et al., 2005).

Phase II trials of a new meningococcal quadrivalent A, C, W135 and Y tetanus toxoid conjugate in children aged 12–14 months and 3–5 years have now been reported (Prieler et al., 2007). The vaccine contains 5 µg of each capsular polysaccharide conjugated directly to tetanus toxoid (W + Y) or via a spacer (A+ C). An interim analysis showed that one month following one dose of meningococcal A, C, W135 and Y tetanus toxoid conjugate vaccine, 100% of children aged 12–14 months and 3–5 years had rSBA titres ≥ 8 for all capsular groups.

A meningococcal quadrivalent A, C, W135 and Y CRM_{197} conjugate has also been developed. In a study in which two to ten year old children were randomized 1:1 to receive either the quadrivalent conjugate vaccine or quadrivalent polysaccharide vaccine as a single-dose schedule (Black et al., 2006), an interim analysis from 311 subjects showed significantly higher proportions of subjects with hSBA titres ≥ 4 in the quadrivalent conjugate cohort versus the polysaccharide cohort; for group A 80% versus 45%, group C 94% versus 71%, group W135 92% versus 66% and group Y 92% versus 66%. The immunogenicity of this quadrivalent conjugate vaccine has also been assessed in toddlers aged 12–23 months as a single dose compared to 3–5 year old recipients of quadrivalent polysaccharide vaccine as a single-dose schedule (Shinefield et al., 2006). An interim analysis of 91 toddlers vaccinated with the conjugate vaccine versus 35 children vaccinated with polysaccharide vaccine demonstrated significantly higher proportions of subjects with hSBA titres ≥ 4 for group A 82% versus 54% and for group C 90% versus 43%, but similar proportions for group W135 86% versus 80% and group Y 66% versus 60%. A study in British and Canadian infants, who received either quadrivalent conjugate vaccine or MCC at two and four months of age, demonstrated that over 80% of quadrivalent conjugate vaccine recipients had hSBA titres ≥ 4 following vaccination for groups C, Y and W135 and over 60% for group A (Snape et al., 2008b). The hSBA GMT post-vaccination was higher following MCC 325 (95% CI 198–532) versus quadrivalent conjugate vaccination 52 (95% CI 36–74).
6.2.3 Monovalent group A conjugate vaccines

In 2001, a ten-year grant was awarded to WHO and PATH from the Bill and Melinda Gates Foundation to support the Meningitis Vaccine Project (MVP), with the goal of eliminating meningococcal epidemics in sub-Saharan Africa through vaccination. The candidate vaccine had to meet a defined profile: induce long-lasting immunity after one dose; interrupt transmission (herd immunity); be produced in accordance with the highest standards for quality control, production and rigorous licensure process; be available as quickly as possible, and have enough capacity for approximately 25 million doses per year over 10 years at a cost that would facilitate widespread use in Africa. To fit this profile, a monovalent group A conjugate, manufactured at a cost agreeable to African countries, was agreed upon. The Serum Institute of India, Pune, was selected to manufacture the vaccine (PsA-TT) which contains 10 µg of group A polysaccharide conjugated to 10–20 µg tetanus toxoid and aluminium phosphate as adjuvant. A Phase I study to assess safety, immunogenicity and antibody persistence in adults was performed in India (Kshirsagar et al., 2007). The vaccine was observed to be immunogenic with a ≥ 4-fold increase in rSBA titres in 83% of individuals compared to 72% of the control group who received one dose of meningococcal polysaccharide vaccine A+C. Significantly higher geometric mean concentrations of group A-specific IgG were observed after group A tetanus toxoid conjugate immunization, as compared to bivalent A+C polysaccharide vaccine. After one year, rSBA titres were significantly higher in those receiving group A tetanus toxoid conjugate vaccine in comparison to the control group (Figure 6).

Figure 6: Group A serum bactericidal antibody persistence at weeks 24 and 48 post-vaccination with a group A tetanus toxoid conjugate vaccine in adults

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Following the encouraging results of the Phase I trial, a Phase II trial was performed to assess the responses in African toddlers aged 12 to 23 months. Preliminary data demonstrated significantly greater rSBA titres in toddlers one month after receiving a single dose of group A tetanus toxoid conjugate vaccine compared to a quadrivalent polysaccharide vaccine (Sow et al., 2007). Ninety-six percent of toddlers demonstrated a ≥ 4-fold increase in rSBA titres one month following group A tetanus toxoid conjugate immunization. The assessment of the persistence of rSBA titres has commenced. These data illustrate that the group A tetanus toxoid conjugate vaccine is highly immunogenic in toddlers aged 12 to 23 months in the meningitis belt, and provides further promising results towards the introduction of a monovalent group A conjugate vaccine for sub-Saharan Africa.

6.2.4 Combination vaccines

Combination vaccines provide a useful way to deliver a wide range of vaccines in a single injection. One meningococcal group C and Haemophilus influenzae type b (Hib) tetanus toxoid conjugate vaccine was licensed in the United Kingdom in December 2005 (Electronic Medicines Compendium, 2005). This combination, administered as a single dose, is mainly utilized to boost immunity to meningococcal group C and Hib in toddlers who have previously completed a primary immunization series with other meningococcal group C or Hib conjugate vaccines, such as in the United Kingdom’s childhood immunization programme (Chief Medical Officer et al., 2006). Coadministration of the combined meningococcal group C and Hib conjugate vaccine with the measles, mumps and rubella (MMR) vaccine does not interfere with the immune response, and is safe and well tolerated (Carmona et al., 2006). Limited data are published on the immunogenicity of the combined meningococcal group C and Hib conjugate vaccine as a booster in the second year of life, though large increases in SBA indicating successful priming with induction of immune memory are demonstrated. Whether infants are primed with either a meningococcal CRM<sub>197</sub>-based or tetanus toxoid (TT)-based conjugate, the proportion of infants putatively protected with SBA titres ≥ 8 is similar at 98% to 100% (Electronic Medicines Compendium, 2005). However, if a TT-based conjugate is used to prime in infancy, SBA GMTs and SBA titres ≥ 128 are higher, as opposed to those if primed with a CRM<sub>197</sub>-based conjugate (Electronic Medicines Compendium, 2005). Hib responses are also enhanced when the priming and/or boosting group C conjugate is conjugated to TT (whether as a combined meningococcal group C and Hib conjugate vaccine or a MCC vaccine) (Electronic Medicines Compendium, 2005; Kitchin et al., 2007). The combined meningococcal group C and Hib conjugate vaccine, coadministered with DTaP<sub>3</sub>-HBV-IPV has been demonstrated in a randomized pre-licensure study to induce a higher SBA GMT than a MCC-CRM<sub>197</sub> vaccine, coadministered with DTaP-HBV-IPV/Hib at 2, 4 and 6 months of age (Tejedor et al., 2007). However, a similar study but using a 2, 3 and 4-month schedule demonstrated lower SBA GMTs for the combined meningococcal group C and Hib conjugate vaccine than for a MCC-CRM<sub>197</sub> vaccine (Schmitt et al., 2007).
Other combinations are under development, for example, a Hib-meningococcal bivalent C-Y tetanus toxoid conjugate vaccine which has been investigated as a priming 2, 3, 4-month schedule in Belgium and Germany (Habermehl et al., 2006), as well as an enforcing dose in the second year of life. An Australian study investigated the 2, 4, 6-month schedule, as well as antibody persistence at 12 months of age (Nolan et al., 2007). This combination vaccine is well designed for countries, such as the USA, where capsular groups C and Y cause approximately half of the meningococcal cases (Lingappa et al., 2001; Rosenstein et al., 2001; Pollard & Scheifele, 2004; Kaplan et al., 2006). Of particular importance in the USA is the incidence of meningococcal-disease-peaks during the first year of life, where capsular groups C and Y together account for approximately half of the cases (Rosenstein et al., 1999); thus a combined CY conjugate vaccine administered to infants would, over time, prevent an estimated 48% more cases than a monovalent group C conjugate vaccine (Lingappa et al., 2001).

