Pneumococcal infections cause at least one million deaths worldwide annually, mostly in young children (52). Immunization against Streptococcus pneumoniae has the potential to face this burden of disease. An ideal vaccine should rapidly elicit protective immunity and generate memory cells, which respond efficiently to subsequent antigen exposure. Generation of memory B cells and long lived plasma cells is associated with isotype switching and hypermutation of the immunoglobulin genes, resulting in selection of B cells with high-affinity B-cell receptors (27, 28). The established 23-valent pneumococcal polysaccharide vaccine (PPV-23) induces in adults primarily immunoglobulin M (IgM), with hardly any class switching, and at 7 and 28 days after administration of the PPV booster and compared to an opsonophagocytosis assay. Of group A, 64 to 100% had antibody concentrations of ≥1 μg/ml on day 28 after the booster versus 25 to 94% of group B. Group A had significantly higher antibody concentrations for all PCV-containing serotypes already on day 7, indicating early memory response. Antibody concentrations were in accordance with functional opsonic activity, although opsonic titers varied among individuals. Pneumococcal vaccination was well tolerated. The incidence of airway infections was reduced after priming with PCV (10/year for group A versus 15/year for group B). Following a PPV booster, even patients primarily not responding to PPV showed a rapid and more pronounced memory response after priming with PCV.

Pneumococcal polysaccharide vaccine (PPV) is of limited immunogenicity in infants and immunocompromised patients. Our prospective randomized controlled trial investigated whether priming with pneumococcal conjugate vaccine (PCV) induced specific immunological memory in previously nonresponders to PPV. Of a total of 33 children (2 to 18 years) with polysaccharide-specific immunodeficiency (PSI), group A (n = 16) received two doses of 7-valent PCV in a 4- to 6-week interval, and a booster dose of 23-valent PPV after one year. Group B (n = 17) received two doses of PPV in a 1-year interval exclusively. Specific antibody concentrations for serotypes 4, 5, 6B, 9V, 14, 18C, 19F, and 23F were determined (enzyme-linked immunosorbent assay) before and at 7 and 28 days after administration of the PPV booster and compared to an opsonophagocytosis assay. Of group A, 64 to 100% had antibody concentrations of ≥1 μg/ml on day 28 after the booster versus 25 to 94% of group B. Group A had significantly higher antibody concentrations for all PCV-containing serotypes already on day 7, indicating early memory response. Antibody concentrations were in accordance with functional opsonic activity, although opsonic titers varied among individuals. Pneumococcal vaccination was well tolerated. The incidence of airway infections was reduced after priming with PCV (10/year for group A versus 15/year for group B). Following a PPV booster, even patients primarily not responding to PPV showed a rapid and more pronounced memory response after priming with PCV.
cytic activity (OPA) of the induced anticasrular antibodies. This functional activity of antibodies in killing of pneumococci is thought to correlate better with the efficacy needed to prevent infections than antibody levels measured solely by enzyme-linked immunosorbent assay (ELISA).

MATERIALS AND METHODS

Our study cohort encompassed 33 children fulfilling the following criteria for PSI: recurrent airway infections (e.g., pneumonia or otitis media; more than three per year), a lacking immune response to repeat (at least two; range, two to three) PPV applications, and a normal immune response to protein vaccines. A lacking immune response was defined as pneumococcal serotype-specific IgG antibody concentrations of <1.0 μg/ml in at least five of seven serotypes 4 weeks after the last immunization with PPV. Exclusion criteria were a history of allergic or serious adverse reactions to previous vaccinations. Other exclusion criteria were progressive neurological disease or any current illness. All parents supplied written informed consent prior to the study. Human experimentation guidelines of Good Clinical Practice, the German Drug Act, and the declaration of Helsinki/Hong Kong were followed in the conduct of clinical research. Local ethical committee approval was obtained. During the 1-year period before the PPV-23 booster was administered, clinically defined airway infections were documented for both groups on diary cards.

