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Serologic Responses to ACYW135 Polysaccharide Meningococcal Vaccine in Saudi Children under 5 Years of Age

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An immunization campaign with meningococcal ACYW135 polysaccharide vaccine was conducted in 2003 by the Saudi Arabian Ministry of Health and included a study to evaluate the immune responses in children under 5 years of age in the Al Qassim region of Saudi Arabia. Children who were ≥24 months old were given one dose of tetravalent polysaccharide vaccine, while younger children were given two doses with an interval of 2 to 3 months. Blood samples were collected prevaccination and 1 month after the second dose for children younger than 24 months old and 1 month after the single dose for older children. Serogroup-specific antibody responses were determined by serum bactericidal antibody (SBA) assays and a tetraplex immunoglobulin G (IgG) bead assay. Significant increases in the proportions of individuals who were ≥24 months old with SBA titers of ≥8 were observed pre- to postvaccination for all serogroups. Age-dependent increases in the percentage of individuals with SBA titers of ≥8 1 month postvaccination were observed for each serogroup. Age-dependent increases in postvaccination IgG levels were observed for serogroup A (menA), serogroup W135 (menW), and serogroup Y (menY) but not for serogroup C (menC). Two doses of tetravalent polysaccharide vaccine in individuals who were ≤18 months old were poorly immunogenic for menC, menW, and menY. However, for menA, 42% of the children who were 18 months old were putatively protected with SBA titers of ≥8. A high percentage of subjects who were ≥2 years of age were putatively protected for menA; a similar level was observed for menY for children who were 4 years of age but not for younger children. However, for menC and menW poor levels of putative protection were still evident at 4 years of age.

Bacterial meningitis is a significant health problem worldwide, particularly in young children. Epidemics of serogroup A meningococcal disease occur in the meningitis belt in sub-Saharan Africa, whereas endemic serogroup B, C, and Y disease occurs in most other regions of the world. Plain polysaccharide vaccines containing either serogroups A and C or serogroups A, C, Y, and W135 have been available for several years, but the serogroup C portion has limited immunogenicity in young children (13). Meningococcal serogroup C conjugate vaccines have been introduced in the United Kingdom (and other countries) and have successfully reduced the incidence of serogroup C disease (8).

Meningococcal disease remains a serious public health issue in Saudi Arabia and occurs in the form of epidemics, usually coinciding with the Hajj and Umra seasons, when 4 million pilgrims visit each year. In 1987, following meningococcal epidemics caused by menA, the Saudi Arabian authorities issued a requirement that all persons applying for Hajj or Umra should be vaccinated with the bivalent A/C vaccine (7). Children under 2 years of age were given monovalent serogroup A vaccine. During the years 2000 to 2002 there was a shift in the epidemic pattern of meningococcal disease during the Hajj season, and there was a predominance of *Neisseria meningitis* menW (15, 25). Globally, an increase in menW disease occurred during the same period. Epidemics of menW disease occurred in Burkina Faso (4, 5), and significant numbers of Hajj-related cases were observed globally due to returning pilgrims (28). Characterization of the menW strains isolated both from Burkina Faso and countries experiencing Hajj-related menW disease demonstrated that they were mainly the same strain, W135:2a:P1.5,2 belonging to the ET-37 clonal complex (5, 24, 29). It is likely that the outbreak in 2000 and the global increase in menW disease were due to expansion of the ET-37 clone that has been in circulation since at least 1970 (24). New genetic lineages of menW that are independent of the Hajj-related strain have been reported (30).

In 2002 the Saudi Ministry of Health decided to use the ACYW135 polysaccharide vaccine to cover menW and menY (6). The vaccine was recommended for those coming for Hajj and for school children in Saudi Arabia. However, it was observed that 58% of reported meningococcal disease occurred in children below the age of 5 years and that 39% occurred in children below the age of 2 years (1). Therefore, in preparation for the Hajj season the Ministry of Health decided to launch a campaign with ACYW135 polysaccharide vaccine, targeting children who were 6 months to 5 years old. The campaign was conducted in 2003 and included a study to evaluate the immune responses to ACYW135. Serum bactericidal antibody (SBA) titers following two doses of tetravalent polysaccharide

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TABLE 1. Compliance with sampling and numbers of paired sera sent for serology by age group

Age group (mo)	No. of children				
	Planned	Agreed to participate	First visit	Second visit	Paired sera sent for serology
6	100	70	61	44	44
12	100	76	63	46	46
18	100	80	67	49	49
24	100	92	79	68	68
36	100	95	81	78	78
48	100	100	98	93	93

vaccine in children under 2 years of age have not been previously reported.

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MATERIALS AND METHODS

Overall design. A follow-up study to evaluate the immunogenicity to tetravalent polysaccharide ACYW135 vaccine given to children up to 5 years old during the campaign was performed. The following six age groups were included in the study: 6, 12, 18, 24, 36, and 48 months. The age range was ±2 weeks for each age group. Children under 24 months of age were vaccinated with two doses with an interval of 2 to 3 months, while older children were vaccinated with one dose. After guardian consent was obtained, blood samples were collected before the first dose and 1 month after the second for children under 2 years old and before the single dose of vaccine and 1 month later for children 2 years old and older. Ethical approval for the study was obtained from the Ethical Committee, Saudi Ministry of Health.

