

Immunogenicity and safety of concomitant administration of meningococcal serogroup B (4CMenB) and serogroup C (MenC-CRM) vaccines in infants: A phase 3b, randomized controlled trial



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ABSTRACT

Background: After implementation of routine infant MenC vaccination, MenB remains a serious cause of meningococcal disease, yet to be targeted by vaccination programs in several countries. This study (NCT01339923) investigated the immunogenicity and safety of MenC CRM-conjugated vaccine (MenC-CRM) concomitantly administered with MenB vaccine (4CMenB).

Methods: Infants (N = 251) were randomised 1:1 to receive 4CMenB and MenC-CRM (Group 1) or MenC-CRM alone (Group 2) at 3 and 5 months (M3, M5) and a booster at 12 months of age (M12), and pneumococcal vaccine at M3, M5, M7, M12. Antibody responses to meningococcal vaccines were measured at M3, M6, M12, and M13. Non-inferiority of MenC-CRM response in Group 1 vs Group 2 was demonstrated at M6 and M13, if the lower limit of the 95% confidence interval (LL95%CI) of the difference in percentage of infants with hSBA titres $\geq 1:8$ was $> -10\%$. Sufficiency of MenB response was achieved if LL95% CI of the percentage of infants with hSBA titres $\geq 1:4$ against fHbp, NadA and PorA strains was $\geq 70\%$ at M6 or $\geq 75\%$ at M13. Adverse events (AEs) were collected for 7 days post-vaccination, and serious AEs (SAEs) and medically attended AEs throughout the study.

Results: Non-inferiority of MenC response in Group 1 vs Group 2 (LL95%CI -6.4% [M6]; -5.2% [M13]) and sufficiency of MenB response in Group 1 (LL95%CI 92%, 90%, 89% [M6]; 97%, 92%, 93% [M13] against fHbp, NadA, PorA, respectively) were demonstrated. Higher rates of mild to moderate solicited AEs were reported in Group 1. Unsolicited AEs and SAEs incidences were similar across groups.

Conclusions: Concomitant administration of MenC-CRM and 4CMenB in infants was immunogenic, resulting in non-inferior responses against MenC compared to MenC-CRM alone and demonstration of sufficient immune response to MenB, after primary and booster vaccination. Reactogenicity was higher for concomitant vaccines administration, but no safety concerns were identified.

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1. Introduction

Neisseria meningitidis is one of the most frequent causes of death in children outside the neonatal period, with the highest disease incidence in infants under 12 months of age [1]. The epidemiology

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of invasive meningococcal disease (IMD) varies regionally and temporally, with six serogroups (A, B, C, W, X, and Y) associated with virtually all cases [2]. Meningococcal disease is endemic in Brazil, predominantly caused by serogroups B and C, with a number of outbreaks occurring since the 1970s [3–8]. In 2010, motivated by the epidemiological situation and the ongoing serogroup C outbreaks, the MenC conjugate vaccine was introduced into the routine infant immunisation schedule in Brazil [9,10]. The introduction of the MenC vaccine into the National Immunisation Programme provided an immediate reduction in incidence

rates of IMD in children aged <2 years, the age group targeted for vaccination [9–11]. However, no early impact was observed in unvaccinated age groups, probably reflecting the lack of a catch-up programme targeting adolescents and young adults—the age groups primarily responsible for carriage and transmission [9]. In children aged <2 years, the estimated incidence of MenB disease is 2.6/100,000 population, with the serogroup causing around 39% of IMD cases in 2013 [12]. Although the incidence of MenB disease in all age categories was reported to decline in the last years [11,13], it still represents a significant burden with potential for outbreaks, such as those seen in several large cities across the country in the late 1980s and 1990s [14].

In May 2015, a multi-component, protein-based serogroup B meningococcal vaccine, 4CMenB, was licenced in Brazil for use in persons aged 2 months up to 50 years [15]. Based on Meningococcal Antigen Typing System (MATS) estimates, the vaccine is predicted to cover 81% (95% confidence interval [CI]: 71–95) of the MenB strains causing IMD in Brazil [16,17]. In this study, we investigated the immunogenicity and safety of concomitant administration of MenC CRM-conjugated vaccine (MenC-CRM) and 4CMenB. The main objective was to evaluate the non-inferiority of the immune response of MenC-CRM when concomitantly administered with the MenB vaccine, compared with the administration of MenC-CRM alone, after primary vaccination and following the booster dose. Other objectives included assessment of the sufficiency of antibody response to 4CMenB after completion of the primary series and following the booster dose, and assessment of reactogenicity and safety of both vaccines.