Group A received two doses of the 7-valent PCV and a booster dose of the 23-valent PPV 1 year after the first PCV-7 dose. Group B received one dose of PPV and another dose of PPV after 1 year without previous PCV priming.

**Vaccines.** The 7-valent PCV (Prevenar, Wyeth-Lederle, Germany) contains the polysaccharide capsular serotypes 4, 9V, 14, 18C, 19F, and 23F at 2 μg/ml and 6B at 4 μg/ml, coupled with a carrier protein, a nontoxic variant of diphtheria toxin (cross-reactive material 197; CRM197). The 23-valent PPV (Pneumovax23, Merieux MSD, Germany) contains serotypes 1 through 5, 6B, 7F, 8, 9N, 9V, 10A, 11A, 12F, 14, 15B, 17F, 18C, 19F, 19A, 20, 22F, 23F, and 33F at 25 μg each. Both vaccines were administered intramuscularly, and local and systemic side effects were recorded on diary cards in the 3 days after each immunization.

**Serological analysis.** Serological determinations were performed blinded at our laboratory. The staff were not aware of age, vaccination status, or medical background of the vaccinees at the time of sampling. Serum antibodies against two highly immunogenic (14 and 19F) and two low-immunogenic serotypes (6B and 23F) and against serotypes 4, 9V, 18C, and the exclusively PPV-23-containing type 5 were determined before and 7 and 28 days after the PPV booster was administered. Serotype-specific IgG antibody concentrations were measured by a modified ELISA technique using Nunc CovaLink microtiter plates (Nunc, Germany) and serum lot 89-SF as the standard serum (FDA, Bethesda, MD). Serum samples were preincubated with 10 μg/ml pneumococcal polysaccharide C (CPS, Statens Serum Institut, Denmark) for blocking of unspecific anti-CPS antibodies. Reference serum 89-SF (provided by C. Frash, Rockville, MD) was used as quality control. We used the SPSS for Windows version 11.0 software package. Probability values (P) <0.05 were considered statistically significant. Nonparametric correlations between antibody titers and opsonic activity were calculated with Spearman’s correlation coefficient.

**RESULTS**

Demographics. Thirty-three patients (age, 2 to 18 years; 19 boys, 14 girls) were enrolled in the study and randomly assigned to group A (16 patients) or B (17 patients) (Table 1). All patients returned completed diary cards. Each pneumococcal vaccination was well tolerated in all patients. Adverse reactions were similar in both groups (Table 2); no severe adverse reaction was observed. The median ages of the two groups differed slightly (the median [range] was 7 [5 to 12] years for group A versus 4 [2 to 14] years for group B; P <0.05).

At study entrance, groups were comparable concerning frequency of airway infections (>3/year; see inclusion criteria) and medical history. Remarkable features of the vaccines included bronchial asthma (group A, 47%; group B, 53%).

| Parameter | Value for indicated group | A (PCV primed) | B (unprimed) |
|-----------|--------------------------|----------------|--------------|
| Age (yr)  |                          | n=16 | n=17         |
| Range     |                          | 5–12 | 2–14         |
| Median    |                          | 7    | 4            |
| Gender    |                          | 8/8  | 11/6         |
| Underlying diagnosis |                  | IgG, deficiency | 10 | 13 |
|           |                          | IgA deficiency | 3  | 2 |
|           |                          | Hypogammaglobulinemia | 1 | 3 |
|           |                          | Asthma bronchiale | 8 | 9 |
|           |                          | Chronic bronchitis | 1  | 1 |
|           |                          | Recurrent pneumonia | 1  | 1 |
|           |                          | Recurrent otitis | 2  | 1 |
|           |                          | Recurrent bronchitis | 1 | 3 |
| Infections during 12 mo prior to PPV-23 booster administration | | Bronchitis | 5 | 8 |
|          |                          | Pneumonia | 5 | 5 |
|          |                          | Otisit | 2  | 1 |
|          |                          | Tonsillitis | 3 | 1 |
|          |                          | Fever of unknown origin | 3 | 4 |