Study population. The study was conducted in the Al Qassim region in the central part of Saudi Arabia, 350 km north of Riyadh and 1,000 km northeast of Mecca. The study was conducted through Primary Health Care (PHC) centers. Five centers were selected randomly from the list of PHC centers in Qassim. Children were recruited randomly from the catchment area of each center, with a planned 100 children in each age group to allow for the expected dropout. The inclusion criteria were as follows: Saudi nationals who were 6 months to 5 years old, with ±2 months allowed for each target age group, and agreement to participation by the guardian. The exclusion criteria included a history of chronic illness, a history of prior meningococcal vaccination, and a prior history of any meningococcal disease or meningitis of other etiology.

Vaccine. The vaccine used in the national campaign was Mencevax ACWY (GSK, Rixensart, Belgium). This vaccine contained 50 μg of each of the four polysaccharides. The vaccine used is licensed for use in children who are more than 2 years old. The study included infants for whom the vaccine is not licensed.

Serology. Serogroup-specific antibody responses were determined by SBA assays as described elsewhere (23) using baby rabbit complement (Pel-Freeze Incorporated, Rodgerson, AZ) as an exogenous source of complement. The following strains were used: for menA, F8238 (A:4:P1.20,9); for menC, C11 (C:16:P1.7-1,1); for menW, M,01.0240070 (W135:NT:P1.18-1,3); and for menY, M,00.0242975 (Y:2a:P1.5,2). SBA titers were expressed as the reciprocal of the final serum dilution giving ≥50% killing at 60 min for menC, menW, and menY and at 90 min for menA. For computational purposes, titers of <4 were assigned a value of 2. A subject with an SBA titer of ≥8 was defined as putatively protected (3).

Serogroup-specific immunoglobulin G (IgG) concentrations were determined as previously described using a tetraplex IgG bead assay (19). The calibration factors of the standard CDC1992 serum for menC-specific IgG used were 91.8 and 24.1 μg/ml for menA and menC, respectively (16), and 25.23 and 28.92 μg/ml for menW and menY, respectively (18). Results were expressed in μg/ml, and the lower limits of detection for menA, menC, menW, and menY were 0.08, 0.06, 0.065, and 0.075 μg/ml, respectively.

Statistical analysis. Serogroup-specific IgG concentrations (GMCs) with 95% confidence intervals were calculated for each group at each sampling time. The proportions with IgG titers of >1 and 2 μg/ml were calculated for each age

TABLE 2. Proportions of individuals for serogroups A, C, Y, and W135 by age group with serum bactericidal antibody titers of ≥8 before and 1 month following vaccination with tetravalent polysaccharide vaccine and ≥fourfold increases pre- to postvaccination

Serogroup	Age group (mo)	No. of individuals/no. of individuals tested (%)			McNemar's exact *P* value for change from pre- to postvaccination
		SBA titer of ≥ 8 prevaccination	SBA titer of ≥ 8 1 mo postvaccination	≥Fourfold increase pre- to postvaccination	
A	6	3/37 (8)	7/37 (19)	6/30 (20)	0.13
	12	10/39 (26)	12/42 (29)	6/35 (17)	0.63
	18	12/43 (28)	19/45 (42)	11/39 (28)	0.15
	24	29/65 (45)	41/51 (80)	23/49 (47)	<0.001
	36	34/75 (45)	56/64 (88)	37/62 (60)	<0.001
	48	47/86 (55)	67/71 (94)	47/68 (69)	<0.001
C	6	3/41 (7)	5/43 (12)	5/40 (13)	0.73
	12	2/42 (5)	5/44 (11)	4/40 (10)	0.38
	18	1/43 (2)	5/47 (11)	4/41 (10)	0.13
	24	5/68 (7)	25/59 (42)	22/59 (37)	<0.001
	36	4/79 (5)	29/71 (41)	28/71 (38)	<0.001
	48	8/90 (9)	40/84 (48)	36/81 (44)	<0.001
W135	6	0/43 (0)	5/43 (12)	5/42 (12)	0.063
	12	1/45 (2)	6/46 (13)	5/45 (11)	0.22
	18	2/49 (4)	9/49 (18)	8/49 (16)	0.04
	24	4/68 (6)	32/68 (47)	32/68 (47)	<0.001
	36	4/78 (5)	42/76 (55)	39/76 (51)	<0.001
	48	11/92 (12)	55/89 (62)	50/88 (57)	<0.001
Y	6	2/40 (5)	6/41 (15)	5/37 (14)	0.063
	12	5/44 (11)	6/43 (14)	5/41 (12)	1.00
	18	2/45 (4)	9/47 (19)	9/43 (21)	0.07
	24	14/67 (21)	32/60 (53)	26/59 (44)	<0.001
	36	28/77 (37)	46/71 (65)	34/69 (49)	<0.001
	48	35/92 (38)	74/85 (87)	63/84 (75)	<0.001