In order to assist in reducing the burden of capsular group A disease in the meningitis belt, a new meningococcal A and C conjugate vaccine combined with DTwP, hepatitis B and Hib (DTwP-HBV/Hib-MenAC) was developed. This vaccine had been shown to be well tolerated following primary vaccination at 2, 4 and 6 months of age (Kerdpanich et al., 2008) but was subsequently withdrawn (http://www.emea.europa.eu/pdfs/human/non_eu_epar/globorix/withdrawalletter.pdf).

The WHO meningococcal disease strategy for the African meningitis belt is, initially, to prioritize mass vaccination campaigns (catch up) with a monovalent meningococcal conjugate vaccine in children older than one year of age. The DTwP-HBV/Hib-MenAC vaccine was intended for primary vaccination of infants in their first year of life and as a booster immunization during their second year of life. Thus, this DTwP-HBV/Hib-MenAC vaccine did not fit into the current WHO vaccination strategy.

Other combination vaccines have been studied containing meningococcal conjugate, for example, with pneumococcal conjugates, but with reduced immunogenicity to the meningococcal portion. A randomized controlled study in British infants (at ages 2, 3 and 4 months) of a combination pneumococcal-meningococcal CRM197-based conjugate vaccine as compared to a MCC-CRM197 demonstrated reduced group C meningococcal immunogenicity (Buttery et al., 2005). Combination vaccines are of importance for the future as more vaccines are added into the infant immunization schedule. Over the next years, the repertoire of combination vaccines that include meningococcal antigens will expand.
6.2.5 Immunological memory and herd immunity

Concerns had been raised with the possibility that, due to meningococcal disease occurring very rapidly after acquisition of the pathogen (Edwards et al., 1977), immune memory may not protect. It is well established that, following an accelerated 2, 3 and 4-month schedule of group C conjugate vaccine, specific antibody levels and rSBA titres wane rapidly (Richmond et al., 1999; MacLennan et al., 2000; Borrow et al., 2002). In the toddler cohort two years after a single dose of group C conjugate vaccine, rSBA titres $\geq 8$ were only demonstrated in 37% toddlers (Snape et al., 2005). However, in 13 to 16 year olds who received a single dose of group C conjugate vaccine as part of the United Kingdom catch-up campaign, rSBA GMTs remained elevated for the first 1.8 years (Borrow et al., 2006b). In a study of 274 13 to 15 year olds vaccinated at least three years previously with a single dose of MCC, 75% of subjects had hSBA titres of $\geq 8$ (Snape et al., 2006). Waning of antibody therefore appears to be age dependent, with poorer persistence in the younger age groups in whom efficacy declines most rapidly. This raises the question of whether long-term protection requires the persistence of SBA, rather than relying on its rapid production as a result of immune memory after exposure to the meningococcus. In infants vaccinated in either a 2, 3, 4 or 2, 4, 6-month schedule, protection wanes with the decline of SBA and although immune memory, as evidenced by the ability to mount a booster response is present (Borrow et al., 2002), it does not appear to provide protection from disease (Trotter et al., 2004). This failure is not specific to group C conjugate vaccines as a similar failure to protect by Hib conjugate vaccines, despite the presence of immune memory, has been shown when given under the accelerated 2, 3 and 4-month schedule (Goldblatt et al., 1998; Goldblatt et al., 1999; Ramsay et al., 2003b; Ramsay et al., 2004).

Studies in neonatal murine models investigating responses to human infant vaccines have shown that the neonatal bone marrow has a limited capacity to support the establishment of long-lived antibody secreting plasmocytes (Pihlgren et al., 2001) which are important for the maintenance of circulating antibody. It is not currently known whether the induction of long-lived plasma cells is also limited in human infants, but short-lived antibody responses are a hallmark of early-life immunization with meningococcal group C conjugate vaccines. Thus immune memory, in the absence of circulating antibody, is not sufficient to confer full protection against encapsulated bacteria, such as meningococci, that require functional antibodies rapidly after exposure.

Although meningococcal polysaccharide vaccines only produce a short-term, transient effect on carriage (Hassan-King et al., 1988), group C conjugate vaccines have been shown to significantly reduce the prevalence of group C carriage (Maiden & Stuart, 2002) which has been corroborated by a similar reduction in the attack rate in unvaccinated individuals (Ramsay et al., 2003a). In developed countries, meningococcal carriage is low in childhood although it starts to increase from five years of age, peaking in 15 to 19 year olds (Gold et al., 1978; Cartwright et al., 1987); therefore catch-up campaigns including the teenage years have a dramatic impact on the carriage of group C meningococci, leading to herd immunity. Mucosal antibodies are elicited following conjugate vaccination, and these may play a role in prevention of acquisition of carriage of group C strains (Borrow et al., 1999; Zhang et al., 2001).
6.2.6 Clinical risk groups

6.2.6.1 Asplenia

Asplenic individuals are known to be at increased risk of infection with encapsulated bacteria such as N. meningitidis (Eraklis et al., 1967; Krivit, 1977). The immune response of 103 asplenic individuals in the United Kingdom has been investigated following MCC vaccine (Balmer et al., 2004b). Asplenic individuals had significantly lower SBA GMTs when compared to an age-matched control group. However, 80% of asplenic individuals achieved the putative protective SBA titre ≥ 8. A significant reduction in SBA GMT or the number of subjects with a SBA titre ≥ 8 was observed if the reason for splenectomy was a medical cause, or if vaccination occurred < 10 years after splenectomy.