**TABLE 1. Demographics, underlying diagnosis, and clinical diagnosed infections of subjects unresponsive to pneumococcal polysaccharide vaccine**
IgG2 subclass deficiency (group A, 59%; group B, 76%), and 18% of group A suffered from IgA deficiency. After PCV vaccination of group A, there were fewer upper and lower airway infections during the year prior to the administration of the PPV-23 booster (10 cumulative in group A versus 15 in group B and a median of 10, with 5 remarkable episodes of pneumonia in the PCV-7-primed group versus none in the other).

**Immunogenicity.** Complete blood sample series were available from all 33 subjects for analysis of the immune responses to the booster dose of PPV. Initial geometric mean antibody concentrations were higher in group A after two PCV-7 applications (0.64 to 3.5 μg/ml) compared to group B after one PPV-23 application (0.17 to 1.13 μg/ml) with significance for the serotypes 4, 6B, 14, 19F, and 23F (Table 3). Following administration of the the PPV booster, antibody concentrations increased even more and were significantly higher in group A: on day 7, group A had 1.76 to 8.39 μg/ml versus 0.21 to 1.67 μg/ml in group B with significance for all serotypes except the non-PCV serotype 5 (Table 2). On day 28, antibody GMCs increased higher from baseline values in group A (1.25- to 2.1-fold) than in group B (1.2- to 2.1-fold) (Fig. 1). Also after 28 days, median antibody GMCs increased higher from baseline values in group A (1.9- to 7.5-fold), compared to 1.6- to 3.3-fold in group B (P < 0.01 for serotype 4) (Fig. 2).

In summary, the antibody response after PCV administration and after booster vaccination was higher and faster in the PCV-primed group A for all PCV-related serotypes as compared to the unprimed group.

For the exclusively PPV-23 serotype 5, there was no significant difference between groups at any time (Table 3). Before the PCV boost, the GMCs were 0.64 μg/ml in group A (0.54 to 0.75) and 0.73 μg/ml (0.48 to 0.99) in group B. On day 7, the group A GMC was 0.73 μg/ml (range, 0.48 to 0.99) and that for group B was 0.95 μg/ml (range, 0.31 to 1.58). On day 28, the GMCs were 1.25 μg/ml (range, 0.92 to 1.58) for group A and 1.21 μg/ml (range, 0.33 to 2.10) for group B. For group A, the rise of antibody levels from baseline values was 1.4-fold on day 7 and 1.95-fold on day 28 (P < 0.05 each). For group B, there

| Symptom                        | No. (%) of infants with symptom in: |
|--------------------------------|-------------------------------------|
|                                | Group A: PCV-7 priming | Group B: solely PPV-23 |
|--------------------------------|----------------------------|--------------------------|
| Redness                        | 6 (35)                    | 6 (35)                   |
| Induration                     | 2 (12)                    | 3 (18)                   |
| Swelling                       | 2 (12)                    | 1 (6)                    |
| Pain at injection site         | 8 (47)                    | 9 (53)                   |
| Irritability                   | 2 (12)                    | 1 (6)                    |
| Drowsiness                     | 2 (12)                    | 1 (6)                    |
| Lack of appetite               | 0                         | 3 (18)                   |
| Body temperature of >38.0°C    | 2 (12)                    | 1 (6)                    |