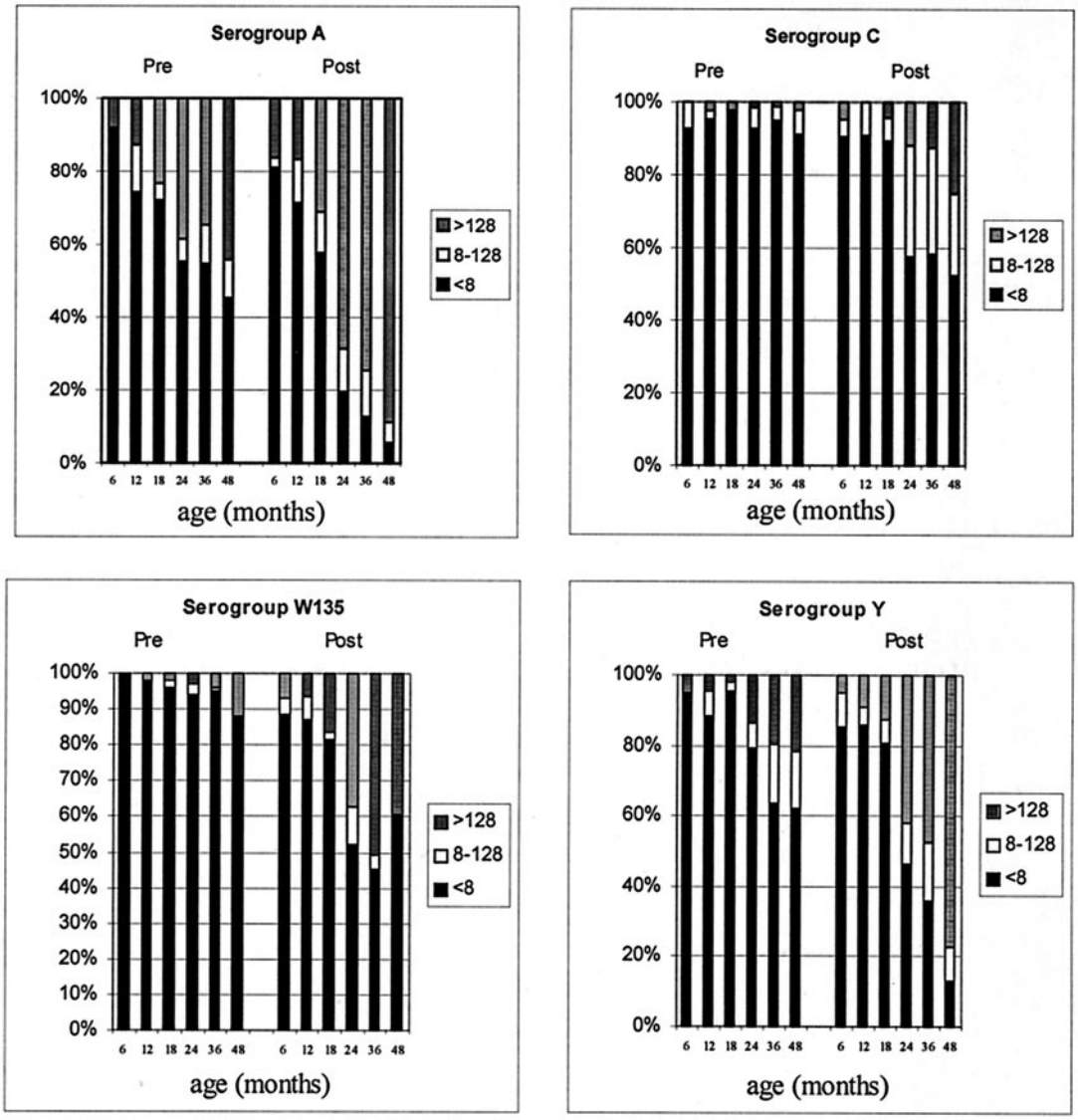

FIG. 1. Percentages of subjects before and 1 month following vaccination at serum bactericidal titer cutoffs of <8, 8 to 128, and >128.

group, and values for different ages were compared using a chi-square test or Fisher's exact test as appropriate and tested for trend by age. The proportions of individuals with SBA titers of ≥8 were calculated and compared between age groups using a chi-squared test and test for trend by age. The significance of changes in the proportions of individuals with SBA titers of ≥8 pre- to postvaccination was calculated using McNemar's exact test. Paired *t* tests were used to compare IgG titers pre- to postvaccination.

RESULTS

Sample compliance. Table 1 summarizes the distribution of the recruited children by age group. A high dropout level was noticed in younger age groups, mainly due to refusal to give blood samples, especially in the second visits. Due to the large number of assays required and the limited serum volumes, some samples were not sufficient to test for all serogroups; therefore, the numbers of results for each age group vary by serogroup.

Serum bactericidal antibody titers. The SBA titers were not normally distributed because a large proportion of the values were <4. Therefore, the SBA data were analyzed by examining the proportions of individuals with SBA titers with ≥eight- or fourfold increases from pre- to postvaccination (Table 2). The percentages of individuals at different SBA cutoffs are shown in Fig. 1.

Significant increases ($P < 0.001$) in the proportions of individuals who were ≥24 months old with SBA titers of ≥8 were observed pre- to postvaccination for all serogroups. If the data for 6- to 18-month-old children were combined, increases in the numbers of individuals with SBA titers of ≥8 were observed pre- to postvaccination (for menA, $P = 0.01$; for menC, $P = 0.03$; for menW, $P < 0.001$; for menY, $P = 0.01$).