6.2.6.2 Pre-term infants

Pre-term infants are at greater risk of infection than those born at full term due to the relative immaturity of their immune system. A follow up of meningococcal group C disease in the United Kingdom following the introduction of the MCC vaccine found that, of 21 subjects with vaccine failure vaccinated at less than one year of age, two (10%) had a history of prematurity (Auckland et al., 2006). This proportion is similar to that found in a study of Hib conjugate vaccine failures, in which 12% of those vaccinated at less than one year were premature (Heath et al., 2000). Pre-term infants have been shown to generate adequate responses and equivalent protective antibody titres to term infants following administration of routine infant vaccines (Bernbaum et al., 1985; D’Angio et al., 1995; Ramsay et al., 1995; Kristensen et al., 1996). This led to the recommendation that pre-term infants should receive the routine infant immunization schedule in accordance with their chronological age; for example, in Australia (The Australian Immunization Handbook, 2003), the United Kingdom (Department of Health, 2006) and the USA (Saari et al., 2003), provided they are well and there are no contradictions to immunization.

There are, however, several studies of premature infants which report decreased antibody responses to a number of vaccine antigens including, diphtheria, pertussis, tetanus toxoid and Hib (Conway et al., 1993; Kirmani et al., 2002; Heath et al., 2003). However, in the United Kingdom, the immune response of premature infants has been shown to elicit comparable protective SBA titres to term infants following immunization with a monovalent group C conjugate vaccine in the 2, 3, 4-month schedule, when coadministered with either whole-cell pertussis (Collins et al., 2005) or acellular pertussis vaccines (Slack et al., 2001; Slack et al., 2005). An Italian study of a 3, 5 and 11-month booster dose of monovalent group C conjugate vaccine also showed comparable protective SBA titres between pre-term and term infants (Esposito et al., 2007).

6.2.6.3 Human immunodeficiency virus

Meningococcal infection, although rare in human immunodeficiency virus (HIV)-positive individuals, has been observed (Brindle & Simani, 1991; Nitta et al., 1993; Couldwell, 2001; Pearson et al., 2001), with reports of disseminated meningococcal which may present with a variety of clinical manifestations, such as pneumonia or arthritis (Nitta et al., 1993). It has been postulated that epidemics of HIV infection may trigger outbreaks of meningococcal disease. A case-control study in western Uganda demonstrated no association between HIV infection and meningococcal epidemics (Kipp et al., 1992). No association between meningococcal disease and
HIV status was found by Brindle and Simani, who retrospectively reviewed 36 cases of bacteriologically-proven meningococcal disease where HIV serological status was known (Brindle & Simani, 1991). Limited data are available for immune responses following meningococcal vaccination in HIV-infected children. A United Kingdom study in 51 children (median age 7.3 years, range 1.2 to 15.7 years), of whom 74% were on antiviral therapy, reported only 49% of the children having protective SBA titres ≥ 8 following a single dose of MCC vaccine (Ruggeberg et al., 2002). Data were available in 11 of the non-responders who received a booster dose, of whom 63% had SBA titres ≥ 8. Therefore, the immunogenicity of MCC vaccines is compromised in this patient group.

6.2.6.4 **Standard and intensive chemotherapy**

After treatment of patients with acute leukaemia there is a decrease in vaccine-specific antibody and an increased susceptibility to certain vaccine-preventable diseases. In a United Kingdom study of 59 children, median age 5.4 years (range 1.1 to 15.9) with acute lymphoblastic leukaemia (ALL) or acute myeloid leukemia (AML), 96% had rSBA titres ≥ 8 following a single dose of group C conjugate vaccine administered ≥ 6 months following standard chemotherapy (Patel et al., 2007a). Before revaccination, only 12% had rSBA titres ≥ 8. Therefore protection can be achieved following revaccination of children after completion of standard chemotherapy for acute leukaemia.

Another United Kingdom study demonstrated that all 38 children after hematopoietic stem cell transplant (HSCT) had rSBA titres ≥ 8 following revaccination with group C conjugate at a median age of 13 years of age (range 3.7 to 18.8) (Patel et al., 2007b). Only 11% had rSBA titres ≥ 8 before revaccination. Thus, revaccination of paediatric HSCT recipients provides good levels of protection.

6.2.6.5 **Malaria, malaria chemoprophylaxis and sickle-cell trait**

The immune response to capsular polysaccharide antigens, including meningococcal polysaccharide, is suppressed by malaria (Williamson & Greenwood, 1978). Antibody levels are better maintained in children who are receiving chemoprophylaxis for malaria as opposed to those who are not, as malaria increases the γ-globulin turnover (Cohen et al., 1961). Two years following vaccination with a bivalent meningococcal A/C polysaccharide vaccine, in Gambian children aged one to three years of age, the mean serogroup A-specific IgG concentrations were significantly higher in 26 children who had received chemoprophylaxis (Maloprim (pyrimethamine/dapsone) once every 14 days during the rainy season) as opposed to 27 children who received placebo (Ceesay et al., 1993). Similar findings have also been found in Nigerian children one to two years of age following meningococcal bivalent A and C polysaccharide vaccine, where the malaria chemoprophylaxis used was chloroquine, with control children receiving vitamin C (Bradley-Moore et al., 1985). Subjects with the haemoglobin genotype AS (sickle-cell trait) have been reported to have higher antibody responses (Greenwood et al., 1980a) and better antibody persistence (Greenwood et al., 1980c) to group C polysaccharide vaccine compared to those with the genotype AA.
7. Effectiveness

7.1 Polysaccharide vaccines

7.1.1 Group C

In 1969 and 1970, two large-scale field trials of group C vaccine were performed among American military recruits (Artenstein et al., 1970; Gold & Artenstein, 1971). Immunization was offered to every fifth man listed on the platoon rosters with the remainder acting as controls. Therefore, a total of 28,245 recruits from both trials received group C polysaccharide vaccination, with 114,481 men included as unimmunized controls. The route of vaccination was either needle or jet gun. During the eight-week follow up, 73 culture-confirmed cases of group C occurred among the controls (an attack rate of 0.64 per 1000) as compared to two cases among the vaccinated, an attack rate of 0.07/1000 (excluding one case that occurred nine days after vaccination). Vaccine efficacy from the combined data set was 89%. The protection induced by the group C polysaccharide vaccine was group-specific. In the first trial there was no reduction in disease due to group B in the vaccinated group, while in the second trial, there were no cases of group B disease in the vaccinated or control groups. From 1972, meningococcal polysaccharide vaccine has been administered to all incoming American recruits, and meningococcal disease caused by strains with capsular groups contained in the vaccine has been eliminated as an important health problem in the American army. Routine immunization of Italian military personnel from 1987 with meningococcal polysaccharide vaccine has also resulted in good control of disease (Biselli et al., 1993).

