Serotype-specific effectiveness and correlates of protection for the 13-valent pneumococcal conjugate vaccine: a postlicensure indirect cohort study

Nick J Andrews, Pauline A Waight, Polly Burbidge, Emma Pearce, Lucy Roalfe, Marta Zancolli, Mary Slack, Shamez N Ladhani, Elizabeth Miller, David Goldblatt

Summary

Background **Efficacy of the 13-valent pneumococcal conjugate vaccine (PCV13) was inferred before licensure from an aggregate correlate of protection established for the seven-valent vaccine (PCV7). We did a postlicensure assessment of serotype-specific vaccine effectiveness and immunogenicity in England, Wales, and Northern Ireland to derive the correlates of protection for individual serotypes.**

Methods **We assessed vaccine effectiveness against invasive pneumococcal disease using the indirect cohort method. We measured serotype-specific IgG concentration in infants after they were given two priming doses of PCV7 (n=126) or PCV13 (n=237) and opsonophagocytic antibody titre from a subset of these infants (n=100). We derived correlates of protection by relating percentage protection to a threshold antibody concentration achieved by an equivalent percentage of infants. We used multivariable logistic regression to estimate vaccine effectiveness and reverse cumulative distribution curves to estimate correlates of protection.**

Findings **For the 706 cases of invasive pneumococcal disease included in the study, PCV13 vaccine effectiveness after two doses before age 12 months or one dose from 12 months was 75% (95% CI 58–84). Vaccine effectiveness was 90% (34–98) for the PCV7 serotypes and 73% (55–84) for the six additional serotypes included in PCV13. Protection was shown for four of the six additional PCV13 serotypes (vaccine effectiveness for serotype 3 was not significant and no cases of serotype 5 infection occurred during the observation period). The vaccine effectiveness for PCV13 and PCV7 was lower than predicted by the aggregate correlate of protection of 0·35 µg/mL used during licensing. Calculated serotype-specific correlates of protection were higher than 0·35 µg/mL for serotypes 1, 3, 7F, 19A, 19F, and lower than 0·35 µg/mL for serotypes 6A, 6B, 18C, and 23F. Opsonophagocytic antibody titres of 1 in 8 or higher did not predict protection.**

Interpretation **PCV13 provides significant protection for most of the vaccine serotypes. Although use of the aggregate correlate of protection of 0·35 µg/mL has enabled the licensing of effective new PCVs, serotype-specific correlates of protection vary widely. The relation between IgG concentration after priming and long-term protection needs to be better understood.**

Funding **Public Health England and UK Department of Health Research and Development Directorate.**

Lancet Infect Dis 2014; 14: 839–46

Published Online July 18, 2014 http://dx.doi.org/10.1016/S1473-3099(14)70822-9

See Comment page 784

Statistics, Modelling, and Economics Department, Health Protection Services (N J Andrews PhD), **Immunisation, Hepatitis, and Blood Safety Department, Health Protection Services** (P A Waight BSc, S N Ladhani MD, E Miller FRCPath), **and Respiratory and Vaccine Preventable Bacteria Reference Unit, Microbiology Services** (M Slack FRCPath), **Public Health England, London, UK; and Immunobiology Section, UCL Institute of Child Health, London, UK** (P Burbidge BSc, E Pearce BSc, L Roalfe BSc, M Zancolli PhD, Prof D Goldblatt MB ChB)

Correspondence to: Prof David Goldblatt, UCL Institute of Child Health, London WC1N 1EH, UK **d.goldblatt@ucl.ac.uk**

Introduction

Since 2010, the seven-valent pneumococcal conjugate vaccine (PCV7) has been replaced in many countries by higher valency vaccines containing ten (PCV10, GlaxoSmithKline, Brentford, UK) or 13 (PCV13, Pfizer, New York, NY, USA) serotypes. This change in vaccine became necessary as a result of the change in serotypes causing invasive pneumococcal disease, partly driven by the use of PCV7. The high effectiveness of PCV7 at reducing vaccine-type invasive pneumococcal disease[1] has been partly offset by an increase in invasive pneumococcal disease caused by non-vaccine serotypes.[2,3] The serotypes that emerged as major causes of invasive pneumococcal disease after the widespread use of PCV7 include many of the additional serotypes in PCV10 and PCV13.

Assessment of vaccine effectiveness for the newer, extended-valency PCVs is of particular interest because, unlike PCV7, PCV10, and PCV13 were licensed on the basis of immunogenicity data alone. Head-to-head studies of PCV7 and the new vaccines with immunogenicity endpoints were deemed acceptable for licensing the new conjugates,[4] largely because of the existence of a correlate of protection for PCV7. An anticapsular polysaccharide antibody concentration of 0·35 µg/mL measured by ELISA aggregated across all the seven serotypes in PCV7 is regarded as predictive of protection against invasive pneumococcal disease.[5] This correlate was derived from three randomised trials of PCV7 or an experimental nine-valent conjugate (Wyeth, Collegeville, PA, USA) done in California, USA,[6] in an Indigenous American population,[7] and in South Africa,[8] by correlation of post-primary IgG concentrations aggregated across the three studies with aggregate efficacy against invasive pneumococcal disease.[5]

Individual serotype-specific correlates could not be derived from the efficacy trials because even the largest PCV7 trial showed significant serotype-specific efficacy

for only three of the seven serotypes, with wide CIs because of small numbers of cases of invasive pneumococcal disease.[9] Researchers therefore derived an aggregate estimate, although serotype-specific differences in the amount of antibody needed to protect against invasive pneumococcal disease and otitis media are now recognised.[10,11] Pooled data from the heterogeneous populations investigated in the efficacy trials[6–8] were used to narrow the confidence limits around the point estimate of efficacy, despite different levels of protection seen in these three populations. With effectiveness estimates for the additional serotypes in the extended-valency conjugates now available, the possibility exists to derive serotype-specific correlates of protection. More precise estimates will both aid decision making relevant to vaccine schedules and inform the licensure of next-generation extended-valency conjugates.