Age-dependent increases in both prevaccination and postvaccination SBA titers were suggested by the percentages of individuals with different SBA titers shown in Fig. 1 and were

TABLE 3. Serogroup-specific IgG concentrations for serogroups A, C, Y, and W135 by age group before and 1 month following vaccination with tetravalent polysaccharide vaccine

Serogroup	Age group (mo)	Prevaccination		1 mo postvaccination	
		No.	GMC (μg/ml) (95% confidence interval)	No.	GMC (μg/ml) (95% confidence interval)
A	6	43	2.4 (1.8–3.1)	44	4.1 (3.1–5.5)
	12	46	1.9 (1.4–2.5)	44	7.5 (5.2–11.0)
	18	49	2.9 (2.4–3.5)	49	9.5 (6.7–13.5)
	24	68	1.6 (1.2–2.1)	67	7.3 (4.9–10.8)
	36	78	2.6 (1.9–3.4)	77	10.9 (7.7–15.3)
	48	93	2.6 (2.1–3.2)	92	10.4 (7.8–13.8)
C	6	43	1.9 (1.4–2.6)	44	2.8 (2.3–3.4)
	12	46	1.9 (1.4–2.5)	46	3.4 (2.7–4.3)
	18	49	2.3 (1.8–2.9)	49	3.4 (2.7–4.3)
	24	68	1.7 (1.4–2.0)	67	3.0 (2.5–3.8)
	36	78	1.9 (1.6–2.3)	77	2.4 (1.8–3.2)
	48	92	2.0 (1.7–2.4)	91	3.4 (2.7–4.2)
W135	6	43	1.0 (0.7–1.3)	44	1.3 (1.0–1.6)
	12	46	0.9 (0.7–1.2)	46	1.7 (1.4–2.3)
	18	49	0.9 (0.7–1.1)	49	1.7 (1.4–2.1)
	24	68	0.7 (0.6–0.9)	68	1.6 (1.3–2.1)
	36	78	0.9 (0.8–1.1)	77	1.8 (1.3–2.3)
	48	93	0.9 (0.8–1.2)	93	2.2 (1.6–2.9)
Y	6	43	0.4 (0.2–0.5)	43	0.6 (0.5–0.8)
	12	46	0.3 (0.2–0.5)	46	0.7 (0.5–0.9)
	18	49	0.4 (0.3–0.5)	49	0.9 (0.7–1.1)
	24	68	0.3 (0.2–0.3)	66	1.1 (0.8–1.6)
	36	78	0.4 (0.3–0.4)	77	1.1 (0.8–1.6)
	48	93	0.4 (0.3–0.5)	92	2.0 (1.4–2.9)

confirmed by analysis of the proportions of individuals with SBA titers of ≥8 both pre- and postvaccination for all serogroups ($P \leq 0.005$) with the exception of prevaccination menC ($P = 0.42$). For all serogroups there was an increase in the percentage of individuals with a ≥fourfold increase in the SBA titer pre- to postvaccination with increasing age ($P < 0.001$ for all serogroups).

Meningococcal serogroup-specific IgG. The serogroup-specific IgG GMCs and 95% confidence intervals are shown in Table 3. The levels of menA-specific IgG were higher than the levels for the other serogroups. Significant increases ($P < 0.05$) were seen from pre- to postvaccination in all age groups for each serogroup except menW-specific IgG at 6 months.

Analysis of serogroup-specific IgG concentrations by age showed that, unlike the SBA titers, there were no age-dependent increases in prevaccination concentrations for any serogroup. Postvaccination there were clear increases with age in GMCs for menA and menY ($P \leq 0.001$) and a small increase for menW ($P = 0.02$). There was no increase with age for menC ($P = 0.96$).

The proportions of individuals with serogroup-specific IgG levels greater than a range of concentrations (0.25 to 500 μg/ml) pre- and postvaccination are shown as reverse cumulative distributions in Fig. 2. For menA and menC there were similar proportions of individuals with IgG concentrations of >1 or 2 μg/ml both pre- and postvaccination, which is reflected in the similar distribution frequencies shown in Fig. 2A and B. For menW and menY there were lower proportions of individuals with IgG concentrations of >1 or 2 μg/ml than there were for menA or menC. For menY, only 61% and 45% of the individuals who were 48 months old achieved concentrations of >1 and 2 μg/ml following vaccination, respectively, compared to 97% and 89% for menA.

An age-dependent increase in the proportions of individuals achieving a concentration of >1 μg/ml was observed only for menY postvaccination ($P < 0.001$), confirming the age-dependent response illustrated in the reverse cumulative distribution plot (Fig. 2D).

DISCUSSION

In 6-, 12-, and 18-month-old children, two doses of tetravalent polysaccharide vaccine gave poor responses against menC, menW, and menY; however, for menA, two doses resulted in 42% putatively protected children (SBA titer, ≥8) at 18 months of age. A high percentage of 2-year-old children were putatively protected following a single dose of menA polysaccharide, and a similar percentage was observed for menY for the 4-year-old group but not for younger groups. However, for menC and menW poor responses were still evident at 4 years of age. The SBA responses observed for menC, menW, and menY, although poor compared to the responses observed for menA, do appear to be age dependent, and an increase in the proportions of children with SBA titers of ≥8 was seen in individuals who were 24 months old or older. An SBA titer of ≥8 determined by an assay using baby rabbit complement has been proposed as a short-term putative correlate of protection following menC conjugate vaccination (3, 9). For menA the only proposed correlate of protection is from the Finnish efficacy trials of menA polysaccharide vaccine and is 2 μg/ml of