2. Methods

This study was conducted as part of a Phase 3b open-label study investigating the immunogenicity and safety of the licensed vaccine 4CMenB administered according to reduced schedules in infants or catch-up series in children (clinicaltrials.gov: NCT01339923). This part of the study had procedures and objectives different from the larger study and was carried out between April 2011 and December 2014, at four sites in Brazil. The protocol was approved by the local institutional review boards and ethics committees prior to the start of the trial. The study was conducted in accordance with the Declaration of Helsinki and the principles of Good Clinical Practice. Written informed consent was obtained from a parent or guardian of each infant prior to enrolment in the study.

2.1. Infants

Healthy infants aged 83–104 days were enrolled in the study if they were available for all scheduled visits and their parent/guardian provided written informed consent. Infants were excluded from participation if any of the following criteria were met: previous MenB, MenC or pneumococcal vaccination; previous *N. meningitidis* disease; history of allergy or hypersensitivity to any of the vaccine components; an acute or chronic infection in the 7 days prior to vaccination; fever the day before vaccination, or antibiotic treatment 6 days prior to vaccination; known or suspected alteration of the immune system; severe or chronic disease; receipt of blood products in the 90 days prior to vaccination; receipt of other vaccines in the 7 days prior to vaccination; current participation in other clinical trials; or family or household member of trial staff.

2.2. Study design and vaccines

Infants were randomly assigned in a 1:1 ratio using a web-based randomisation system to receive either 4CMenB and MenC-CRM

(Group 1), or MenC-CRM alone (Group 2), at 3, 5 and 12 months of age (M3, M5 and M12). All infants also received the 10-valent pneumococcal non-typeable *Haemophilus influenzae* protein D conjugate vaccine (PHiD-CV) at M3, M5, M7 and M12. A 0.5 mL dose of 4CMenB (*Bexsero*[™], GSK Vaccines, Italy; lot numbers: 101601H, 101601E, 113001AD, 101601D, 113001AA, IB139201, 101601G, 090101A5, 090101A9) contained 50 µg of each of three purified MenB antigens (factor H binding protein [fHbp], Neisseria heparin binding antigen [NHBA], and Neisserial adhesin A [NadA]), and 25 µg of outer membrane vesicles (OMV) from *N. meningitidis* strain NZ98/254 (PorA). A 0.5 mL dose of the glycoconjugate MenC-CRM vaccine (*Menjugate*[™], GSK Vaccines, Italy; lot numbers: 583011, 938011) contained 10 µg of meningococcal C oligosaccharide, conjugated to CRM₁₉₇. A 0.5 mL dose of PHiD-CV vaccine (*Synflorix*[™], GSK Vaccines, Belgium; lot numbers: SPNA189CN, ASPNA410AH) contained 1 µg of each capsular polysaccharide for serotypes 1, 5, 6B, 7F, 9V, 14, and 23F, and 3 µg for serotype 4 each individually conjugated to protein D of non typeable *H. influenzae*, and 3 µg of capsular polysaccharide of serotypes 18C and 19F conjugated to tetanus and diphtheria toxoids, respectively. MenC-CRM and PHiD-CV were administered intramuscularly into the left thigh, with a spacing of at least 2.5 cm between injection sites. 4CMenB was administered into the right thigh.

2.3. Immunogenicity

Serum samples were taken at pre-vaccination (M3), one month after the second dose (M6), prior to administration of the booster dose (M12), and one month after the booster dose (M13) (Fig. 1). Immunogenicity against MenB was assessed using serum bactericidal antibody assay using human complement (hSBA) against four test strains: H44/76 (fHbp), 5/99 (NadA), NZ98/254 (PorA) and M10713 (NHBA). The study was designed to comply with the European Medicines Agency recommendations for clinical evaluation of the vaccine, but at the time the protocol was established, no suitable indicator strain had been identified for NHBA, and therefore sufficiency of the immune response was only planned to be assessed against the fHbp, NadA and PorA strains. Immunogenicity against MenC was also evaluated by hSBA. Analysis was performed at GSK Clinical Sciences Laboratory, Marburg, Germany (MenC and NHBA test strain) and Public Health England Laboratory, Manchester, UK (fHbp, NadA and PorA test strains). Immune responses were assessed as the percentage of infants with hSBA titres $\geq 1:4$ (MenB test strains) or $\geq 1:8$ (MenC). These thresholds represented the widely accepted surrogates for protection and the established endpoints measurements for clinical trials of meningococcal vaccines [18,19]. Geometric mean titres (GMTs) and ratios (GMRs) were also calculated.