PCV13 replaced PCV7 in the UK on April 1, 2010. For both vaccines, a 2+1 schedule was used (at 2, 4, and 12 months). We previously reported on the effectiveness of PCV13 against vaccine-type invasive pneumococcal disease in the first 15 months after introduction in a case-control (indirect cohort) study.[12] Here, we extend the estimates of effectiveness to 3·5 years after introduction. Additionally, using data from immunogenicity studies of PCV13 and PCV7 in infants in the UK and previously published data for the effectiveness of PCV7,[13] we aimed to derive individual serotype-specific estimates of correlates of protection for the vaccine serotypes and to calculate the first serotype-specific functional correlates of protection based on opsonophagocytic killing.

For the **ELISA protocol** see http://www.vaccine.uab.edu/ELISA%20Protocol.pdf

Methods

Setting

We did a postlicensure indirect cohort study to investigate the serotype-specific effectiveness and correlates of protection for PCV13. We used cases of invasive pneumococcal disease (diagnosed in infants by culture of *Streptococcus pneumoniae* from a normally sterile site or by DNA detection in pleural fluid or cerebrospinal fluid) for which a serotype was identified. Cases were reported during a sufficiently long surveillance period (3·5 years and about 700 cases) to produce serotype-specific estimates of vaccine effectiveness that we could use to produce and assess correlates of protection based on antibody measurements from available clinical trial data (n=126 for PCV7 serotypes; n=237 for PCV13 serotypes).

Procedures

To assess vaccine effectiveness, we used all cases of invasive pneumococcal disease in the cohort eligible for PCV13 vaccination in England, Wales, and Northern Ireland identified up to Oct 31, 2013, through enhanced national surveillance by Public Health England (responsible for surveillance in these countries; formerly known as the Health Protection Agency until 2013) and in whom the serotype of the infecting isolate was known.[3] To enhance study power, individuals born on or after April, 2008, were included as long as the onset of invasive pneumococcal disease was on or after March 30, 2010, because they might have received PCV13 as a booster dose. We obtained vaccination history, clinical risk group, and prematurity status from general practitioners through a postal questionnaire and telephone calls. Only individuals aged at least 2·5 months were included, and those with serogroup information only were excluded. For the assessment of vaccine effectiveness against the extra serotypes in PCV13 (including 6C), PCV7 serotypes were excluded; for the assessment of PCV13 vaccine effectiveness against PCV7 serotypes, any child who received PCV7 was excluded.

To derive correlates of protection, we used serum samples from children who had received PCV7 or PCV13 in two immunogenicity studies (Findlow and colleagues[6] and EudraCT 2010-023865-22/NCT01425372) by the UK National Vaccine Evaluation Consortium (NVEC). These studies were done in two representative populations in the middle of England and on the outskirts of London that have been used consistently by NVEC for vaccine studies to inform UK immunisation policy. For PCV7 and PCV13 serology, serotype-specific IgG was measured at age 5 months after PCV7 (n=126)[6] or PCV13 (n=237; EudraCT 2010-023865-22/NCT01425372) administration at ages 2 months and 4 months.

We used ELISA to assay serum samples for antibodies to 13 vaccine-type capsular polysaccharides (1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F) at the University College London Institute of Child Health (London, UK), a WHO reference laboratory for pneumococcal serology, as previously described.[14] We obtained prevaccination IgG titres from infants in the first year of life from historical data generated before the introduction of PCV7 in the UK.[15] In a random subset of serum samples (n=100), we measured functional antibodies to the 13 vaccine serotypes in a multiplexed opsonophagocytic assay, as previously described.[14] Values are expressed as an opsonophagocytic antibody titre equivalent to the reciprocal of the serum dilution needed to produce 50% killing of the relevant serotype.

Statistical analysis

We calculated vaccine effectiveness using a case-control design wherein the cases are individuals with vaccine-type invasive pneumococcal disease and controls are individuals with invasive pneumococcal disease caused by the non-PCV13 serotypes (Broome or indirect cohort method),[16] as described previously for UK PCV7 and PCV13 studies.[12,13] We used logistic regression to adjust for age (2·5–5, 6–12, 13–17, 18–23, 24–35, 36–47, and 48–56 months) and year of infection (2010, 2011, 2012, and 2013), and to examine the need to adjust for clinical risk group (since underlying comorbidities or prematurity might affect vaccine effectiveness).

Vaccine doses given within 14 days of onset of invasive pneumococcal disease were not counted and children with a single dose given within 14 days were excluded. We assessed vaccine effectiveness on the basis of the number of PCV13 doses received before age 12 months and from age 12 months, as well as by aggregated categories of at least one dose given at any age and at least two doses given before age 12 months or one dose from age 12 months onwards. We calculated serotype-specific vaccine effectiveness using these aggregated vaccination criteria (to enhance power) and for a category defined as two doses before age 12 months. PCV7 doses were not counted for assessment of PCV13 effectiveness, so most individuals with an incomplete number of PCV13 doses for age will have received previous PCV7 doses.