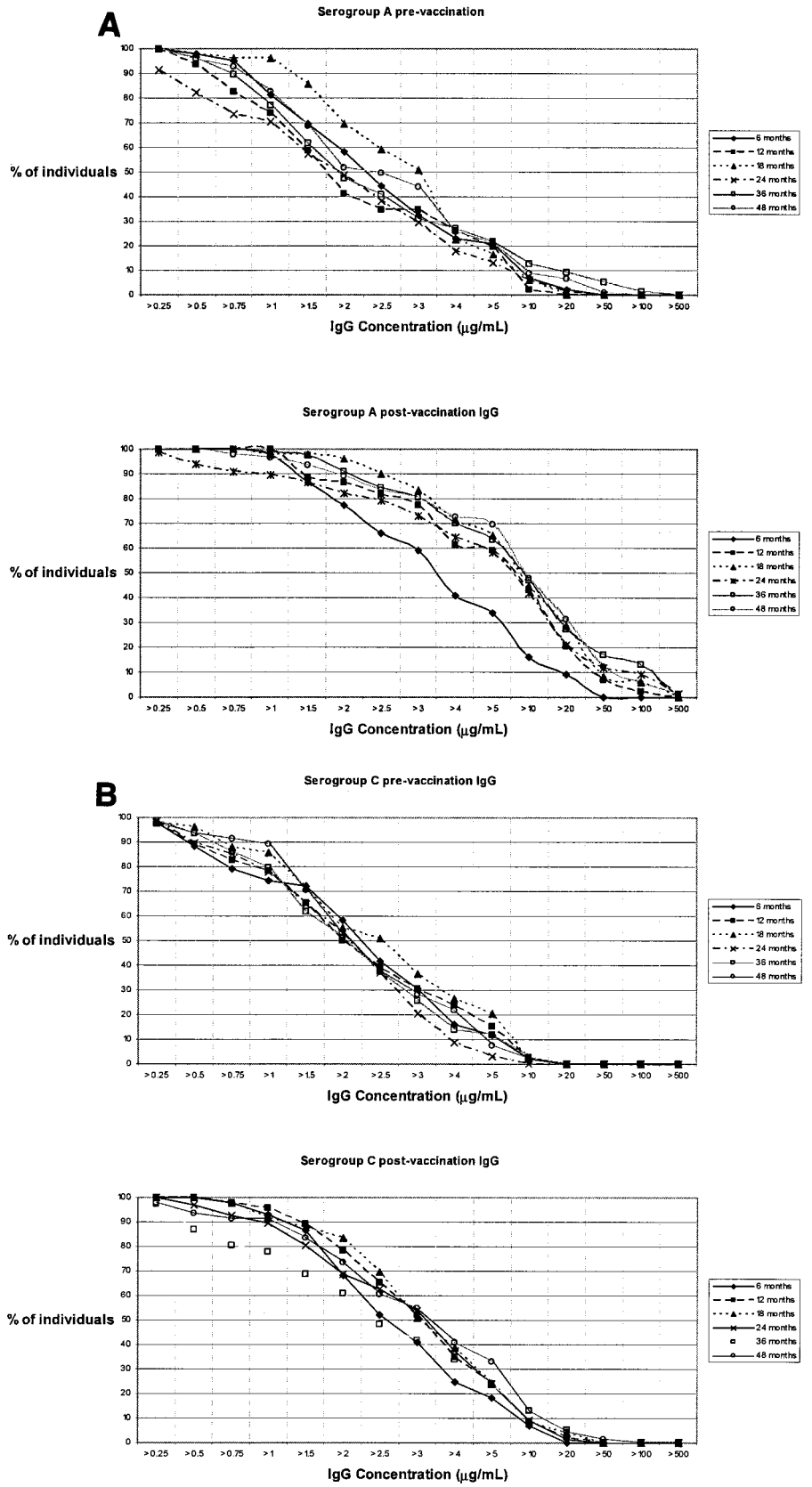

FIG. 2. Serogroup-specific IgG concentrations for serogroups A (A), C (B), W135 (C), and Y (D) by age group, before and 1 month following vaccination with tetravalent polysaccharide vaccine.