2.4. Safety

Infants were observed for 30 min following each vaccination for any immediate adverse reactions. Local and systemic adverse reactions, and unsolicited adverse events (AEs) were recorded for 7 days following each vaccination. Local reactions were assessed separately for each vaccine in Group 1. Medically attended AEs, solicited reactions persisting after Day 7, AEs leading to premature withdrawal from the study, and serious AEs (SAEs) were recorded throughout the study. Severity of AEs (mild, moderate, severe) and relatedness to the study vaccine (not related, possibly related, probably related) were determined by the investigator.

2.5. Statistical analysis

All analyses were performed on the per protocol set, i.e. all infants who correctly received the vaccinations, provided evalu-

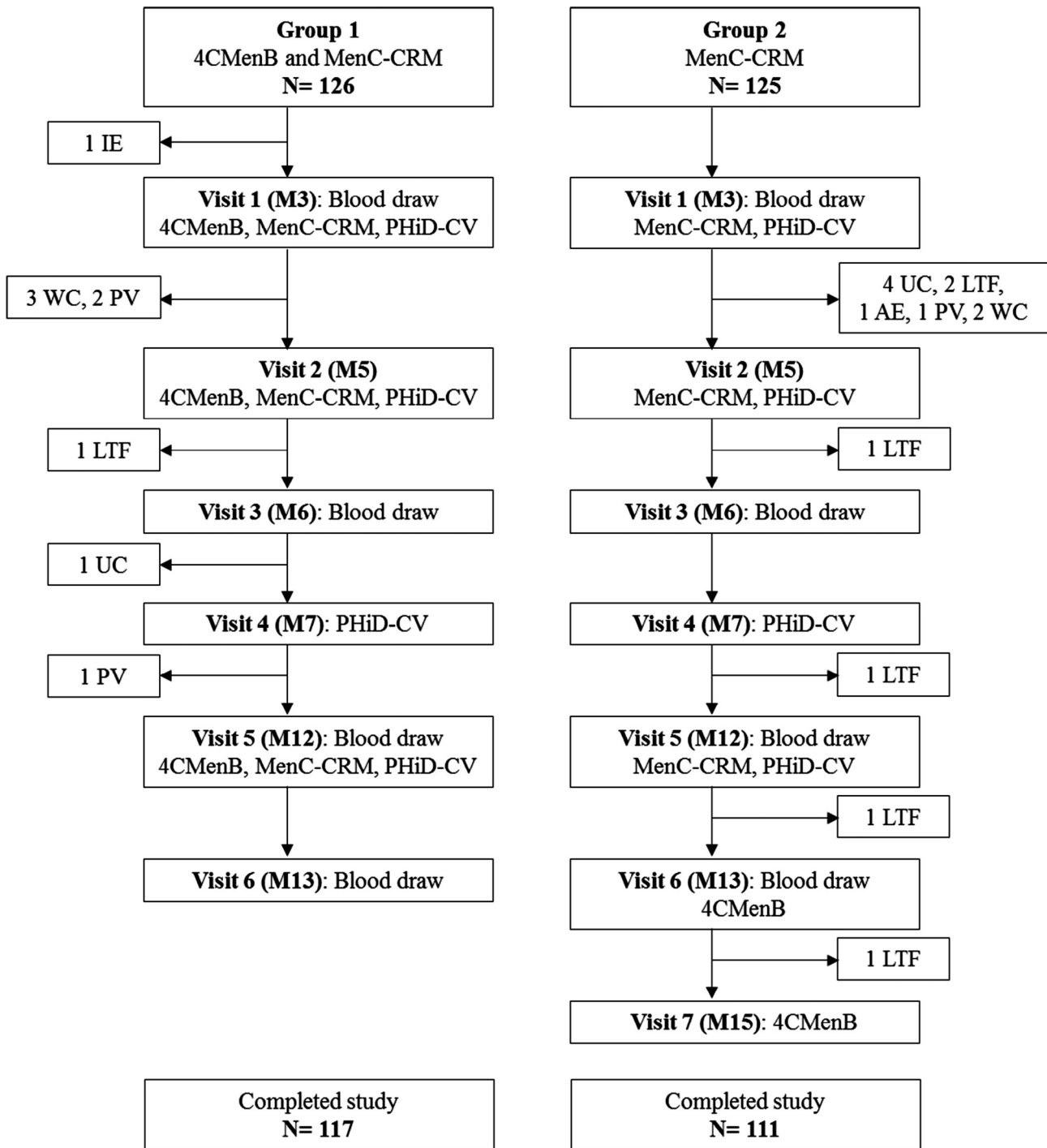


Fig. 1. Study design and subject disposition flowchart. N, number of infants in each group; M, month; PHiD-CV, 10-valent pneumococcal polysaccharide conjugate vaccine; AE, adverse event; IE: inappropriate enrolment; LTF, lost to follow-up; PV, protocol violation; UC, unable to classify; WC, withdrew consent.

able serum samples at one month after the primary series (M6) and had no major protocol deviations. Non-inferiority of concomitantly administered MenC-CRM compared with its separate administration was demonstrated if the lower limit of the two-sided 95% CI of the difference between the percentage of infants with hSBA titres $\geq 1:8$ in the co-administered group compared with the MenC-CRM group was greater than -10% . With a sample size of 100 evaluable infants in each group, the power for demonstrating non-inferiority was 83% after the primary vaccination series, given an underlying responder rate of 95%. The power was 97%

for an underlying responder rate of 98%. For percentages of infants with hSBA titres above the thresholds, GMTs and GMRs, 95% CIs were calculated using the Clopper-Pearson method. Sufficiency of immune response to 4CMenB vaccination was achieved if the lower limit of the two-sided 95% CI for the percentage of infants with hSBA titres $\geq 1:4$ against all three of the fHbp, NadA and PorA test strains was $\geq 70\%$ following the primary series or $\geq 75\%$ following the booster vaccination (Group 1). Safety was evaluated descriptively. All statistical analyses were performed using SAS v9.1 or higher.