The first evaluation of efficacy of group C polysaccharide vaccination in young children was conducted as a prospective field trial in 1974 during a large group C epidemic in São Paulo, Brazil (Taunay et al., 1974; Taunay et al., 1978). Approximately 67,000 children aged 6 to 36 months were randomized to receive either a dose of group C polysaccharide vaccine or, as a control, diphtheria-tetanus toxoid. During the 17 month follow up, there were 31 culture- or serologically-confirmed cases among the vaccinated group and 45 cases among the controls (efficacy = 31%, 95% CI 11 to 58). For children aged 6 to 23 months, vaccine efficacy was 12% (95% CI 55 to 62). For children 24 to 36 months, vaccine efficacy was 55% (95% CI 4 to 72). Children aged two years in this study showed significant anti-group C Ig antibody responses, as measured by the Farr assay (Farr, 1958) to vaccination (Amato Neto et al., 1974). The low efficacy in this trial has since been attributed to the possibility that the vaccine used was sub-optimal (Frasch, 1995).
The efficacy of group C polysaccharide has been evaluated post-licensure. In Quebec, Canada, approximately 1.6 million doses of meningococcal polysaccharide vaccine were administered to persons aged 6 months to 20 years, in response to an increase in the incidence of meningococcal group C disease. The incidence rates of group C disease among vaccinated and unvaccinated populations were compared, and for children less than two years there were eight cases in the vaccinated group and none in the unvaccinated group ($P = 0.38$) (De Wals et al., 2001). Vaccine efficacy estimated for this age group therefore, was negative, but with very wide confidence intervals. In children aged two to nine years, vaccine efficacy was 41% (95% CI 106 to 79) while among subjects aged five to 20 years, vaccine efficacy was 65% during the first two years of follow up (95% CI 20 to 84). This fell to 0% between three and five years after vaccination (95% CI 5 to 65). The efficacy of group C polysaccharide vaccination has been shown to be age-related, being low in infants and young children while high in adults. The exact age when children develop protective efficacy remains unknown.

### 7.1.2 Group A

There have been seven controlled field trials, all conducted in the 1970s, measuring the efficacy of meningococcal group A polysaccharide vaccines in Egypt (Wahdan et al., 1973; Wahdan et al., 1977), Finland (Mäkelä et al., 1975; Peltola et al., 1977), Sudan (Erwa et al., 1973) and Upper Volta and Mali (Saliou et al., 1978). The interpretation of these trial data is hampered by the small number of cases and also the short duration of observation. Even so, high levels of vaccine efficacy were observed in all studies in all age groups, including infants.

The effectiveness of the group A polysaccharide vaccine has also been reported from observational studies performed during mass immunization campaigns in epidemic conditions. The effects of the vaccine on the course of a group A epidemic in Bamako, Mali, between January and April 1981 were reported (Binkin & Band, 1982). Almost two-thirds of the estimated 671 000 population of Bamako were vaccinated with a single dose of bivalent group A and C polysaccharide. Those aged one to 30 years were targeted. The vaccine was effective in limiting further spread of the epidemic, and the attack rate among those who received vaccine was lower than that in the unvaccinated (0.7/10 000 versus 4.7/10 000).

In 1979, Nigeria suffered a group A epidemic for the third year in succession. Due to limited vaccine supplies, immunization was only carried out in children over one year of age from villages who had at least two cases of meningitis. A population of 10 000 was immunized with a group A and C polysaccharide vaccine, and subsequently there were 10 cases of meningococcal disease. Of these 10 cases, two occurred in vaccinated individuals, but were not vaccine failures, as their symptoms commenced on the day of vaccination (Greenwood & Wali, 1980b).
Vaccine effectiveness was also studied in a group A epidemic in Auckland, New Zealand in 1985–1986 (Lennon et al., 1992). The age group targeted for vaccination were those aged three months to 13 years, with special emphasis on reaching populations at highest risk (Maori and Pacific Island Polynesian children). Two doses of monovalent group A polysaccharide vaccine were administered to ages three, to 23 months with those aged two to 13 years receiving a single dose. Although coverage was high, approximately 90% for a single dose whatever the age group, only approximately 26% of children under two years of age received their second dose. There were no cases of group A disease (100% efficacy) observed among children following a single dose of vaccine at 18 months of age or older. Seven cases occurred in those children vaccinated between three and 18 months, with efficacy during the first year of follow up at 52% (95% CI 330 to 95), falling to 16% (95% CI 538 to 90) after one year. Of those children who received their second dose, none developed disease. One of the controlled field trials in Egypt followed children aged six to 15 years for two years following immunization (Wahdan et al., 1977). There was high efficacy for the first year but no efficacy between one and two years. However, these data must be interpreted with caution as a sub-optimal, low molecular weight polysaccharide was used in that study. Efficacy was also shown to wane over time in a retrospective study in Ouagadougou, Burkina Faso, where approximately 103 000 infants and children aged three months to 16 years were vaccinated in response to a group A epidemic (Reingold et al., 1985). The results of case-control studies conducted 1, 2 and 3 years after vaccination indicated overall efficacy of 87% for year one, 70% for year two, and 54% for year three. In the first year after vaccination, age had no effect on the efficacy rate observed, but three years following vaccination, efficacy was only 8% for those children less than four years of age, compared to 67% for children aged four to 16 years of age. It can be concluded from the above data that at least two doses of group A polysaccharide vaccine are required to elicit protection in those aged two years of age, and that the high efficacy of group A polysaccharide vaccine among infants and young children is quite different to that seen for group C polysaccharide vaccines.

7.1.3 Groups Y and W135

The licensure of the quadrivalent meningococcal polysaccharide vaccines was obtained through immunogenicity data based on ≥ 4-fold rises in rSBA responses (WHO, 1980), and from this the efficacy of the group Y and W135 components was inferred. There are no data on the efficacy of groups Y and W135 polysaccharides.
7.2 Conjugate vaccines

7.2.1 Group C

The efficacy of these monovalent MCC vaccines was originally inferred on the basis of the immunogenicity data and extrapolating from the success of conjugate vaccine technology in decreasing Hib disease. Within 12 to 18 months of vaccine introduction, there was a marked decline observed in the number of cases and number of deaths caused by group C disease in the age groups targeted for immunization (Miller et al., 2001; Gray et al., 2006). Formal estimates of age-specific vaccine efficacy in England up to September 2001 were approximately 90% or above for all vaccinated age groups (Miller et al., 2001; Ramsay et al., 2001). No notable change occurred in group B disease during the group C conjugate campaign. When effectiveness was measured again more than one year after vaccination there was a significant decline in effectiveness for infants vaccinated in the routine infant immunization programme (Trotter et al., 2004). The effectiveness in infants vaccinated at 2, 3, and 4 months of age significantly declined from the first year (88 (95% CI 58 to 93)) to up to June 2006 where there was no demonstrable efficacy (7 (95% CI 3733 to 85)). For those infants aged five to 11 months in the catch up who received two doses of group C conjugate vaccine, effectiveness remained high (91 (95% CI 8 to 100)) within the first year and 84 (95% CI 31 to 97) after more than one year. In toddlers, the youngest group to receive a single dose, efficacy declined to 71% (40 to 93) from 89% (64 to 98) in the first year. The confidence intervals for both the infant and toddler effectiveness data are wide and thus the true extent of protection is uncertain. For those three to 18 years of age, efficacy remained high at 92 (95% CI 85 to 96) more than one year on from vaccination.