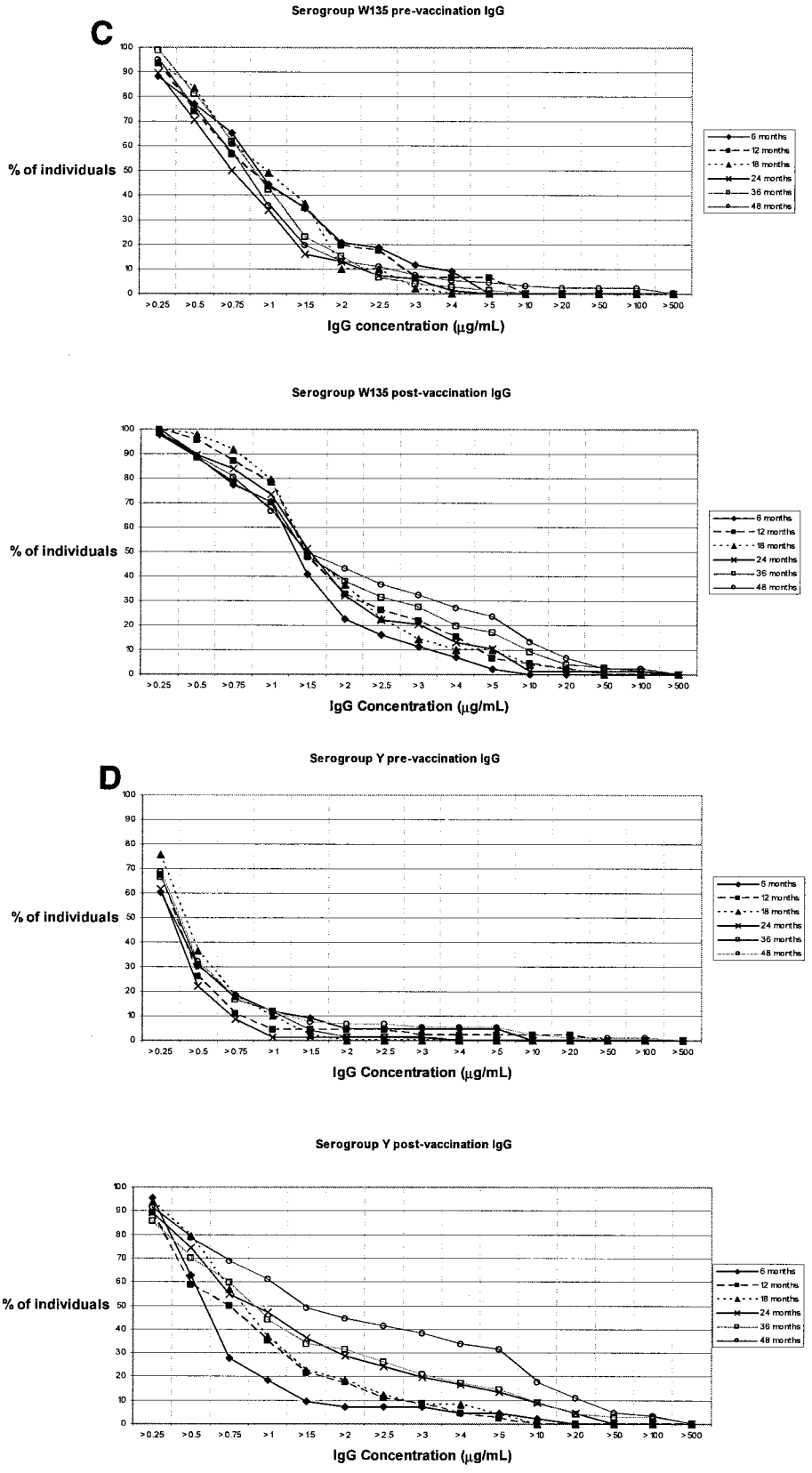

FIG. 2—*Continued*.

immunoglobulin as determined by radioimmunoassay (22, 26). There are currently no putative correlates of protection for menW or menY.

Prior to vaccination, an age-dependent increase in the proportion of children with SBA titers of ≥8 was observed for menA, menW, and menY but not for menC. There are no previous studies to compare the data for menW and menY; however, the menC data are similar to data for age-matched United Kingdom children, in which no increase in the proportion with SBA titers of ≥8 was observed (32). However, the setting of the study does appear to have an influence, as in age-matched United States children a general increase in the percentage with menC SBA titers of ≥4 (using human complement) was observed (14). An age-dependent increase in the percentage with SBA titers of ≥4 was also observed in United States children for menA (14), which is similar to the data in this study for Saudi children.

A Finnish study (27) that looked at the immunogenicity of one dose of tetravalent polysaccharide vaccine revealed over 90% responders for menA (≥fourfold increase in the SBA titer or from undetectable to detectable SBA titer) in children 6 to 23 months of age. For menC, menW, and menY in the 18- to 23-month age group there were 90, 85, and 79% responders, respectively. These data are not directly comparable with the data reported here as only one dose of vaccine was used in the Finnish study and two doses were used for the <24-month-old children in the present study. It is well documented that repeated doses of menC polysaccharide lead to hyporesponsiveness (13, 21); however, to date, there have been no reported studies for menW and menY. This could not be categorically ascertained in the present study, as blood was not collected following the first dose for children who were <24 months of age. However, the proportions of individuals with SBA titers of ≥8 postvaccination for children who were 6 to 18 months old were similar for menC, menW, or menY, and the values for children who were ≥24 months old were significantly greater, suggesting that menW and menY polysaccharides behave like the menC polysaccharide rather than the menA polysaccharide, for which a steady increase in the proportion of individuals with SBA titers of ≥8 was observed with increasing age. Repeated doses of menA polysaccharide lead to boosting in adults (17), but it is still unclear if this occurs in young children or if, similar to the response to repeated doses of menC polysaccharide, hyporesponsiveness is induced. An inherent problem when ≥fourfold increases in the SBA titer prevaccination to postvaccination are used is the presence of SBA titers in the prevaccination samples. This biases the number of responders; for example, 40% of 24-month-old children have greater SBA titers (≥128) prevaccination. The percentage of individuals seroconverting for menC in the Finnish study (90% for the 18- to 23-month-old children) also seems very high, especially as menC polysaccharide has not been reported to protect children who are <2 years of age (2, 31).

We found that the immunogenicity of the serogroups differed as follows, from the most immunogenic to the least immunogenic: menA, menY, menW, menC. In contrast, Lepow et al. (20) found that for children less than 5 years of age receiving one dose of a tetravalent polysaccharide, a greater percentage responded to menW and menY than to menA or menC. However, a similar number of responders to menC (40% of 2- to 5-year-old children) was observed, which is similar to our findings. This is also supported by the vaccine effectiveness reported following a mass immunization campaign against menC disease in Quebec, where the effectiveness was 41% for ages 2 to 9 years (12). Our data for menC are also similar to those reported in a previous study of bivalent polysaccharide in young children in the United Kingdom (10). A small French study performed with older children (ages, 3 to 13 years) and with a single dose of tetravalent polysaccharide demonstrated that all 21 children responded, as measured by a ≥fourfold increase in the SBA titer pre- to post vaccination for all four serogroups (11).