Vaccine effectiveness for PCV7 was based on previously published data,[13] but we reanalysed these to obtain estimates using the aggregated category of at least two doses given before age 12 months or one dose from age 12 months. We also estimated vaccine effectiveness for PCV7 against serotype 6A after retrospective testing of 6A isolates was done to distinguish 6A from 6C.[13]

To calculate correlates of protection, we used reverse cumulative distribution curves of vaccine serotype-specific IgG antibody in two ways. First, we applied the currently accepted correlate of protection (0·35 µg/mL) to predict vaccine effectiveness both for aggregate PCV7 or PCV13 serotypes from aggregate reverse cumulative distribution curves and, for individual serotypes, from individual reverse cumulative distribution curves. Second, using the estimates of vaccine effectiveness from the present study, we derived the aggregate and serotype-specific protective levels, as previously described.[17] Briefly, we applied the point estimate and 95% CIs of the percentage protection afforded by the vaccine (ie, vaccine effectiveness) to the distribution of serotype-specific IgG 1 month after the priming series of vaccinations to derive an IgG protective level for each individual serotype. For the aggregate correlate of protection, we combined serotype-specific values for the seven serotypes in PCV7, the additional six serotypes in PCV13, or all serotypes in PCV13. We used historical data for prevaccination antibody titres[15] to investigate the effect of correcting the correlate estimates for pre-existing antibodies, as described by Siber and colleagues.[17] Where possible, we used serotype-specific reverse cumulative distributions of the opsonophagocytic antibody titres to derive functional serotype-specific correlates of protection.

Role of the funding source

The funder of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

Results

Of 716 serotyped cases of invasive pneumococcal disease, eight were assigned a serogroup only (serogroup 7) and were therefore excluded, and two other individuals had an unknown vaccination status. The remaining 706 cases included 30 PCV7 serotypes, 292 with the additional six PCV13 serotypes or 6C, and 414 with non-PCV13 serotypes. The serotype distribution varied by age, because of vaccine effects and possible natural differences in serotype distribution by age (table 1, appendix p 1). As expected, vaccination status varied by age, suggesting the need to adjust for age when estimating vaccine effectiveness. A full breakdown by vaccine status for the assessment of vaccine effectiveness by serotype is shown in the appendix (pp 2–4).

See Online for appendix

The estimated vaccine effectiveness for the PCV13 serotypes not in PCV7 (plus 6C) at the 2+1 schedule was 79% (95% CI 25–94), although the 95% CIs were wide because only patients old enough to have completed this schedule could be included (table 2). Vaccine effectiveness against PCV7 serotypes for at least two doses before age 12 months or one dose from age 12 months onwards was high at 90% (34–98), although precision was low because of low numbers. Vaccine effectiveness for at least two doses before age 12 months or one dose from 12 months onwards was 73% (55–84) against the additional serotypes included in PCV13 (plus 6C), but varied by serotype (table 2).

	Age range (months)							Total
	2·5–5	6–12	13–17	18–23	24–35	36–47	48–56	
Serotype								
Non-PCV13	55	139	75	49	46	37	13	414
All PCV13 serotypes (plus 6C)	45	67	33	32	69	36	10	292
4	0	0	0	2	0	0	0	2
6B	2	2	0	1	2	0	0	7
9V	0	0	1	0	0	0	0	1
14	1	0	1	0	1	0	0	3
18C	1	1	0	0	0	1	0	3
19F	3	4	0	2	0	0	0	9
23F	0	2	0	1	1	0	1	5
1	3	2	4	4	14	9	4	40
3	4	7	7	11	18	5	2	55
5	0	0	0	0	0	0	0	0
6A*	2	4	1	0	2	1	0	10
6C	2	1	2	1	0	1	0	7
7F	22	11	3	4	11	8	0	59
19A	5	33	13	6	20	11	3	91
PCV13 vaccinated†								
No	24	47	33	17	65	45	14	249
Yes	71	158	75	64	50	28	9	451

PCV13=13-valent pneumococcal conjugate vaccine. *Includes three serotyped as type 6 A/C. †Six individuals who received one dose within 14 days before onset were excluded.

Table 1: **Serotype distribution and numbers of individuals with at least one PCV13 dose by patient age for the 706 cases of invasive pneumococcal disease used in the calculations of vaccine effectiveness**

	Age range (months)	Cases vaccinated: unvaccinated	Controls vaccinated: unvaccinated	Adjusted vaccine effectiveness* (95% CI)
PCV13 serotypes not in PCV7 (plus 6C)† (1, 3, 6A, 6C, 7F, and 19A)				
At least one dose	2·5 to ≤56	105:155	330:80	73% (57 to 83)
At least two doses before age 12 months or one dose on or after age 12 months	4 to ≤56	76:146	280:76	73% (55 to 84)
One dose (before age 12 months)	2·5 to <13	28:43	48:24	60% (12 to 82)
Two doses (before age 12 months)	4 to <13	23:34	118:20	80% (43 to 93)
One dose (on or after age 12 months)	13 to ≤56	28:112	65:56	73% (50 to 85)
2 + 1 dosing schedule	13 to ≤56	13:112	71:56	79% (25 to 94)
PCV7 serotypes‡ (4, 6B, 9V, 14, 18C, 19F, and 23F)				
At least one dose	2·5 to ≤56	18:4	248:8	83% (35 to 96)
At least two doses before age 12 months or one dose on or after age 12 months	4 to ≤56	10:2	205:5	90% (34 to 98)
Pooled estimate for all PCV13 serotypes (plus 6C)				
At least two doses before age 12 months or one dose on or after age 12 months	4 to ≤56	NA§	NA§	75% (58 to 84)
1				
At least two doses before age 12 months or one dose on or after age 12 months	4 to ≤56	9:28	280:76	84% (54 to 95)
Two doses (before age 12 months)	4 to <13	1:1	118:20	NA¶
3				
At least two doses before age 12 months or one dose on or after age 12 months	4 to ≤56	21:28	280:76	26% (–69 to 68)
Two doses (before age 12 months)	4 to <13	3:2	118:20	66% (–322 to 92)
6A‖				
At least two doses before age 12 months or one dose on or after age 12 months	4 to ≤56	1:7	280:76	98% (64 to 99·8)
Two doses (before age 12 months)	4 to <13	1:3	118:20	96% (41 to 99·8)
7F				
At least two doses before age 12 months or one dose on or after age 12 months	4 to ≤56	5:35	280:76	91% (70 to 98)
Two doses (before age 12 months)	4 to <13	3:11	118:20	76% (–122 to 97)
19A				
At least two doses before age 12 months or one dose on or after age 12 months	4 to ≤56	30:53	280:76	67% (33 to 84)
Two doses (before age 12 months)	4 to <13	14:17	118:20	62% (–55 to 90)