**TABLE 2.** Number of infants experiencing systemic reactions in the 3 days following each immunization

| PnC                     | GMC (μg/ml) (95% CI) | Group A (PCV-7 primed) (n = 16) | Group B (PPV-23) (n = 17) |
|-------------------------|----------------------|--------------------------------|--------------------------|
|                         | Day 0    | Day 7   | Day 28  | Day 0   | Day 7   | Day 28  | Comparison of groups A and B |
| 4                       | 0.75 (0.45–1.05)    | 3.59** (0.00–8.25) | 5.62* (1.80–9.44) | 0.39 (0.17–0.61) | 0.60* (0.21–0.99) | 1.02 NS (0.50–1.54) | ** *** *** |
| 5                       | 0.64 (0.54–0.75)    | 0.91* (0.74–1.08) | 1.25* (0.92–1.58) | 0.73 (0.48–0.99) | 0.95 NS (0.31–1.58) | 1.21 NS (0.33–2.10) | NS NS NS |
| 6B                      | 0.64 (0.00–1.44)    | 1.76** (0.00–3.85) | 2.24 NS (0.18–4.29) | 0.18 (0.02–0.33) | 0.27* (0.00–0.62) | 0.32 NS (0.07–0.57) | *** *** *** |
| 9V                      | 0.82 (0.19–1.45)    | 2.37** (0.00–7.22) | 4.28** (0.00–10.4) | 0.43 (0.00–2.18) | 0.83* (0.00–1.68) | 0.83 NS (0.00–2.17) | NS * ** |
| 14                      | 3.50 (2.30–4.49)    | 8.39** (0.00–18.4) | 13.00 (0.00–28.7) | 1.13 (0.54–1.73) | 1.48* (0.86–2.11) | 1.75 NS (1.38–2.12) | *** *** *** |
| 18C                     | 0.86 (0.00–2.09)    | 2.35* (0.63–4.08) | 2.94 NS (1.54–4.33) | 0.39 (0.20–0.59) | 0.78* (0.41–1.15) | 1.13 NS (0.29–1.96) | NS * * |
| 19F                     | 2.30 (0.00–5.27)    | 7.20** (0.00–15.02) | 10.59* (2.35–18.8) | 0.79 (0.34–1.24) | 1.67* (1.25–2.10) | 2.60** (1.22–3.99) | *** *** |
| 23F                     | 0.88 (0.11–1.63)    | 2.93* (0.00–7.22) | 3.54 NS (0.25–6.85) | 0.17 (0.03–0.31) | 0.21* (0.00–0.62) | 0.28 NS (0.00–0.88) | ** *** |

*PnC, pneumococcal serotype; n, number of patients; NS, not significant. *, P < 0.05; **, P < 0.01; ***, P < 0.001.

*FIG. 1. Increase of specific pneumococcal antibodies after vaccination.*
was a 1.3-fold rise on day 7 and a 1.66-fold rise on day 28, which reached no statistical significance.

Opsonophagocytic activity. OPA was determined for the low-immunogenic serotype 23F before and 7 and 28 days after the booster vaccination. Prior to immunization, median opsonic titers were 1:4 (1:4 to 1:2,048) for group A and 1:4 (1:4 to 1:32) for group B. On day 7, group A showed a median activity of 1:4 (1:4 to 1:1,024) and group B of 1:4 (1:4 to 1:1,024). On day 28, median opsonic titers were 1:256 (1:4 to 1:2,048) for group A and 1:4 (1:4 to 1:512) for group B.

There was a direct correlation (Spearman \( r = 0.72; P < 0.01 \)) between antibody concentrations and functional OPA (Fig. 2). OPA varied highly, especially in group A, reflecting high antibody activity after conjugate vaccination. However, most individuals with a minimum antibody concentration of 1 \( \mu \text{g/ml} \) had high OPA.

Seroprotection. A positive immune response to pneumococcal serotypes was defined as an antibody concentration of \( \geq 1.0 \) \( \mu \text{g/ml} \) and an opsonophagocytic titer of >1:64. Accordingly, after 28 days, the percentage of responders was determined for each group and revealed significantly more responders for low-immunogenic serotypes 6B and 23F in group A, with 80.0% and 73.7% of patients, compared to group B, with 25.0% each (\( P < 0.01 \)). There was no significant difference between responders in each group for the highly immunogenic serotypes 14 and 19F, even though the antibody concentration was significantly higher in the primed group. Interestingly, even for serotype 5, a higher percentage of responders was found in the primed group A (64.3% versus 43.7% in group B).