Age-dependent increases in serogroup-specific IgG concentrations were observed following vaccination for menA, menW, and menY. The concentrations observed for menA were greater than the concentrations observed for the other serogroups. There is no known IgG correlate of either short- or long-term protection following polysaccharide vaccination. For menA, 2 μg/ml of anticapsular total antibody has been reported to be a correlate of protection following polysaccharide vaccination (22, 26). In this study ≥77% of individuals achieved an IgG concentration of >2 μg/ml in response to the menA portion of the polysaccharide vaccine. Low levels of menY-specific IgG were observed in comparison to the other serogroups. The response to the menY portion of the polysaccharide vaccine was shown to be age dependent, but only 61% and 45% of the children who were 48 months old had IgG concentrations of >1 and 2 μg/ml, respectively. However, 87% of the children in the same age group had menY SBA titers of ≥8, and 75% had a ≥fourfold increase in the SBA titer pre- to postvaccination.

In conclusion, two doses of tetravalent polysaccharide vaccine in children <24 months old gave poor protection against menC, menY, and menW; however, there was reasonable protection against menA from 18 months of age. For one dose from 24 months to 48 months there was a trend to an age-dependent response for all serogroups. These data led to discontinuation of the use of tetravalent vaccine in children less than 2 years of age in Saudi Arabia and have wider implications for the use of tetravalent polysaccharide vaccines in African countries.

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REFERENCES

1. **Al-Mazrou, Y. Y., E. K. Musa, M. N. Abdalla, M. H. Al-Jeffri, S. H. Al-Hajjar, and O. M. Mohamed.** 2003. Disease burden and case management of bacterial meningitis among children under 5 years of age in Saudi Arabia. Saudi Med. J. **24:**1300–1307.
2. **Amato, N. V., H. Finger, E. C. Gotschlich, R. A. Feldman, C. A. de Avila, and S. R. Konichi.** 1974. Serological response to serogroups C meningococcal vaccine in Brazilian preschool children. Rev. Inst. Med. Trop. Sao Paulo **16:**149–153.
3. **Andrews, N., R. Borrow, and E. Miller.** 2003. Validation of serological correlate of protection for meningococcal C conjugate vaccine using efficacy estimates from post-licensure surveillance in England. Clin. Diagn. Lab. Immunol. **10:**780–786.
4. **Anonymous.** 2001. Epidemics of meningococcal disease, African meningitis belt, 2001. Wkly. Epidemiol. Rec. **76:**281–288.