PCV=pneumococcal conjugate vaccine. NA=not applicable. *Adjusted by age (2·5–5, 6–12, 13–17, 18–23, 24–35, 36–47, or 48–56 months) and year of infection (2010, 2011, 2012, or 2013). †No cases of serotype 5 were recorded. ‡Only children born from January, 2010, who did not receive any PVC7 doses were included in this analysis. §Pooled estimate, no single ratio of vaccinated:unvaccinated cases and controls possible. ¶Too few cases to calculate. ‖Includes three cases typed as 6A/C.

Table 2: **Vaccine effectiveness estimates, by serotype and schedule**

Children with comorbidities were infected with non-PCV13 serotypes more often than were healthy children (appendix p 1). The vaccine effectiveness against the additional serotypes included in PCV13 (plus 6C) for healthy children with at least two doses before age 12 months or one dose from 12 months onwards was 79% (95% CI 59 to 90), compared with –12% (–307 to 69) for those in a risk group (interaction p=0·008). Despite this interaction, adjustment for risk group status made little difference to the overall (risk and non-risk combined) vaccine effectiveness estimates; for example, vaccine effectiveness for at least two doses before age 12 months or one dose from 12 months onwards was reduced from 73% (95% CI 55–84) to 71% (47–84).

The predicted and observed values for vaccine effectiveness were not concordant (table 3). The predicted vaccine effectiveness was higher than the observed vaccine effectiveness for individual serotypes 1, 3, 7F, 19A, and 19F, as well as the aggregate PCV13 serotypes plus serotype 6C combined (figure) and the aggregate PCV7 serotypes combined before the introduction of PCV13 (82% observed *vs* 93% predicted; table 3), suggesting that 0·35 µg/mL is an underestimate of the true correlate of protection. For serotypes 6A, 6B, 18C, and 23F, predicted values for vaccine effectiveness were below the observed vaccine effectiveness, suggesting that 0·35 µg/mL is an overestimate and less antibody is required for protection. The point estimates for the correlates of protection were higher than 0·35 µg/mL for serotypes 1, 3, 7F, 9V, 19A, 14, and 19F (table 3), and for the aggregate PCV13 serotypes plus serotype 6C the point estimate was 0·98 µg/mL. By contrast, the correlate of protection for serotype 4 was exactly 0·35µg/mL and for 6A, 6B, 18C, and 23F the correlates were less than 0·35 µg/mL. Adjustment for pre-existing antibody in unvaccinated infants had little effect on either the predicted vaccine effectiveness or the calculated correlate of protection for most serotypes because, generally, IgG concentrations were low in vaccine-naive individuals (data not shown).

	Vaccine effectiveness (95% CI)	Predicted vaccine effectiveness at 0·35 µg/mL ELISA cutoff*	Calculated correlate of protection in µg/ml for ELISA* (95% CI)	Calculated correlate of protection in titres for opsonophagocytic antibody* (95% CI)
PCV13				
1	84% (54 to 95)	96%	0·78 (0·47 to 1·68)	4 (4 to 8)
3	26% (–69 to 68)	97%	2·83 (1·16 to ∞)	39 (14 to ∞)
6A†	98% (64 to 99·8)	90%	0·16 (0·08 to 1·05)	4 (4 to 824)
7F	91% (70 to 98)	98%	0·87 (0·40 to 1·80)	769 (373 to 1502)
19A	67% (33 to 84)	95%	1·00 (0·60 to 2·47)	48 (15 to 234)
5	..	89%	..	..
Extra serotypes in PCV13 (plus 6C)	73% (55 to 84)	97%	1·19 (0·97 to 1·64)	..
Extra serotypes in PCV13 (plus 6C), excluding 3	80% (65 to 89)	97%	1·04 (0·68 to 1·42)	..
All PCV7 serotypes	90% (34 to 98)	98%	0·59 (0·34 to 2·45)	..
All PCV13 serotypes, (plus 6C)	75% (58 to 84)	97%	0·98 (0·77 to 1·25)	..
All PCV13 serotypes (plus 6C), excluding 3	82% (68 to 89)	97%	0·81 (0·70 to 1·10)	..
PCV7				
4	97% (65 to 99·8)	98%	0·35 (0·20 to 1·17)	70 (52 to 329)
6B	58% (3 to 82)	31%	0·16 (0·08 to 2·54)	97 (4 to 1003)
9V	70% (–25 to 93)	86%	0·62 (0·19 to ∞)	201 (4 to ∞)
14	98% (88 to 99·5)	98%	0·46 (0·25 to 1·12)	4 (4 to 92)
18C	96% (81 to 99)	83%	0·14 (0·09 to 0·40)	4 (4 to 284)
19F	75% (37 to 90)	96%	1·17 (0·62 to 4·62)	430 (260 to 909)
23F	78% (23 to 94)	60%	0·20 (0·08 to 1·50)	231 (4 to 890)
All PCV7 serotypes	82% (72 to 89)	93%	0·63 (0·51 to 0·79)	..