Additionally, to further investigate the kinetics of immune response, we determined the number of nonresponders to all pneumococcal serotypes, remaining below the level of 1.0 \( \mu \text{g/ml} \) for each pneumococcal serotype (Table 4). As early as on day 7 postvaccination, between 0 (serotypes 14 and 19F) and 33.3% (serotypes 6B, 18C, and 23F) of group A subjects remained below this critical threshold compared to between 5.9% (serotype 19F) and 82.4% (serotype 23F) of group B subjects. For the non-PCV serotype 5, the percentage was similar in both groups (57.1% in group A versus 62.5% in group B).

**DISCUSSION**

Pneumococcal conjugate vaccines are immunogenic in infants and induce long-term protection by inducing systemic anamnestic IgG response (10, 37, 43). This has been demonstrated by studying antibody responses to pure polysaccharide antigens in infants who previously received conjugate vaccines (10, 34). Significant responses to plain polysaccharide vaccine challenge are generally not seen in unprimed infants. Thus, in

![FIG. 2. Pneumococcal opsonophagocytosis titers versus antibody concentrations (\( r = 0.72; P < 0.01 \)).](image)

**TABLE 4. Percentage of subjects with IgG antibody concentrations below 1.0 \( \mu \text{g/ml} \) before and 7 and 28 days after pneumococcal polysaccharide booster vaccination**

| PnC | Group A (PCV-7 primed) | Group B (unprimed) |
|-----|------------------------|---------------------|
|     | Day 0 | Day 7 | Day 28 | Day 0 | Day 7 | Day 28 |
| 4   | 68.8  | 6.7   | 6.7   | 87.5  | 70.6  | 56.3   |
| 5   | 85.7  | 57.1  | 35.7  | 75.0  | 62.5  | 56.3   |
| 6B  | 43.8  | 33.3  | 20.0  | 100   | 76.5  | 75.0   |
| 9V  | 68.8  | 20.0  | 13.3  | 81.3  | 47.1  | 62.5   |
| 14  | 6.3   | 0.00  | 0.00  | 31.3  | 17.6  | 6.3    |
| 18C | 68.8  | 33.3  | 13.3  | 81.3  | 47.1  | 37.5   |
| 19F | 12.5  | 0.00  | 0.00  | 43.8  | 5.9   | 6.3    |
| 23F | 37.5  | 33.3  | 26.7  | 100   | 82.4  | 75.0   |

*PnC, pneumococcal serotypes.*
### TABLE 5. Serotype-specific IgG antibody GMCs of patients previously not responding to pneumococcal polysaccharide vaccination, stratified for age

| Group A (PCV-7 primed) | Group B (PPV-23 only) |
|------------------------|-----------------------|
| **PnC**                | **PnC** |
| **Group**              | **Day 0** | **Day 7** | **Day 28** | **Group** | **Day 0** | **Day 7** | **Day 28** |
| **2–5 yr old**         | 6.99 (5.86–8.55) | 2.37* (1.76–3.13) | 0.77b | 1.75 (1.29–2.36) | 0.88 (0.62–1.24) | 0.69 (0.48–1.15) |
| **5 yr old**           | 3.70 (2.63–5.03) | 1.07 (0.72–1.57) | 0.39 | 1.25 (0.90–1.74) | 0.82 (0.62–1.09) | 0.59 (0.45–0.77) |
| **0–2 yr old**         | 0.75 (0.54–1.01) | 0.28 (0.14–0.55) | 0.17 | 0.89 (0.62–1.21) | 0.62 (0.44–0.89) | 0.41 (0.29-0.56) |
| **≥7 yr old**          | 1.29 (0.61–2.36) | 0.42 (0.23–0.74) | 0.26 | 0.88 (0.53–1.46) | 0.62 (0.37–1.04) | 0.39 (0.22–0.70) |

### Notes
- *PnC pneumococcal serotype-specific antibody GMCs, number of patients.
- **PnC**: pneumococcal polysaccharide in certain risk groups (22, 30, 32, 39, 41, 46, 55). However, antibody concentrations are not the only correlate of a protective immunity. As we know from *Haemophilus influenzae*, conjugate vaccine efficacy is also determined by differences in the kinetics of immune response (17). Demonstration of B-cell memory has largely been based on a rapid and strong antibody response to a dose of plain polysaccharide vaccine after priming with a conjugate vaccine (32, 33, 54).