5. **Anonymous.** 2002. Meningococcal disease, serogroup W135, Burkina Faso. Wkly. Epidemiol. Rec. **77:**141–156.
6. **Anonymous.** 2002. Meningococcal vaccines: polysaccharide and polysaccharide conjugate vaccines. Wkly. Epidemiol. Rec. **77:**331–339.
7. **Anonymous.** 1992. Pilgrimage to Mecca (Hajj) 1992. Commun. Dis. Rep. Wkly. **2:**43.
8. **Balmer, P., R. Borrow, and E. Miller.** 2002. Impact of meningococcal C conjugate vaccine in the UK. J. Med. Microbiol. **51:**717–722.
9. **Borrow, R., N. Andrews, D. Goldblatt, and E. Miller.** 2001. Serological basis for use of meningococcal serogroup C conjugate vaccines in the United Kingdom: reevaluation of correlates of protection. Infect. Immun. **69:**1568–1573.
10. **Borrow, R., P. Richmond, E. B. Kaczmarski, A. Iverson, S. L. Martin, J. Findlow, M. Acuna, E. Longworth, R. O'Connor, J. Paul, and E. Miller.** 2000. Meningococcal serogroup C-specific IgG antibody responses and serum bactericidal titers in children following vaccination with a meningococcal A/C polysaccharide vaccine. FEMS Immunol. Med. Microbiol. **28:**79–85.
11. **Cadoz, M., J. Armand, F. Arminjon, R. Gire, and C. H. Lafaix.** 1985. Tetravalent (A,C,Y,W135) meningococcal vaccine in children: immunogenicity and safety. Vaccine **3:**340–342.
12. **De Wals, P., G. De Serres, and T. Niyonsenga.** 2001. Effectiveness of a mass immunization campaign against serogroup C meningococcal disease in Quebec. JAMA **285:**177–181.
13. **Gold, R., M. L. Lepow, I. Goldschneider, T. L. Draper, and E. C. Gotschlich.** 1975. Clinical evaluation of group A and group C meningococcal polysaccharide vaccines in infants. J. Clin. Investig. **56:**1536–1547.
14. **Goldschneider, I., E. C. Gotschlich, and M. S. Artenstein.** 1969. Human immunity to the meningococcus. I. The role of humoral antibodies. J. Exp. Med. **129:**1307–1326.
15. **Hajjeh, R. A., and J. Lingappa.** 2000. Meningococcal disease in the Kingdom of Saudi Arabia: an evaluation of disease surveillance and control after an outbreak of serogroup W-135 meningococcal disease. CDC Rep. **23:**2000.
16. **Holder, P. K., S. E. Maslanka, L. B. Pais, J. Dykes, B. D. Plikaytis, and G. M. Carlone.** 1995. Assignment of *Neisseria meningitidis* serogroup A and C class-specific anticapsular antibody concentrations to the new standard reference. Clin. Diagn. Lab. Immunol. **2:**132–137.
17. **Jokhdar, H., R. Borrow, A. Sultan, M. Adi, C. Riley, E. Fuller, and D. Baxter.** 2004. Immunologic hyporesponsiveness to serogroup C but not serogroup A following repeated meningococcal A/C polysaccharide vaccination in Saudi Arabia. Clin. Diagn. Lab. Immunol. **11:**83–88.
18. **Joseph, H., P. Balmer, M. Bybel, T. Papa, R. Ryall, and R. Borrow.** 2004. Assignment of *Neisseria meningitidis* serogroup A, C, W135 and Y anticapsular total IgG, IgG1 and IgG2 concentrations to reference sera. Clin. Diagn. Lab. Immunol. **11:**1–5.
19. **Lal, G., P. Balmer, H. Joseph, M. Dawson, and R. Borrow.** 2004. Development and evaluation of a multiplex flow cytometric assay for quantitation of serum antibodies to *Neisseria meningitidis* serogroups A, C, Y and W-135. Clin. Diagn. Lab. Immunol. **11:**272–279.
20. **Lepow, M. L., J. Beeler, M. Randolph, J. S. Samuelson, and W. A. Hankins.** 1986. Reactogenicity and immunogenicity of a quadrivalent combined meningococcal polysaccharide vaccine in children. J. Infect. Dis. **154:**1033–1036.
21. **MacDonald, N. E., S. A. Halperin, B. J. Law, B. Forrest, L. E. Danzig, and D. M. Granoff.** 1998. Induction of immunologic memory by conjugated vs plain meningococcal C polysaccharide vaccine in toddlers: a randomized controlled trial. JAMA **280:**1685–1689.
22. **Mäkelä, P. H., H. Käyhty, P. Weckström, E. Sivonen, and O.-V. Renkonen.** 1975. Effect of group A meningococcal vaccine in army recruits in Finland. Lancet **ii:**883–886.
23. **Maslanka, S. E., L. L. Gheesling, D. E. LiButti, K. B. J. Donaldson, H. S. Harakeh, J. K. Dykes, F. F. Arhin, S. J. Devi, C. E. Frasch, J. C. Huang, P. Kriz-Kuzemenska, R. D. Lemmon, M. Lorange, C. C. Peeters, S. Quataert, J. Y. Tai, and G. M. Carlone.** 1997. Standardization and a multilaboratory comparison of *Neisseria meningitidis* serogroup A and C serum bactericidal assays. Clin. Diagn. Lab. Immunol. **4:**156–167.
24. **Mayer, L. W., M. W. Reeves, N. Al-Hamdan, C. T. Sacchi, M. K. Taha, G. W. Ajello, S. E. Schmink, C. A. Noble, M. L. C. Tondella, A. M. Whitney, Y. Al-Mazrou, M. Al-Jeffri, A. Mishkhis, S. Sabban, D. A. Caugant, J. Lingappa, N. E. Rosenstein, and T. Popovic.** 2002. Outbreak of W135 meningococcal disease in 2000: not emergence of a new W135 strain but clonal expansion within the electrophoretic type-37 complex. J. Infect. Dis. **185:**1596–1605.
25. **Memish, Z. A.** 2002. Meningococcal disease and travel. Clin. Infect. Dis. **34:**84–90.
26. **Peltola, H., H. Mäkelä, H. Käyhty, H. Jousimies, E. Herva, K. Hallstrom, A Sivonen, O. V. Renkonen, O. Pettay, V. Karanko, P. Ahvonen, and S. Sarna.** 1997. Clinical efficacy of meningococcus group A capsular polysaccharide vaccine in children three months to five years of age. N. Engl. J. Med. **297:**686–691.
27. **Peltola, H., A. Safary, H. Kayhty, V. Karanko, and F. E. Andre.** 1985. Evaluation of two tetravalent (ACYW135) meningococcal vaccines in infants and small children: a clinical study comparing immunogenicity of O-acetyl-negative and O-acetyl-positive group C polysaccharides. Pediatrics **76:**91–96.
28. **Taha, M. K., M. Achtman, J. M. Alonso, B. Greenwood, M. Ramsay, A. Fox, S. Gray, and E. Kaczmarski.** 2000. Serogroup W135 meningococcal disease in Hajj pilgrims. Lancet **356:**2159.
29. **Taha, M. K., I. Parent du Chatelet, M. Schlumberger, I. Sanou, S. Djibo, F. de Chabalier, and J. M. Alonso.** 2002. *Neisseria meningitidis* serogroups W135 and A were equally prevalent among meningitis cases occurring at the end of the 2001 epidemics in Burkina Faso and Níger. J. Clin. Microbiol. **40:**1083–1084.
30. **Taha, M. K., D. Giorgini, M. Ducos-Galand, and J. M. Alonso.** 2004. Continuing diversification of *Neisseria meningitidis* W135 as a primary cause of meningococcal disease after emergence of the serogroup in 2000. J. Clin. Microbiol. **42:**4158–4163.
31. **Taunay, A. E., R. A. Feldman, C. O. Bastos, P. A. A. Galvao, J. S. Morais, and I. O. Castro.** 1978. Evaluation of the protective effect of meningococcal serogroup C vaccine in infants aged 6–36 months. Rev. Inst. Adolfo Lutz **38:**77–82.
32. **Trotter, C., R. Borrow, N. Andrews, and E. Miller.** 2003. Seroprevalence of meningococcal serogroup C bactericidal antibody in England and Wales in the pre-vaccination era. Vaccine **21:**1094–1098.

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