Calculated correlates of protection are based on the estimates of vaccine effectiveness estimates for the measure defined as at least two doses given before age 12 months or one dose given on or after age 12 months. PCV=pneumococcal conjugate vaccine. *Calculations based on the proportion above the cutoff from 1 month post-second dose samples from studies of PCV13 and PCV7. †Includes cases typed as 6A/C.

Table 3: **Predicted vaccine effectiveness at the 0·35 µg/mL correlate of protection and calculated correlates of protection for ELISA and opsonophagocytic antibody assays**

We also derived serotype-specific correlates of protection for functional antibody (table 3). For serotypes 1, 6A, 14, and 18C the point estimate for the correlate was below the lower limit of the assay and was thus expressed as a titre of 4, although for some results the CIs were wide. For the other nine serotypes the point estimates varied widely, ranging from 39 (serotype 3) to 769 (serotype 7F). Absolute titres between serotypes should be compared with caution because the sensitivity of each serotype to killing in each assay varies. With such wide variation between the correlates for different serotypes, a single aggregate correlate would be unlikely to have any biological plausibility, thus we made no attempt to derive one for functional antibody.

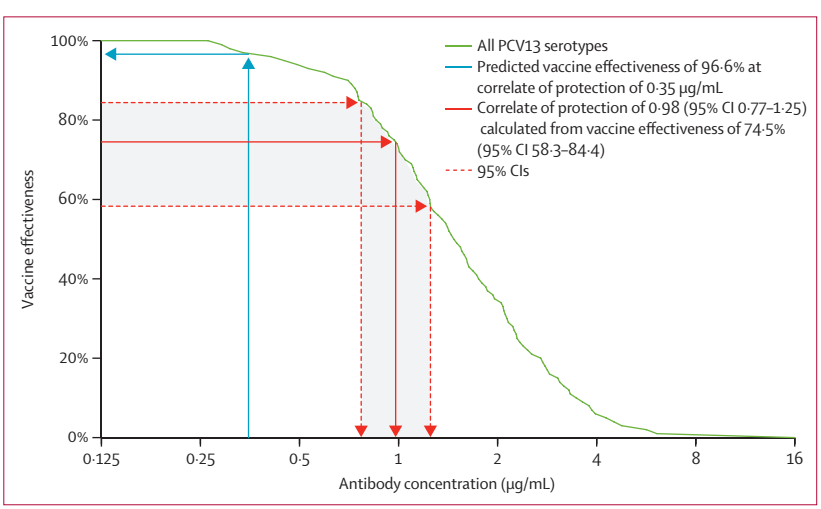

Figure: **Reverse cumulative distribution curve for the geometric mean of all PCV13 serotype-specific IgG from the PCV13 serology study (EudraCT 2010-023865-22/NCT01425372)**
Predicted vaccine effectiveness at a correlate of 0·35 µg/mL and observed vaccine effectiveness against all serotypes for individuals given at least two doses of vaccine before age 12 months or one dose on or after 12 months onwards.

Discussion

The findings of our postlicensure indirect cohort study of the use of PCV13 over 3·5 years in England, Wales, and Northern Ireland show generally good vaccine effectiveness for four of the six new serotypes in PCV13 and high vaccine effectiveness against the PCV7 serotypes (panel). Of particular importance is evidence of significant protection against serotypes 7F and 19A, which have been major causes of replacement disease,[2,3] and protection against serotype 1, which is an important cause of invasive pneumococcal disease in developing countries and for which existing clinical trial data have been contradictory.[18,19]

For serotype 6A, the estimate for vaccine effectiveness of 98% (95% CI 64–99·8) for PCV13 was higher than our

Panel: Research in context

Systematic review

We searched PubMed for articles published in English between Jan 1, 2000, and May 30, 2014, that included the terms "*Streptococcus pneumonia*", "pneumococcus", "pneumococcal vaccine", or "conjugate vaccine", and the term "correlates of protection". We assessed for relevance any studies of pneumococcal vaccines in human beings that sought to derive or validate a correlate of protection. We identified no studies that address the validity of the single existing aggregate correlate of protection applied to the different serotypes in the seven-valent pneumococcal conjugate vaccine (PCV7) and higher valency vaccines when used routinely in a national immunisation programme, although results from one postlicensure study[11] in the UK suggested a mismatch between efficacy for serotype 6B predicted from immunogenicity data and observed protection.