There are only limited data on the kinetics of immunological memory in high-risk groups. In our study, the anamnestic immune response after the PPV booster vaccination was demonstrated by the significantly more rapid and higher antibody increase already within 7 days in the PCV-primed group A. Moreover, the majority of patients primed with PCV-7 had protective pneumococcal antibody levels (≥1.0 μg/ml) for all PCV-7 included serotypes already on day 7. This threshold value also implicates long-term protection. Even for the weak immunogenic serotypes 6B, 18C, and 23F, about two-thirds of our subjects reached this threshold compared to 24%, 53%, and 18%, respectively, in the unprimed group (Table 3). Obviously, the kinetics of pneumococcal antibodies are serotype dependent, an observation also supported by others (15). As there was only a marginal additional rise in antibody concentrations from day 7 to day 28 in these patients, the IgG antibody peak might have been reached even earlier. This observation is of special clinical importance when natural contact and the organisms occurs and rapid protection is required (e.g., in unvaccinated immunocompromized patients as post-exposure immunization). Also, group B individuals previously not responding to PPV-23 demonstrated a moderate immune response seven days after the PPV booster. This might be also due to having grown one year older in the meantime. While group A subjects showed a further increase of specific pneumococcal antibodies (significant for serotypes 4, 5, 9V, 14, and 19F) 28 days after PPV-23 administration, in group B, this was observed only for the highly immunogenic serotype 19F and in a much smaller amount.

Overall the development of a polysaccharide-specific memory seems to be influenced by the age at priming with the glycoconjugate, the route of immunization, the time and features of the booster, and other factors, as also demonstrated in an early-life murine model reproducing the main features of infant responses to pneumococcal vaccination (7, 24). Two other studies in healthy senior subjects could not find a benefit from a PPV-23 booster 1 (42) or 6 months (35) after PCV-7 immunization. The subsequent administration provided no additional antibody response. On the other hand, in a Dutch study 383 children (1 to 7 years old) with recurrent otitis media were immunized with either PCV followed 6 to 7 months later by PPV-23, or by hepatitis A or B vaccine (50). In the PPV-23-boosted group, antipneumococcal antibodies against PCV-containing serotypes were much higher. Also a study in 386 healthy United Kingdom infants showed results in favor of the PPV-23 booster. They received three doses of PCV-7 or placebo in the first year of life, followed by a PPV-23 booster at
13 to 16 months of age. The PPV-23 booster resulted in a 3.4- to 51.7-fold rise of pneumococcal anticapsular IgG antibodies depending on the serotype and immunization schedule (10).

PCV repeated priming like in our protocol has the potential to support a booster response, improving immunogenicity and efficacy. On the other hand, there is evidence that even a single dose of a conjugate vaccine may increase antibody avidity (5, 6), and that higher avidity antibodies are more cross-reactive with closely related pneumococci serotypes. Also, prevaccination antibodies are more cross-reactive than postvaccination antibodies (44). In our study, we documented the frequency of airway infections in our subjects in order to estimate clinical efficacy prior to the PPV-23 challenge. Interestingly, there was a marked reduction of previously frequent infections in the PCV-primed patient group (Table 1).