For those vaccinated following their routine 2, 4, and 6-month schedule in Spain, a similar decline in effectiveness was demonstrated. Vaccine effectiveness in infants fell from 98.4 (95% CI 95.7 to 99.4) within one year from vaccination, to 78.0 (95% CI 3.1 to 95.0) one year following vaccination (Larrauri et al., 2005). Good disease-control has been maintained in England and Wales and Spain, with only low numbers of group C cases. From January 2000 to December 2003 in England and Wales, all cases of group C disease have been followed up (Auckland et al., 2006). There was no evidence of immunodeficiency in the vaccine failures and the case-fatality rate (7.5%) was similar to that in unvaccinated cases (10.6%). Serum bactericidal antibody titres in convalescent sera and the avidity indices in acute sera were significantly higher in vaccine failures than in unvaccinated cases (6.1-fold higher for SBA titres, P = 0.03 and 3.2-fold higher for avidity indices, P = 0.001). This was consistent with an anamnestic response in the vaccine failures, suggesting that group C disease occurred despite the presence of immune memory.

Introduction of group C conjugate vaccine in the province of Quebec City, Canada, was undertaken during 2001 (De Wals et al., 2004a). Approximately 81% of the population between two months and 21 years of age were immunized, of which more than 1.52 million with MCC vaccine and 51 782 with polysaccharide. For immunization of infants at two months of age, three doses were recommended at 2, 4 and 6 months of age; for those aged four to 11 months two doses, and for all those aged one year or over a single dose (National Advisory Committee on Immunization (NACI), 2001). The overall efficacy was 97% in 2002. Following the experience in the United Kingdom and elsewhere, and cost-effectiveness studies (De Wals et al., 2004b), different provinces opted for different MCC schedules (Public Health Agency of Canada, 2007) with most opting for a single dose at 12 months.
In the Netherlands, group C conjugate vaccine was introduced into their routine immunization programme, in September 2002, in a one-dose schedule at 14 months of age. In addition, a catch-up campaign was conducted from June to November 2002 targeting nearly three million children up to 19 years of age. The reasons for commencing at 14 months of age and not immunizing infants were based partially on the evidence of herd immunity as demonstrated in the United Kingdom, as well as a low incidence of group C disease in the under one year olds. Up to February 2007, no vaccine failures have been reported, and the number of group C cases has fallen from 276 in 2001 to four in 2006 (de Greeff et al., 2006; De Greeff et al., 2007). Both the Flanders and Wallonie regions in Belgium also adopted a single-dose strategy from 12 months of age (EU-IBIS, 2007). Australia also introduced a single dose early in 2003 at 12 months of age, with a catch up to 20 years; however, impact was more difficult to assess due to disease incidence already falling before vaccine introduction in New South Wales. Nevertheless, there was over 75% reduction in disease from 213 cases in 2002, to 50 in 2005 (Booy et al., 2007).
8. Safety

8.1 Polysaccharide vaccines

Meningococcal polysaccharide vaccines, whether bivalent A and C or quadrivalent A, C, Y and W135, have been administered to millions of subjects, including military personnel, as part of mass vaccination programmes, certain at-risk groups and international travellers (CDC MMWR, 2000). These polysaccharide vaccines are safe and well-tolerated. Pain and redness at the injection site are the most commonly-reported adverse events (up to 40%). These local reactions are typically of mild severity and last between one and two days. Transient low-grade fever is reported in less than 5% of vaccinees and occurs more commonly in infants (Gold et al., 1975; Peltola et al., 1978). Higher fevers (>38.4°C.) occur in less than 1% of immunized persons. Severe reactions are uncommon (Gold et al., 1975; Peltola et al., 1976; Makela et al., 1977; Peltola et al., 1978; Hankins et al., 1982; Ambrosch et al., 1983; Lepow et al., 1986; Roberts & Bryett, 1988; Scheifele et al., 1994; Yergeau et al., 1996; Aseffa et al., 2007; Bentsi-Enchill et al., 2007), consisting of wheezing or urticaria in an estimated one per 1 000 000 doses, or anaphylaxis in <1 per 1 000 000 doses (CDC MMWR, 2000). There are also rare reports of Guillain-Barré syndrome (GBS) or other neurological disorders, such as optic neuritis, paresthesia or convulsions, with onset temporally associated with vaccination. In most of the reported instances, multiple injections of different vaccines were given to the patients, so the role of meningococcal vaccination was uncertain (CDC MMWR, 2000). Studies of vaccination during pregnancy have not reported adverse effects among either newborns or pregnant women (de Andrade Carvalho et al., 1977; McCormick et al., 1980; Leston et al., 1998); thus it is not deemed necessary to alter recommendations for meningococcal polysaccharide vaccination during pregnancy.

8.2 Monovalent group C conjugate vaccines

During the United Kingdom MCC vaccine campaign, three manufacturers’ vaccines were utilized, each of which had been evaluated for safety in a comprehensive series of pre-licensure studies (Richmond et al., 1999; MacLennan et al., 2000; Richmond et al., 2001a; Richmond et al., 2001b; Southern et al., 2006). From these pre-licensure safety studies, transient headache of mild to moderate severity was the most commonly-reported adverse event with the highest rate (12%) in the first three days after vaccination. Local reactions at the injection site consisted mostly of pain, tenderness and occasional redness. These tended to be of mild to moderate severity, were maximal on the third post-vaccination day, and typically resolved within a day. Pre-existing allergies did not appear to affect reactogenicity (Southern et al., 2006). Post-licensure passive surveillance through reports of adverse vaccine events from health professionals to the United Kingdom’s Medicines Control Agency/Committee on Safety of Medicines indicated a rate of one adverse event per 2875 doses of MCC vaccine distributed in the first 10 months of the British campaign (Medicines Control Agency et al., 2000).
Nearly all of these adverse events consisted of transient headache, local reactions, pyrexia or dizziness. Anaphylaxis was reported at a rate of one per 500,000 doses distributed. While some new reactions to MCC vaccine were identified (such as headache, nausea, vomiting, abdominal pain and malaise in all age groups), these were generally not serious, and the balance of risks and benefits for MCC vaccines was considered overwhelmingly favourable by The Medicines and Healthcare Products Regulatory Agency (MHRA); (http://www.mhra.gov.uk/home/groups/pl-p/documents/websitesresources/con2022528.pdf).

During the enhanced post-licensure surveillance, health professionals were requested to report any suspected reaction to the MCC vaccine, whatever the severity, to the United Kingdom licensing authorities. A possible association was observed between MCC vaccination and convulsions and purpura. Hospital admissions for convulsions and purpura for the period November 1999 to September 2003 in children from the south-east of England were linked to vaccine records for MCC vaccine, diphtheria/tetanus/pertussis and measles/mumps/rubella. A total of 1715 children with convulsions and 363 with purpura were linked to vaccination records. The results showed that there was no evidence of an increased relative incidence of convulsions or purpura, respectively, two or four weeks following MCC vaccination (Andrews et al., 2007).