Interpretation

Our study combines postlicensure estimates of serotype-specific vaccine effectiveness for the 13-valent pneumococcal conjugate vaccine (PCV13) with immunogenicity data, both generated in the target infant population in England, Wales, and Northern Ireland. We used the vaccine effectiveness of PCV13 after its introduction into routine infant immunisation to critically assess the original aggregate correlate of protection. Our results show that there are significant differences between the correlates of protection for the different vaccine serotypes such that the aggregate correlate is imprecise. Our results also suggest that no single aggregate correlate of protection based on functional assays will be useful because significant differences exist between the serotypes. Our findings will raise questions about the most appropriate correlates to use for the licensing of the next generation of vaccines.

previous estimate for the cross-protection induced by the 6B component of PCV7 (31%, 95% CI –117 to 78), although cross-protection against 6C, which we anticipated because of the inclusion of 6A in PCV13,[20] could not be estimated because of small numbers. For serotype 3, which was removed from the 11-valent precursor of PCV10 after an otitis media trial did not show serotype-specific efficacy,[21] the point estimate suggested some protection against invasive pneumococcal disease, although 95% CIs were wide and crossed zero. No cases of serotype 5 were recorded during the surveillance period, precluding any analysis. We were unable to obtain a precise estimate of vaccine effectiveness in children with comorbidities; however, protection was significantly lower than in healthy children. Children with comorbidities were infected with non-PCV13 serotypes more often than were healthy children, as previously noted.[22]

No other analyses of serotype-specific vaccine effectiveness after the introduction of PCV13 have been reported. However, data presented in an abstract[23] from the US Centers for Disease Control and Prevention (CDC) show estimates of vaccine effectiveness that are higher than those that we have derived for some serotypes. However, these data, and the CDC's previous estimates of vaccine effectiveness for PCV7,[1] which were also higher than those in the UK, were obtained by use of a conventional case-control design with an attempt to adjust for potential cofounders that were regarded as risk factors for being diagnosed with invasive pneumococcal disease. Since the CDC PCV7 study[1] showed significant protection against non-PCV7 serotypes in children with comorbidities, vaccine effectiveness might well have been overestimated because of residual confounding. The test-negative design we have used has the advantage of well matched controls, but can overestimate vaccine effectiveness because of serotype replacement. However, we have previously shown that this bias should be small (<5 percentage points) in the UK setting.[13] Underestimation of vaccine effectiveness is possible if crossprotection is present, which is why we included 6C with the PCV13 serotypes in our analysis.

The key question that our study addresses is whether the correlate of protection used for licensing PCV13 adequately predicted vaccine effectiveness. If the 0·35 µg/mL correlate of protection threshold correctly predicted the aggregate vaccine effectiveness for the original PCV7 serotypes, then the introduction of PCV13 would have led to 98% protection. In reality, the aggregate effectiveness against these seven serotypes after the introduction of PCV13 was lower (90%, 95% CI 34–98). When we recalculated an aggregate correlate of protection for the PCV7 serotypes (using relevant values for IgG concentration after receipt of PCV13), the correlate of protection was 0·59 µg/mL. This finding suggests that the 0·35 µg/mL correlate for PCV7 serotypes is too low and does not predict vaccine performance with much precision. The pooled estimates of vaccine effectiveness for PCV13 serotypes were also somewhat lower than predicted from immunogenicity data—an aggregate correlate derived for all PCV13 serotypes (plus 6C) would be 0·98 µg/mL. This fairly high correlate of protection was partly caused by the high estimated correlate of protection for serotype 3, for which the vaccine had the lowest effectiveness. However, even after removal of this serotype from the estimate of vaccine effectiveness, the aggregate correlate of protection for the remaining PCV13 serotypes was 0·81 µg/mL, still substantially higher than the 0·35 µg/mL threshold.

The aggregate 0·35 µg/mL correlate of protection was an imprecise predictor of the probable efficacy of individual serotypes. Although the 95% CIs for the estimates of vaccine effectiveness were wide, if an IgG concentration of 0·35 µg/mL after the priming dose were a true correlate of serotype-specific protection then PCV13 should have been more effective against serotypes 1, 3, 7F, 9V, 19A, and 19F than our results showed. For these serotypes, calculated correlates of protection were all substantially higher than 0·35 µg/mL. For serotype 3, for example, a much higher

serum IgG concentration (2·83 μg/mL) would be needed for protection. Such a high concentration is rarely attained from vaccination, which accounts for the poor efficacy against otitis media in a recent clinical trial.[21] The fairly high correlate of protection for serotype 19F (1·17 μg/mL) is biologically plausible, since in-vitro data show that high IgG concentrations are needed to achieve complement deposition and killing for this serotype.[24] By contrast, the estimated correlates of protection for serotypes 6A, 6B, 18C, and 23F were low, in the range of 0·14–0·20 μg/mL, suggesting that less antibody is needed to protect against invasive pneumococcal disease caused by these serotypes. These findings are consistent with our previously reported estimates of correlates of protection for serotype 6B.[11]

Functional correlates of protection, assessed with the pneumococcal opsonophagocytic killing assay,[14] have not been well defined, but are crucial, since more emphasis is now being placed on opsonophagocytic antibodies for the assessment of new pneumococcal vaccines.[25] When the original ELISA correlates were reported, researchers noted that an opsonic antibody titre of 1 in 8 accorded with the IgG correlate of protection, an assertion that seems to have gained some acceptance.[5] However, this titre, which was derived from the validated correlate of protection for *Neisseria meningitidis* serogroup C conjugate vaccines, has not itself been validated.[26] The results of our study suggest that no single threshold can be applied for all PCV13 serotypes, because the performance of each individual serotype-specific functional assay varies. 13 different bacterial isolates are used for the serotype-specific opsonophagocytic antibody assays and each has different sensitivity to being killed. Some isolates (eg, serotype 7F) are very sensitive to killing with many unvaccinated individuals showing high opsonophagocytic antibody titres, leading to high correlates of protection, whereas other isolates (eg, serotypes 1, 14, and 18C) are difficult to kill, resulting in low opsonophagocytic antibody correlates of protection. The differences between the strains (used widely in many laboratories[14]) make it impossible to define a single aggregate correlate as has been done for IgG. Moreover, correlates ranked by IgG will not necessarily match those ranked by functional titres.