3. Results

A total of 251 infants were enrolled in the study, 126 in Group 1 and 125 in Group 2 (Fig. 1). Across the two groups, 89–93% of infants completed the study. The main reasons for not completing the study were withdrawal of consent, lost to follow-up, AE, and protocol violation (Fig. 1). Demographics (age and race) and baseline characteristics (height, weight, and body mass index) for infants in the two groups were very similar, although more females were enrolled in Group 1 (Supplementary Table 1).

3.1. Immunogenicity

Co-administration of MenC-CRM with 4CMenB was found to be non-inferior to administration of MenC-CRM alone, one month after completion of the primary series and before and one month after the booster dose (Table 1), in terms of percentages of responders. Across the groups, 99–100% of infants had hSBA titres $\geq 1:8$ against MenC one month after the second vaccination (primary series), and all infants achieved these titres following the booster dose. Following the primary series, GMTs against MenC in Group 2 (MenC-CRM) were higher than in Group 1 (4CMenB and MenC-CRM) with the 95% CI of the GMT ratio (Group 1 over Group 2) not including 1 (Table 2). Antibody levels increased again following booster vaccination in both groups, but no significant difference between Group 1 and 2 was observed in terms of GMTs against MenC, as shown by overlapping 95% and GMT vaccine group ratios (Table 2).

A robust immune response was seen against MenB test strains. In Group 1, 95–97% of infants achieved hSBA titres $\geq 1:4$ against the fHbp, NadA and PorA test strains after completion of the primary series (Fig. 2). This rose to 97–100% after receipt of the booster dose. Sufficiency of antibody response was achieved against all three test strains after the primary and booster doses (lower limit of 95% CIs were 92%, 90%, and 89% after the primary series and 97%, 92% and 93% after the booster against fHbp, NadA and PorA, respectively). The percentages of infants with hSBA titres $\geq 1:4$,