A study published in 2003 also suggested that the relapse rate of nephrotic syndrome was increased after administration of MCC vaccine (Abeyagunawardena et al., 2003). However, using an active population-based surveillance system, in conjunction with a record-linkage study of MCC vaccination of 53 children with nephrotic syndrome who experienced 162 relapses, there was no association between vaccination and relapse of nephrotic syndrome (Taylor et al., 2007). Five cases of GBS were reported following MCC vaccine during the United Kingdom catch-up campaign when millions of doses were administered. This number of cases in the United Kingdom was considered by the United Kingdom Department of Health to be lower than the expected background rate, and no reports of GBS in association with MCC vaccination have since been received in the United Kingdom.

### 8.3 Meningococcal group A conjugate vaccines

Limited data are currently available from pre-licensure studies. In a Phase I study in Indian adults, the most frequent local and systemic solicited reactions within seven days after vaccination were pain, redness, swelling, headache, fatigue, malaise and arthralgia, which were not different from the licensed meningococcal bivalent polysaccharide or tetanus toxoid control vaccines. No serious adverse events were reported during the one year safety follow-up period (Kshirsagar et al., 2007). In a Phase II study in the Gambia and Mali where 200 toddlers received the group A conjugate vaccine, there was no significant safety issue and, to date, the five SAEs reported were unrelated to the study vaccines (Sow et al. 2007).
8.4 Meningococcal quadrivalent conjugate vaccines

The quadrivalent polysaccharide-diphtheria conjugate vaccine has been administered to over 7000 persons during pre-licensure studies. Vaccination was found to be safe and well tolerated (Campbell et al., 2002; Keyserling et al., 2005; Lagos et al., 2005; Pichichero et al., 2005). In randomized studies of quadrivalent polysaccharide and conjugate vaccines, local reactions were more common in the adolescent conjugate recipients (72.3%) than in the adolescent polysaccharide recipients (34.7%) (Keyserling et al., 2005), but similar for two to 10 year old children in studies in Chile and the USA for conjugate recipients compared to polysaccharide recipients (Lagos et al., 2005; Pichichero et al., 2005). The overall frequency of systemic reactions was similar for both conjugate and polysaccharide recipients in adolescents (Keyserling et al., 2005) and children (Pichichero et al., 2005). Less than 5% of conjugate or polysaccharide recipients experienced serious systemic reactions, defined as high fevers or headache, fatigue, malaise, chills or arthralgias requiring bed rest, anorexia (missing three or more meals), three or more episodes of vomiting, or five or more episodes of diarrhoea, or the presence of rash or seizures.

During March 2005 to April 2007 19 confirmed cases of GBS with onset within six weeks of quadrivalent polysaccharide-diphtheria conjugate vaccination were reported to the Vaccine Adverse Event Reporting System (VAERS) (CDC MMWR, 2006; WHO, 2007b). An increased rate of GBS has not been observed after vaccination with meningococcal quadrivalent polysaccharide or diphtheria toxoids, the two principal components of quadrivalent polysaccharide-diphtheria conjugate vaccine. Although these data suggest a small increased risk of GBS after the administration of quadrivalent polysaccharide-diphtheria conjugate vaccine, the inherent limitations of VAERS and the uncertainty regarding background incidence rates for GBS require that these findings must be viewed with caution. Therefore, additional surveillance and investigation of American cases are planned to determine whether the occurrence of GBS might be causally associated with quadrivalent polysaccharide-diphtheria conjugate vaccination.

8.5 Combination vaccines

8.5.1 Meningococcal group C tetanus conjugate/Haemophilus influenzae type b tetanus conjugate

Few published data are available on the reactogenicity of the meningococcal group C tetanus conjugate/Haemophilus influenzae type b tetanus conjugate combination. From pre-licensure studies after vaccination at 2, 3 and 4 months (Schmitt et al., 2007) or at 2, 4 and 6 months (Tejedor et al., 2007) concomitantly with DTaP-HBV-IPV, redness and drowsiness/irritability were the most commonly-reported local and general solicited symptoms, with no difference seen as compared to the MCC conjugate vaccine group. There were no SAEs deemed to be related to the combined meningococcal group C/Hib conjugate vaccine. There are no published data on the administration of the combined meningococcal group C/Hib conjugate vaccine beyond the second year.
The success of conjugate vaccines has led to the introduction of Hib, pneumococcal and meningococcal conjugate vaccines into the immunization schedules of numerous countries. The development of these conjugate vaccines has raised concerns regarding impairment or interference of responses, as well as safety concerns, but there is also the potential to enhance responses to the coadministered antigens. Few randomized studies have been published on the use of different manufacturers’ conjugates using different carrier proteins, and it is difficult to make comparisons between different studies due to potential differences in batch numbers of vaccines, populations, temporal variations, vaccination techniques, etc.

The utilization of whole-cell pertussis (wP) versus acellular pertussis (aP) has been shown to affect the magnitude of the rSBA GMT of a group C tetanus toxoid conjugate (Kitchin et al., 2007). In this randomized study, infants received either a wP or aP, and each group was further randomized to receive either a MCC-CRM$_{197}$ or MCC-TT vaccine. There were no differences in the proportion of subjects with rSBA titres ≥ 8 following the accelerated 2, 3 and 4-month schedule, but the rSBA GMT was significantly lower for the MCC-TT conjugate when given with aP versus wP. No effect on the group C-CRM$_{197}$ conjugate was demonstrated, regardless of whether aP or wP were coadministered.

A randomized study of pre-school and school leaver cohorts given one of the three MCC vaccines administered either together with, before, or after vaccination with diphtheria-tetanus (DT) or diphtheria toxoid (Td) booster vaccines, showed all three MCC vaccines were highly immunogenic in any combination with DT or Td (Burrage et al., 2002). However, the rSBA GMT of the MCC-TT vaccine was reduced when DT or Td was administered before the conjugate vaccine.
10. Serogroup B vaccines

10.1 Outer membrane vesicle vaccines

Group B meningococci are a major cause of invasive meningococcal disease in the Americas and in many European countries, and over the last 30 years there have been epidemics in Brazil (Sacchi et al., 1992), Chile (Cruz et al., 1990), Cuba (Sierra et al., 1991), New Zealand (Martin et al., 1998) and Norway (Bjune et al., 1991). Epidemics of meningococcal group B disease often commence slowly and then persist for 5–10 years or even longer. The incidence during an outbreak is usually around 5–10 cases/100 000, although it can be much higher in certain age groups, ethnic groups or geographical areas. Most cases during a group B epidemic are clonal and share the same PorA subtype. Therefore, for the development of a vaccine against a single group B strain, the PorA outer membrane protein (OMP) is considered as the most important protein with regard to protection, although, in addition, SBA responses to other OMPs and antigens are also induced (Rosenqvist et al., 1995). OMV vaccines can be tailor-made for the control of a particular epidemic and are usually based on a clinical isolate from the actual epidemic (Holst et al., 2005). Immunization with an OMV vaccine induces SBA activity which correlates with protection (Holst et al., 2003), and the licensure of group B vaccine will be on the basis of SBA activity, together with safety data without large-scale efficacy studies (Borrow et al., 2006a).