The method that we used in this study to derive serotype-specific correlates of protection reproduces that used to derive an aggregate correlate of protection for PCV7 serotypes from the three efficacy trials,[6–8] namely comparison of IgG concentrations 1 month after completion of the primary schedule. The vaccine effectiveness measure combines the periods after primary schedule and after booster. The booster dose of PCV is highly immunogenic, but this immunogenicity is not factored into the derivation of correlates of protection. Notably, despite PCV responses being higher after a booster than after the priming doses, the estimate of vaccine effectiveness after one dose of PCV13 in the second year of life was similar to the vaccine effectiveness after the two-dose primary series. This finding shows that the notion of a threshold concentration of circulating serotype-specific IgG that is protective is overly simplistic.

Despite this conceptual problem, and the difficulty in translating the aggregate correlate of protection of 0·35 μg/mL derived for PCV7 serotypes to individual PCV13 serotypes, the PCV7 correlate of protection has permitted the licensure of vaccines containing highly protective additional serotypes, the efficacy of which had not been shown in trials before licensing. With the potential need for more serotypes to be added to existing vaccines, for which efficacy data will be similarly unavailable, discarding the currently accepted 0·35 μg/mL correlate of protection without a substitute would be unwise. We do not propose that the criteria for licensure for higher valency vaccines should necessarily be changed in view of our findings. New PCVs would still need to be licensed on the basis of non-inferiority in head-to-head studies with existing licensed PCVs, as is the established practice.[4] However, the serotype-specific correlates of protection that we have calculated provide a more accurate prediction of the probable protection afforded by PCV13 for the common serotypes. Expectations about the efficacy of the vaccine against these serotypes must be recalibrated in view of our data. Although continued use of a simplistic serological correlate of protection after primary schedule might be necessary for pragmatic reasons, the biological mechanisms underlying individual protection and induction of herd protection merit further study.

Contributors

EM and PAW were responsible for data collection and data management. DG, PB, and EP generated the ELISA data. DG, LR, and MZ generated the opsonophagocytic antibody data. MS is the microbiological lead for pneumococcal surveillance. SNL is the clinical lead for pneumococcal surveillance and coordinated the PCV13 immunogenicity study. NJA did the statistical analysis and DG, NJA, and EM wrote the report. All authors contributed to the study design and read and approved the final version of the report.

Declaration of interests

DG and MS have served on ad-hoc advisory boards for Pfizer, GlaxoSmithKline, and Merck, and the University College London Institute of Child Health laboratory (London, UK; employer of DG, PB, EP, LR, and MZ) receives contract research funding from Pfizer, GlaxoSmithKline, and Merck. The Public Health England Respiratory and Vaccine Preventable Bacteria Reference Unit, Colindale, London, UK (employer of MS), has received research funding from Pfizer and GlaxoSmithKline. SNL has worked on clinical trials on behalf of St George's University of London (London, UK) for vaccine manufacturers including GlaxoSmithKline and Pfizer, but has received no personal remuneration. EM, PAW, and NJA declare no competing interests.

Acknowledgments

The national surveillance of invasive pneumococcal disease is funded by Public Health England. The field and laboratory work for the immunogenicity study were supported by a grant from the UK Department of Health Research and Development Directorate (grant number 039/0031). The views expressed in this publication are those of the authors and not necessarily those of the UK Department of Health. Public Health England has approval under PIAG Section 60 of the Health and Social Act 2001 (which has been subsumed into the National Information Governance Board for Health and Social Care with Section 60—now Section 251 of the NHS Act 2006) to process confidential information from patients for the purposes of monitoring the efficacy and safety of vaccination programmes. We thank the many general

practitioners, hospital doctors, and laboratory staff in England and Wales who provided information about their patients. We also thank the administrative staff of the Public Health England Immunisation Department for their assistance in the follow-up of invasive pneumococcal disease cases and in the running of the clinical trials, the vaccine research nurses in Hertfordshire and Gloucestershire who recruited, bled, and vaccinated the individuals included in the study, and the technical staff in the Public Health England Respiratory and Vaccine Preventable Bacteria Reference Unit for serotyping the pneumococcal isolates. We also thank staff at the John Radcliffe Hospital Oxford (Oxford, UK) for reporting invasive pneumococcal disease cases in southern England serotyped by their laboratory.

References

1 Whitney CG, Pilishvili T, Farley MM, et al. Effectiveness of seven-valent pneumococcal conjugate vaccine against invasive pneumococcal disease: a matched case-control study. *Lancet* 2006; **368:** 1495–502.

2 Weinberger DM, Malley R, Lipsitch M. Serotype replacement in disease after pneumococcal vaccination. *Lancet* 2011; **378:** 1962–73.

3 Miller E, Andrews NJ, Waight PA, Slack MP, George RC. Herd immunity and serotype replacement 4 years after seven-valent pneumococcal conjugate vaccination in England and Wales: an observational cohort study. *Lancet Infect Dis* 2011; **11:** 760–68.

4 WHO. Recommendations for the production and control of pneumococcal conjugate vaccines. Technical Report Series 927. Geneva: World Health Organization, 2005.

5 Jodar L, Butler J, Carlone G, et al. Serological criteria for evaluation and licensure of new pneumococcal conjugate vaccine formulations for use in infants. *Vaccine* 2003; **21:** 3265–72.