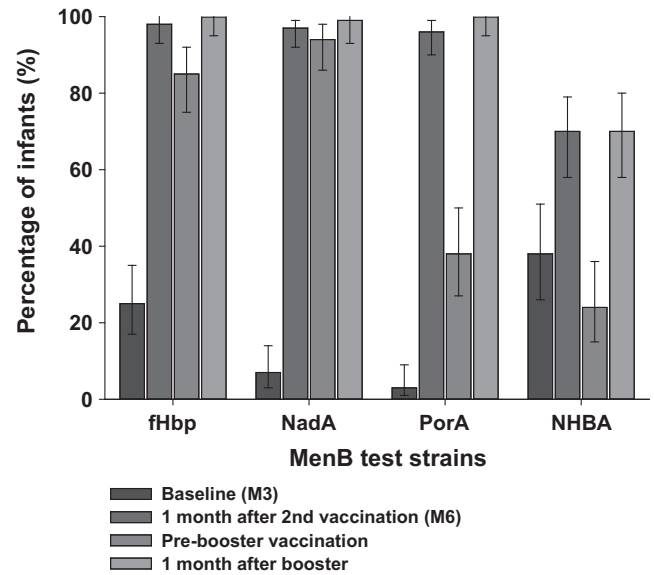


Fig. 2. Percentage of infants in Group 2 (95% confidence intervals) with hSBA titres $\geq 1:4$ against MenB test strains fHbp, NadA, PorA and NHBA, by timepoint (per-protocol cohort).

against the NHBA test strain were 70% (95% CI: 58–79) after the primary series and 70% (95% CI: 58–80) after the booster dose. GMTs for immune responses against the four MenB test strains increased at one month after primary vaccination with respect to baseline values. Antibody levels then declined up to seven months after the receipt of the second vaccine dose, while remaining above baseline levels, except in the case of the NHBA strain. At one month after the booster dose, antibody GMTs were higher than following the primary series, for all four MenB test strains (Supplementary Table 2).

Table 1

Non-inferiority of co-administered 4CMenB and MenC-CRM to MenC-CRM in terms of MenC-CRM immune response, by timepoint (per-protocol cohort).

Timepoint	% of infants with hSBA titres $\geq 1:8$ against MenC				Vaccine group difference % (95% CI)
	Group 1		Group 2		
	N	% (95% CI)	N	% (95% CI)	
M3	67	12% (5%–22%)	51	6% (1%–16%)	6% (–5.5%–17%)
M6	85	99% (94%–100%)	72	100% (95%–100%)	–1% (–6.4%–3.9%)
M12	81	91% (83%–96%)	63	90% (80%–96%)	1% (–8.9%–11.7%)
M13	70	100% (95%–100%)	47	100% (92%–100%)	0% (–5.2%–7.6%)

Group 1, infants receiving 4CMenB and MenC-CRM at 3, 5 and 12 months of age; Group 2, infants receiving MenC-CRM at 3, 5 and 12 months of age; M, month, N, number of infants in each group for whom analyses were carried out; %, percentage of infants with hSBA titres $\geq 1:8$ against MenC; CI, confidence interval. Vaccine group differences are calculated as the difference in percentages between the Group 1 and 2. Bold font indicates that the non-inferiority criterion (lower limit of two-sided 95% CI $> -10\%$) was met.

Table 2

Geometric mean titres (95% confidence intervals) against MenC for infants in Group 1 (4CMenB and MenC-CRM) and Group 2 (MenC-CRM alone), by timepoint (per-protocol cohort).

Timepoint	Antibody geometric mean titres against MenC				Vaccine group ratios (95% CI)
	Group 1		Group 2		
	N	GMT (95% CI)	N	% (95% CI)	
M3	67	2.95 (2.23–3.90)	51	2.48 (1.91–3.22)	1.09 (0.78–1.52)
M6	85	583 (458–742)	72	897 (724–1112)	0.64 (0.46–0.88)
M12	81	39 (30–51)	63	58 (41–83)	0.68 (0.44–1.05)
M13	70	1228 (972–1552)	47	1634 (1208–2209)	0.75 (0.52–1.1)

Group 1, infants receiving 4CMenB and MenC-CRM at 3, 5 and 12 months of age; Group 2, infants receiving MenC-CRM at 3, 5 and 12 months of age; M, month; N, number of infants in each group for whom analyses were carried out; GMT, geometric mean titer; CI, confidence interval. Vaccine group ratios are calculated as the GMT ratios between Group 1 and Group 2.