The Cuban vaccine which is registered in 19 countries, mostly in Latin America, is formulated from a phenotype B:4:P1.15 strain belonging to the ST32 clonal complex. The vaccine formulation included group C polysaccharide and alum (Sierra et al., 1991). Following two doses of this OMV vaccine in 10 to 14 year olds, the efficacy was found to be 83% (95% CI 42 to 95) (Sierra et al., 1991). The same vaccine was trialled in São Paulo, Brazil, again as a 2-dose schedule, and the efficacy was found to vary with the age group studied (de Moraes et al., 1992). Efficacy was 37% (95% CI 100 to 73) in the three to 23 month olds, 47% (95% CI 72 to 84) in the 24 to 47 month olds and 74% (95% CI 16 to 92) in the four to seven year olds. In Rio de Janeiro, Brazil, the same OMV vaccine showed efficacy of 41% (95% CI 96 to 82) in six to 23 month olds and 14% (95% CI 165 to 72) in 24 to 47 month olds (Noronha et al., 1995).

In Norway, an epidemic of a ST32 clonal complex group B meningococci commenced in the late 1980s. The phenotype of the epidemic and vaccine strain was B:15:P1.7,16. The OMV vaccine was formulated with alum. Efficacy was found to be 57% (95% CI 28 to NR). This OMV vaccine is also being utilized, in a 3-dose schedule, for an outbreak of ST32 phenotype B:14:P1.7,16 disease where around 2700 children aged 12 months to five years were vaccinated in one administrative region (Department of Seine-Maritime) in France (Holst et al., 2005; WHO, 2008).
In 1991 a group B epidemic occurred in New Zealand (Baker et al., 2001). Case numbers and population rates rose from 53 (1.5 per 100 000 population) in 1990 to a high of 650 (17.4 per 100 000 population) in 2001 (Dyet & Martin, 2005). The highest rates of disease occurred in Pacific peoples under 20 years of age, but the highest percentages of cases occurred in European and Maori New Zealanders. The epidemic strain was characterized as B:4:P1.7-2,4, ST-41/44 complex, hence an OMV vaccine prepared from a representative group B strain would be useful for elimination of the epidemic. Through a public-private partnership, an OMV vaccine from the epidemic strain was developed for the New Zealand Ministry of Health, and vaccine trials began with a phase I trial in adults and progressed through to large safety and immunogenicity studies in infants and older children (Oster et al., 2005; Thornton et al., 2006; Hosking et al., 2007; Oster et al., 2007) The vaccine was found to be safe, well-tolerated and immunogenic. The vaccination campaign commenced in July 2004 and, since the introduction of the vaccine, there has been a significant decrease in the numbers of annual cases caused by the epidemic strain (O’Hallahan et al., 2006). The vaccine effectiveness has been estimated, using an observational cohort study and a two-year follow-up period, as 80.0% (95% CI 52.5 to 91.6) for children aged six months to < 5 years and 84.8% (95% CI 59.4 to 94.3) for those aged six months to < 3 years (Galloway et al., 2008).

In order to increase the potential coverage of a particular OMV vaccine, OMVs from two strains have been combined. Two different bivalent OMV vaccines have been studied in clinical trials, one containing OMVs from B:4:P1.19,15 and B:4:P1.7,4 strains (Boutriau et al., 2007), and the other from B:15:P1.7,16 and B:4:P1.7,4 strains (Sandbu et al., 2007). The immune response induced by both bivalent vaccines demonstrated the induction of SBA against the respective homologous strains.

Data relating to the safety of meningococcal OMV vaccines based on their usage in Cuba, France, New Zealand and Norway were reviewed in December 2007 (WHO, 2008). A carefully designed safety-monitoring programme had been set up in New Zealand to ascertain possible SAE following the OMV vaccination of around one million people aged less than 20 years from July 2004 onwards, including 200 000 vaccinees monitored through linkages to hospital admission records. The various surveillance methods used to detect potential SAEs consistently found no evidence of such effects attributable to vaccination. In Norway, although OMV vaccines had never been introduced into routine use because of waning of the meningococcal disease epidemic, there had been extensive use of the vaccine in trials among teenagers and young adults. In some age cohorts, up to 40% of the population had been vaccinated. Despite media reports on possible increased risk of myalgic encephalomyelitis (ME), also called chronic fatigue syndrome, the results from these trials provided no specific causes for concern with respect to SAE. A case–control study had been conducted of ME in Norway, involving all 273 cases in whom ME had been diagnosed at the two major Norwegian referral hospitals, in the 1972–1977 birth cohorts, of whom 201 participated in the case-control study (Magnus et al., 2008). This study had been prompted by media reports of a possible increased risk of ME associated with the use of meningococcal B vaccine. A random sample of 889 controls was drawn from the general population, of whom 389 participated in the study. About 45% of both case and control groups had received meningococcal vaccine, and the study thus provided no evidence of an increased risk of ME associated with vaccination (relative risk = 1.06, 95% CI 0.67 to 1.66). The most widely used OMV vaccine has been that produced in Cuba. Over 55 million doses have been used in Cuba and other Latin American countries over the past 20 years. Data from the original Phase III study involving over 100 000 people showed no evidence that an excess of SAE had been identified in the vaccinated group.
10.2 Expectations for a vaccine with broad coverage against group B strains

In order to broaden further the coverage provided by OMV vaccines, a multivalent PorA vaccine was developed in the Netherlands. The vaccine, containing six different PorA OMPs, was based on two strains, each expressing three different PorA OMPs (P1.7,16; P1.5-1,2-2; P1.19,15-1; P1.5-2,10; P1.12-1,13; P1.7-2,4) (van der Ley et al., 1995; Claassen et al., 1996). It was found to be safe, well tolerated and immunogenic in infants (Cartwright et al., 1999) and toddlers and schoolchildren (de Kleijn et al., 2000). To expand coverage further, a nonavalent OMV vaccine has now been developed adding a third trivalent vesicle (containing PorAs P1.7-2,4; P1.22,14; P1.7-1,1 and P1.18-1,3,6) (Van den Dobbelsteen et al., 2007). This nonavalent PorA OMV vaccine is undergoing clinical evaluation.

An OMV vaccine has also been prepared from a strain of *N. lactamica* (Gorringe, 2005). It has been hypothesized that the development of natural immunity to meningococci in young children may be due to cross-protection induced by carriage of the commensal *N. lactamica* which has many common surface structures with *N. meningitidis* but not PorA or capsular polysaccharide. Thus, immunization with an OMV vaccine that lacks PorA may, theoretically, shift the antibody responses to other antigens that are poorly immunogenic in the presence of PorA but capable of eliciting protective antibodies in its absence. Pre-clinical studies have shown that *N. lactamica* killed whole cells, OMV or OMP pools can induce some protection (Oliver et al., 2002). A Phase I study analysing safety and immunogenicity of three doses of a *N. lactamica* OMV vaccine has been completed, and the vaccine was shown to be safe and elicit increases in SBA responses similar to those seen with a meningococcal OMV vaccine against PorA heterologous strains (Gorringe et al., 2008).