6 Findlow H, Borrow R, Andrews N, et al. Immunogenicity of a single dose of meningococcal group C conjugate vaccine given at 3 months of age to healthy infants in the United Kingdom. *Pediatr Infect Dis J* 2012; **31:** 616–22.

7 O'Brien KL, Moulton LH, Reid R, et al. Efficacy and safety of seven-valent conjugate pneumococcal vaccine in American Indian children: group randomised trial. *Lancet* 2003; **362:** 355–61.

8 Klugman KP, Madhi SA, Huebner RE, Kohberger R, Mbelle N, Pierce N, for the Vaccine Trialists Group. A trial of a 9-valent pneumococcal conjugate vaccine in children with and those without HIV infection. *N Engl J Med* 2003; **349:** 1341–48.

9 Black S, Shinefield H, Fireman B, et al. Efficacy, safety and immunogenicity of heptavalent pneumococcal conjugate vaccine in children. Northern California Kaiser Permanente Vaccine Study Center Group. *Pediatr Infect Dis* J 2000; **19:** 187–95.

10 Jokinen JT, Ahman H, Kilpi TM, Makela PH, Kayhty MH. Concentration of antipneumococcal antibodies as a serological correlate of protection: an application to acute otitis media. *J Infect Dis* 2004; **190:** 545–50.

11 Goldblatt D, Southern J, Ashton L, et al. Immunogenicity of a reduced schedule of pneumococcal conjugate vaccine in healthy infants and correlates of protection for serotype 6B in the United Kingdom. *Pediatr Infect Dis J* 2010; **29:** 401–05.

12 Miller E, Andrews NJ, Waight PA, Slack MP, George RC. Effectiveness of the new serotypes in the 13-valent pneumococcal conjugate vaccine. *Vaccine* 2011; **29:** 9127–31.

13 Andrews N, Waight PA, Borrow R, et al. Using the indirect cohort design to estimate the effectiveness of the seven valent pneumococcal conjugate vaccine in England and Wales. *PLoS One* 2011; **6:** e28435.

14 Rose CE, Romero-Steiner S, Burton RL, et al. Multilaboratory comparison of *Streptococcus pneumoniae* opsonophagocytic killing assays and their level of agreement for the determination of functional antibody activity in human reference sera. *Clin Vaccine Immunol* 2011; **18:** 135–42.

15 Goldblatt D, Southern J, Ashton L, et al. Immunogenicity and boosting after a reduced number of doses of a pneumococcal conjugate vaccine in infants and toddlers. *Pediatr Infect Dis J* 2006; **25:** 312–19.

16 Broome CV, Facklam RR, Fraser DW. Pneumococcal disease after pneumococcal vaccination: an alternative method to estimate the efficacy of pneumococcal vaccine. *N Engl J Med* 1980; **303:** 549–52.

17 Siber GR, Chang I, Baker S, et al. Estimating the protective concentration of anti-pneumococcal capsular polysaccharide antibodies. *Vaccine* 2007; **25:** 3816–26.

18 Cutts FT, Zaman SM, Enwere G, et al. Efficacy of nine-valent pneumococcal conjugate vaccine against pneumonia and invasive pneumococcal disease in The Gambia: randomised, double-blind, placebo-controlled trial. *Lancet* 2005; **365:** 1139–46.

19 Klugman KP, Madhi SA, Huebner RE, Kohberger R, Mbelle N, Pierce N. A trial of a 9-valent pneumococcal conjugate vaccine in children with and those without HIV infection. *N Engl J Med* 2003; **349:** 1341–48.

20 Grant LR, O'Brien SE, Burbidge P, et al. Comparative immunogenicity of 7 and 13-valent pneumococcal conjugate vaccines and the development of functional antibodies to cross-reactive serotypes. *PLoS One* 2013; **8:** e74906.

21 Prymula R, Peeters P, Chrobok V, et al. Pneumococcal capsular polysaccharides conjugated to protein D for prevention of acute otitis media caused by both *Streptococcus pneumoniae* and non-typable *Haemophilus influenzae*: a randomised double-blind efficacy study. *Lancet* 2006; **367:** 740–48.

22 Ladhani SN, Slack MP, Andrews NJ, Waight PA, Borrow R, Miller E. Invasive pneumococcal disease after routine pneumococcal conjugate vaccination in children, England and Wales. *Emerg Infect Dis* 2013; **19:** 61–68.

23 Moore M, Link-Gelles R, Guevera R, et al. Case-control study of effectiveness of 13-valent pneumococcal conjugate vaccine (PCV13) against invasive pneumococcal disease (IPD) in the US: preliminary estimates. International Symposium on Pneumococci and Pneumococcal Disease; Hyderabad, India; March 9–14, 2014. Abstr 0329.

24 Melin M, Jarva H, Siira L, Meri S, Kayhty H, Vakevainen M. *Streptococcus pneumoniae* capsular serotype 19F is more resistant to C3 deposition and less sensitive to opsonophagocytosis than serotype 6B. *Infect Immun* 2009; **77:** 676–84.

25 Feavers I, Griffiths E, Baca-Estrada M, Knezevic I, Zhou T. WHO/Health Canada meeting on regulatory considerations for evaluation and licensing of new meningococcal Group B vaccines, Ottawa, Canada, 3–4 October 2011. *Biologicals* 2012; **40:** 507–16.

26 Andrews N, Borrow R, Miller E. Validation of serological correlate of protection for meningococcal C conjugate vaccine by using efficacy estimates from postlicensure surveillance in England. *Clin Diagn Lab Immunol* 2003; **10:** 780–86.