3.2. Safety

Solicited reactions were reported by all infants in Group 1 and 95% of infants in Group 2 after any vaccination. In both groups, local reactogenicity was higher after the first vaccination than after subsequent vaccinations, and was higher in Group 1 than in Group 2 after each vaccination (Supplementary Table 3). The most commonly reported local adverse reaction following any vaccination was tenderness (Table 2). Rates of reporting of tenderness were similar for both vaccines in Group 1 and lower in Group 2. Most local reactions were mild to moderate and transient in duration (see Table 3).

Similarly to local reactions, rates of systemic adverse reactions were highest after the first vaccination, and higher in Group 1 than Group 2 (Fig. 3). The most commonly reported systemic reaction after each vaccination in both groups was unusual crying. Fever (rectal temperature $\geq 38^\circ\text{C}$) was reported by 39–48% of infants in Group 1 after each vaccination, compared with 10–20% in Group 2. After the second vaccination, rectal temperature $\geq 40^\circ\text{C}$ was reported for one infant in Group 1, resolved within 2 days.

At least one unsolicited AE was reported in 82% of infants in Group 1 and 73% in Group 2. The most commonly reported AE by the Medical Dictionary for Regulatory Activities (MEDRA) preferred term was upper respiratory tract infection (47% and 40% of infants in Groups 1 and 2, respectively). Across groups, 10–11% of infants were reported to have AEs that were judged as possibly related to the study vaccination by the investigator. Most of these AEs were solicited AEs continuing beyond the 7-day collection window following vaccination. SAEs were reported in 4% and 6% of infants in Groups 1 and 2, respectively; none of these were considered related to the study vaccination. There were no deaths in the study.

4. Discussion

In this study, co-administration of MenC-CRM vaccine with 4CMenB was found to generate non-inferior antibody responses to MenC-CRM administered alone. Reactogenicity was higher in the group that also received 4CMenB, but no concerning safety signals were identified. A robust antibody response was seen following 4CMenB administration, which met the criteria for sufficiency against three serogroup B test strains after both the primary and booster doses.

Although GMTs for the immune response to MenC were slightly higher when MenC-CRM was administered alone, which could potentially predict a longer persistence of individual protection, all the infants in both groups achieved protective hSBA titres $\geq 1:4$ (the currently accepted surrogate of protection for MenC disease) after the booster dose [18].

The percentage of infants with hSBA titres $\geq 1:4$ after 4CMenB primary series and booster was similar to previous studies in infants, where the vaccine was administered alone or concomitantly with routine infant vaccines [20–24]. For instance, in a study evaluating the immunogenicity of 4CMenB co-administered with routine vaccines according to a 3 + 1 dose schedule in infants (at 2, 4, 6 and 12 months of age), hSBA titres $\geq 1:4$ against fHBP, NadA and PorA strains were recorded in $\geq 64\%$ of infants after two doses of 4CMenB and $\geq 78\%$ of infants after booster vaccination [20].

No concerning safety signals were identified in the current study for either vaccine, although, as expected, reactogenicity was higher in the group that received concomitantly 4CMenB and MenC-CRM compared to MenC-CRM alone. Previous studies have also noted high levels of reactogenicity following receipt of 4CMenB in this age group, especially fever. In a study assessing the reactogenicity of 4CMenB administered to infants at 2, 3, 4, and 12 months of age together with routine paediatric vaccines, the occurrence of fever $\geq 38.5^\circ\text{C}$ after primary vaccination was 70.3%, compared with 27.1% in a group receiving the MenC-CRM vaccine [25]. In a mass vaccination setting, lower fever rates were reported in infants than previously observed in clinical trials (10.9% after a first dose of 4CMenB), although fever was still highest in this age group [26]. Despite the high percentage of infants with fever following 4CMenB vaccination in the current study, only one infant had a rectal temperature $\geq 40^\circ\text{C}$, which resolved within 2 days.