The approach of using PorA-depleted meningococcal OMVs while at the same time upregulating key protective antigens that are reasonably conserved and induce SBA activity, is also undergoing clinical evaluation (Poolman et al., 2006). By upregulating minor antigens such as NspA (a possible adhesin), OMP85 (involved in positioning and folding of other OMPs in the outer membrane), TbpA (part of the transferrin receptor) and Hsf (a possible adhesin), synergy is shown yielding high functional antibody responses (Weynants et al., 2007).

Developments in the meningococcal genomics and proteomics have provided additional approaches to identification of novel vaccine candidate antigens (Pizza et al., 2000; Sun et al., 2000) with the term “reverse vaccinology” being adopted for antigens first identified by computer analysis of the genome (Rappuoli, 2000; Kurz et al., 2003; Capecchi et al., 2004; Mora et al., 2006). Using reverse vaccinology, a large number of novel antigens were identified, five of which were taken forward and expressed in a form suitable for large-scale manufacturing. The antigens included in a five-component vaccine are GNA2132 (Welsh et al., 2003), GNA1030 (Pizza et al., 2000), GNA2091 (Pizza et al., 2000), GNA1870 (also termed factor H binding protein or lipoprotein (LP) 2086) (Masignani et al., 2003; Welsch et al., 2004; Giuliani et al., 2005; Hou et al., 2005; Cantini et al., 2006; Madico et al., 2006) and NadA (Comanducci et al., 2002; Capecchi et al., 2005). When compared to OMV vaccines such as the New Zealand or Norwegian vaccines which induce protective SBA titres against strains with the homologous PorA, the five component vaccine, in mice, induced broader protection that was not PorA-specific (Giuliani et al., 2006). This vaccine has now entered clinical trials.
The lipoprotein LP2086 (also known as factor H or GNA1870) is also being taken forward into a clinical programme as a bivalent vaccine — one from subfamily A and one from subfamily B (Fletcher et al., 2004; Ambrose, 2006).
Meningococcal polysaccharide vaccines for capsular groups A, C, Y and W135 have proved useful in the control of group A epidemics in sub-Saharan Africa (Greenwood & Wali, 1980b; Binkin & Band, 1982) and for group C in the control of disease in the military, and college students (Biselli et al., 1993), but have their limitations in that antibody levels to group A do not persist in children (Reingold et al., 1985), whilst group C polysaccharide is also not efficacious in young children (Taunay et al., 1978). Repeated immunization with group C polysaccharide vaccine results in hyporesponsiveness where antibody levels following a second or third dose are lower than following the initial dose (Gold et al., 1975; Jokhdar et al., 2004) and prior group C polysaccharide vaccination appears to suppress responses to subsequent group C conjugate vaccination, both at one month following MCC vaccination (Richmond et al., 2000; Borrow et al., 2001b) and at six months following vaccination (Southern et al., 2004). Polysaccharide vaccines do not induce immune memory and, most importantly, only produce a short-term, transient effect on nasopharyngeal carriage (Hassan-King et al., 1988).

With the advent of conjugate vaccines which are immunogenic and prime for immune memory even in infants (Richmond et al., 1999), and, of most importance, have been shown to significantly reduce the prevalence of carriage, immunization strategies have been reconsidered (Maiden et al., 2002). Therefore, catch-up campaigns are highly effective in inducing herd immunity where carriage is predominant in cohorts immunized as part of the campaign. Although antibodies persist following group C conjugate vaccination in older children and adults, persistence is poor for those vaccinated in the first year of life (Borrow et al., 2002), and this, together with waning protection (Trotter et al., 2004) following infant vaccination without a booster dose, illustrates the importance of a dose in the second year of life for conjugate vaccines. Group C conjugate vaccines have been hugely successful in dramatically reducing disease in the countries that have instigated immunization programmes together with appropriate catch-up campaigns (Trotter et al., 2004; De Wals et al., 2004a; Larrauri et al., 2005; de Greeff et al., 2006).

11. Implications for immunization
Meningococcal quadrivalent conjugate vaccines are now being implemented into schedules in Canada and the USA (Bilukha & Rosenstein, 2005; National Advisory Committee on Immunization (NACI), 2007). It is hoped that, in populations where these vaccines are utilized, they will generate herd immunity and impact on younger age groups for whom meningococcal quadrivalent conjugate vaccination is not currently recommended. With the development of meningococcal group A conjugate vaccines by the MVP WHO/PATH project, it is hoped that the widespread introduction whereby this conjugate vaccine will be taken to a public-health scale by immunizing the entire population between the ages of one and 29 years of age of the African meningitis belt, starting with Burkina Faso in 2009–2010 (Meningitis Vaccine Project, 2007), will have the same impact as has been demonstrated for the MCC vaccines.
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The World Health Organization has provided technical support to its Member States in the field of vaccine-preventable diseases since 1975. The office carrying out this function at WHO headquarters is the Department of Immunization, Vaccines and Biologicals (IVB).

IVB’s mission is the achievement of a world in which all people at risk are protected against vaccine-preventable diseases. The Department covers a range of activities including research and development, standard-setting, vaccine regulation and quality, vaccine supply and immunization financing, and immunization system strengthening.

These activities are carried out by three technical units: the Initiative for Vaccine Research; the Quality, Safety and Standards team; and the Expanded Programme on Immunization.

The Initiative for Vaccine Research guides, facilitates and provides a vision for worldwide vaccine and immunization technology research and development efforts. It focuses on current and emerging diseases of global public health importance, including pandemic influenza. Its main activities cover: i) research and development of key candidate vaccines; ii) implementation research to promote evidence-based decision-making on the early introduction of new vaccines; and iii) promotion of the development, evaluation and future availability of HIV, tuberculosis and malaria vaccines.

The Quality, Safety and Standards team focuses on supporting the use of vaccines, other biological products and immunization-related equipment that meet current international norms and standards of quality and safety. Activities cover: i) setting norms and standards and establishing reference preparation materials; ii) ensuring the use of quality vaccines and immunization equipment through prequalification activities and strengthening national regulatory authorities; and iii) monitoring, assessing and responding to immunization safety issues of global concern.

The Expanded Programme on Immunization focuses on maximizing access to high quality immunization services, accelerating disease control and linking to other health interventions that can be delivered during immunization contacts. Activities cover: i) immunization systems strengthening, including expansion of immunization services beyond the infant age group; ii) accelerated control of measles and maternal and neonatal tetanus; iii) introduction of new and underutilized vaccines; iv) vaccine supply and immunization financing; and v) disease surveillance and immunization coverage monitoring for tracking global progress.

The Director’s Office directs the work of these units through oversight of immunization programme policy, planning, coordination and management. It also mobilizes resources and carries out communication, advocacy and media-related work.