We are aware of a minor selection bias, since the vaccinees in group A were slightly older than in group B. Individuals up to 5 years of age are regarded as relatively immature, which finds its expression also in vaccination strategies (4). In order to create comparable patient groups, we stratified our study subjects as follows: patients up to 5 years of age and older than 5 years. The analysis of these data confirmed our results with the restriction of smaller group sizes (Table 5). This better immunogenicity of PCV compared to PPV beyond infancy has also been suggested elsewhere (21, 39). Induction of immunologic memory after priming with a pneumococcal conjugate vaccine finds its expression also in vaccination strategies (4).

In order to create comparable patient groups, we stratified our study subjects as follows: patients up to 5 years of age and older than 5 years. The analysis of these data confirmed our results with the restriction of smaller group sizes (Table 5). This better immunogenicity of PCV compared to PPV beyond infancy has also been suggested elsewhere (21, 39). Induction of immunologic memory after priming with a pneumococcal conjugate vaccine finds its expression also in vaccination strategies (4).

In conclusion, following a PPV booster, even patients primarily not responding to solely PPV, showed a rapid and more pronounced memory response after priming with PCV. There was a good correlation between serum antibody GMCs and OPA as a functional surrogate for protection.

REFERENCES

1. Åhman, H., H. Käbyth, H. Lehtonen, O. Leroy, J. Froschle, and J. Eskola. 1998. Streptococcus pneumoniae capsular polysaccharide-diphtheria toxoid conjugate vaccine is immunogenic in early infancy and able to induce immunologic memory. Pediatr. Infect. Dis. J. 17:211–216.

2. Aronson, C. M. Rodriguez, M. C. Senne, G. R. Hamilton, D. M. Musher, and K. E. Nelson. 1996. Effect of human immunodeficiency virus type 1 infection on the antibody response to a glycopeptid conjugate pneumococcal vaccine: results from a randomized trial. J. Infect. Dis. 173:790–793.

3. Ambrosino, D. M., G. R. Silver, R. A. Chilmonczyk, J. B. Jernberg, and R. W. Finberg. 1987. An immunodeficiency characterized by impaired antibody responses to polysaccharides. N. Engl. J. Med. 316:790–793.

4. Anttila, M., J. Eskola, H. Åhman, and H. Käbyth. 1999. Differences in the avidity of antibodies evolved by four different pneumococcal conjugate vaccines in early childhood. Vaccine 17:1970–1977.

5. Anttila, M., J. Escalera, H. Åhman, and H. Käbyth. 1998. Avidity of IgG for Streptococcus pneumoniae type 6B and 23F polysaccharides in infants primed with pneumococcal conjugates and boosted with polysaccharide or conjugate vaccines. J. Infect. Dis. 177:1614–1621.

6. Bjarnason, S. P., H. Jakobsen, G. del Giudice, E. Trannoy, C.A. Siegrist, and I. Jonsdottir. 2005. The advantage of mucosal immunization for polysaccharide-specific memory responses in early life. Eur. J. Immunol. 35:1037–1045.

7. Black, S. B., R. R. Shinfield, J. Hansen, L. Elvin, D. Lauer, and F. Malinoski. 2001. Postlicensure evaluation of the effectiveness of seven valent pneumococcal conjugate vaccine. Pediatr. Infect. Dis. J. 20:1105–1107.

8. Chan, C. Y., D. C. Molrine, S. George, N. J. Tarbell, P. Mauch, L. Diller, S. B. Black, H. R. Shinefield, J. Hansen, L. Elvin, D. Laufer, and F. Malinoski. 2001. Postlicensure evaluation of the effectiveness of seven valent pneumococcal conjugate vaccine. Pediatr. Infect. Dis. J. 20:1105–1107.

9. Concepcion, N. F., and C. E. Frasch. 2000. Immunogenicity and reactivity of a pneumococcal conjugate vaccine administered combined with a Haemophilus influenzae type B conjugate vaccine in United Kingdom infants. Pediatr. Infect. Dis. J. 19:854–862.