Although this was only a small study, as part of a larger clinical trial, the results have paramount importance for public health vaccination policies in countries such as Brazil, where a significant burden of disease is caused by these two major disease-causing serogroups in infants and young children. In several countries (like Italy, United Kingdom and Ireland), 4CMenB is already recommended or used as part of the infant immunisation schedule along MenC. The results of this study showed that concomitant administration of 4CMenB and MenC-CRM vaccines is possible, and this could lead to lowering of the costs of vaccine administration and is likely increase compliance to vaccination schedules due to a limited number of visits. In this study, immune responses were only tested against MenB reference strains, thus limiting conclusions regarding breadth of meningococcal strain coverage. However, previous MATS analysis estimated that 4CMenB provided coverage against 81% (95% CI: 71–95) of the circulating MenB invasive strains in Brazil. Furthermore, 4CMenB antigens are present and expressed in other serogroups' invasive strains, suggesting that vaccination could also provide protection against some meningococcal non-B strains [27,28]. A higher than expected rate of protocol deviations was observed in Group 2, and this led to a relatively small size of the post-booster per-protocol cohort. However, immunogenicity analyses were not impacted by this.

Table 3
Number (percentage) of infants with solicited local adverse reactions up to Day 7 following each vaccination.

	First vaccination			Second vaccination			Booster vaccination		
	Group 1		Group 2	Group 1		Group 2	Group 1		Group 2
	4CMenB N = 123	MenC-CRM N = 123	MenC-CRM N = 117	4CMenB N = 120	MenC-CRM N = 120	MenC-CRM N = 114	4CMenB N = 118	MenC-CRM N = 117	MenC-CRM N = 110
Tenderness	78 (63%)	72 (59%)	54 (46%)	69 (58%)	63 (53%)	39 (34%)	63 (53%)	59 (50%)	38 (35%)
Severe ^a	13 (11%)	10 (8%)	3 (3%)	10 (8%)	10 (8%)	2 (2%)	6 (5%)	3 (3%)	1 (1%)
Erythema	34 (28%)	14 (11%)	16 (14%)	33 (28%)	10 (8%)	16 (14%)	14 (12%)	13 (11%)	8 (7%)
Severe ^a	1 (1%)	0	0	0	0	0	0	0	0
Induration	14 (11%)	4 (3%)	16 (14%)	22 (18%)	4 (3%)	13 (11%)	12 (10%)	7 (6%)	6 (5%)
Severe ^a	0	0	0	0	0	0	0	0	0
Swelling	7 (6%)	4 (3%)	15 (13%)	14 (12%)	5 (4%)	10 (9%)	7 (6%)	7 (6%)	6 (5%)
Severe ^a	0	0	0	0	0	0	0	0	0

N, number of infants in each group for whom analyses were carried out.

^a Severe symptoms were defined as surface diameter >50 mm (erythema, induration and swelling), or 'cried when vaccinated limb was moved' (tenderness).

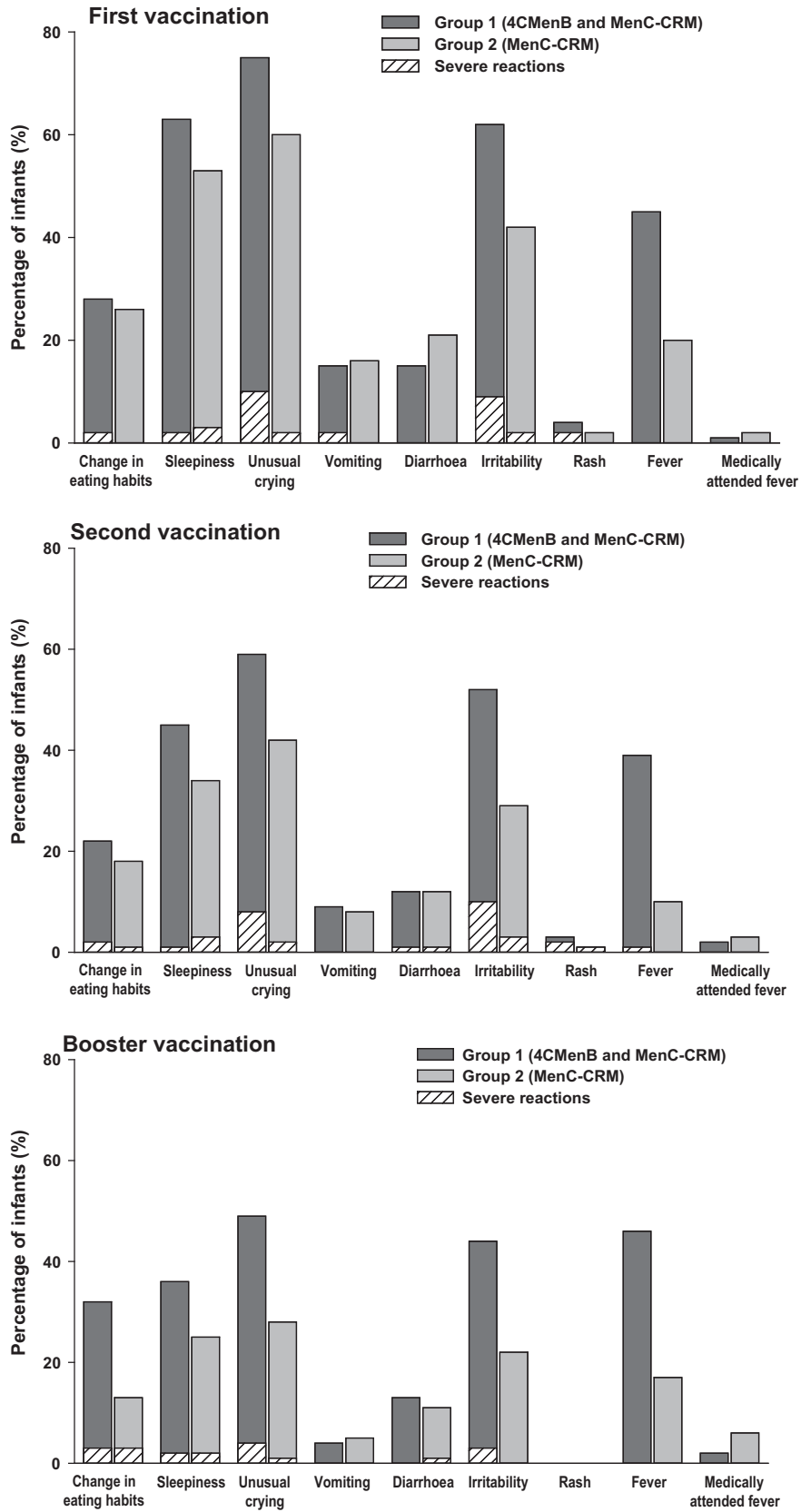


Fig. 3. Percentage of infants reporting systemic adverse reactions after each vaccination. Severe reactions are given in brackets.

In conclusion, concomitant administration of 4CMenB vaccine with MenC-CRM in infants induced robust immune responses against MenB reference strains, without clinically relevant interference with immune responses to MenC. Although increased reactivity was observed when 4CMenB was co-administered with MenC-CRM compared to MenC-CRM alone, with higher rates of fever, the safety profile of the vaccines was not affected. The concomitant administration of the 4CMenB vaccine with MenC-CRM in infants might be an important strategy for the introduction of 4CMenB vaccine in countries that already have MenC vaccines in their routine immunisation schedule, by minimising the number of visits required to deliver each vaccine individually and potentially providing protection against the two predominant IMD-causing serogroups in this age group.

Author contributions

DT designed the study. MAS, LYW and EDMJ conducted the study and collected the data. EJFL participated in the collection/generation of the data and contributed materials/analysis/reagent tools. IM performed the statistical analysis. IM, MC and DT analysed and interpreted the data. All authors reviewed the manuscript and approved the final version for submission.

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Trademark statement

Bexsero, *Menjugate* and *Synflorix* are trademarks of the GSK group of companies.

Conflict of interest

MC was and, DT and IM are employees of the GSK group of companies. DT owns stock/stock options in the GSK group of companies. MAPS reported a grant from the GSK group of companies during the conduct of the study. MAPS received research grant from the GSK group of companies, Pfizer and Takeda and speaker's honoraria from the GSK group of companies, Pfizer and Sanofi-Pasteur outside the submitted work. The institution of LYW received a grant from Novartis during the conduct of this study and from the GSK group of companies outside the submitted work. LYW reported personal fees as member of advisory board for the GSK group of companies, Novartis and MSD outside the submitted work. The institution of FMT received clinical trial fees from Novartis during the conduct of this study, and he received personal fees/non-financial support/grants/other from Pfizer, SPMSD and/or GSK, outside the submitted work. EDM reported a grant from Novartis during the conduct of the study. EJFL declares a grant from Novartis during the conduct of the study.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at <http://dx.doi.org/10.1016/j.vaccine.2017.03.002>.

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