10. Cho, S., L. Smyour, R. Morris, S. Quataert, S. Lockhart, K. Cartwright, and A. Finn. 2000. Immunogenicity and reactogenicity of a pneumococcal conjugate vaccine administered combined with a Haemophilus influenzae type B conjugate vaccine in United Kingdom infants. Pediatr. Infect. Dis. J. 19:854–862.

11. Chaney, N. F., and C. E. Frasch. 2001. Pneumococcal type 22F polysaccharide absorption improves the specificity of a pneumococcal-polysaccharide enzyme-linked immunosorbent assay. Clin. Diag. Lab. Immunol. 8:266–272.

12. Dagan, R., R. Melamed, O. Zamir, and O. Leroy. 1997. Safety and immunogenicity of tetravalent pneumococcal vaccines conjugate vaccines containing 6B, 14, 19F and 23F polysaccharide conjugated to either tetanus toxoid or diphtheria toxoid in young infants and their boosterability by native polysaccharide antigens. Pediatr. Infect. Dis. J. 16:1053–1059.

13. Dagan, R., M. Mualem, R. Melamed, O. Leroy, and P. Yagupsky. 1997. Reduction of pneumococcal nasopharyngeal carriage in early infancy after immunization with tetravalent pneumococcal vaccines conjugated to either tetanus toxoid or diphtheria toxoid. Pediatr. Infect. Dis. J. 16:1060–1064.

14. Dagan, R., R. Melamed, M. Mualem, L. Piglansky, D. Greenberg, O. Abramson, P. M. Mendelman, N. Bohidar, and P. Yagupsky. 1996. Reduction of nasopharyngeal carriage of pneumococci during the second year of life by a heptavalent conjugate pneumococcal vaccine. J. Infect. Dis. 174:1271–1278.

15. Ekström, N., H. Åhman, J. Verho, J. Jokinen, M. Väkeväinen, T. Kilpi, H. Käbyth, and the Finnish Otitis Media Study Group. 2005. Kinetics and...
avidity of antibodies evoked by heptavalent pneumococcal conjugate vac-
cines PncCRM and ProMPIC in the Finnish Ottis Media Vaccine Trial. 
Infect. Immun. 73:369–377.

18. Esoka, J., and M. Antilla. 1999. Pneumococcal conjugate vaccines. Pediatr.
Infect. Dis. J. 18:S33–S51.

17. Granoff, D. M., E. L. Anderson, M. T. Osterholm, S. J. Holmes, J. E. 
McHugh, R. B. Belshe, F. Medley, and T. V. Murphy. 1992. Differences in the 
immunogenicity of three Haemophilus influenzae type b conjugate vaccines 
in infants. J. Infect. Dis. 165:187–194.

16. Jodar, L., J. Butler, G. Carlone, R. Dagan, C. Frasch, and T. Cherian. 1999. 
Serological criteria for evaluation and licensure of new pneumococcal con-
jugate vaccine formulations for use in infants. Vaccine 17:3265–3272.

15. Kauppi, M., J. Eskola, and H. Käyhty. 1995. Anti-capsular pneumococcal 
antibody and phagocytosis of type 14 pneumococcal following 
immunization. Scand. J. Immunol. 36:96–98.

14. Kaniuk, A. C., J. E. Lortan, and M. A. Monteil. 1993. Development of 
heptavalent pneumococcal vaccine conjugated to CRM 197 
and B. M. Greenwood. J. Infect. Dis. 168:575–763.

13. Larsson, P., B. M. Rose, and S. Zielen. 1999. Evidence for induc-
tion of fimbriae in Haemophilus influenzae type b vaccines and the 
role of capsular polysaccharide in the immune response. J. Infect. Dis. 179:109–114.

12. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

11. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

10. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

9. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

8. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

7. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

6. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

5. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

4. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

3. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

2. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.

1. McHeyzer-Williams, M. G., D. J. Driver, and M. G. McHeyzer-Williams. 
1985. Thymus-independent and thymus-dependent responses to 
protein-polysaccharide conjugate vaccines in young and elderly 
adults. Infect. Immun. 65:242–247.