ANTIBODY PERSISTENCE AFTER PRIMARY AND BOOSTER DOSES OF A PENTAVALENT VACCINE AGAINST DIPHTHERIA, TETANUS, ACELLULAR PERTUSSIS, INACTIVATED POLIOVIRUS, *HAEMOPHILUS INFLUENZAE* TYPE B VACCINE AMONG THAI CHILDREN AT 18-19 MONTHS OF AGE

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Abstract. The World Health Organization recommends a booster dose of a pertussis-containing vaccine for children aged 1-6 years, preferably during the second year of life. This study assessed the immunogenicity and safety of a pentavalent combination vaccine containing diphtheria, tetanus, acellular pertussis, inactivated poliovirus, and conjugated-Hib polysaccharide antigens, [(DTaP-IPV//PRP~T (Pentaxim®)], as a booster at 18-19 months of age. Participants had received primary doses of the same vaccine at 2, 4 and 6 months of age. Antibody concentrations were measured immediately before and one month after the booster dose. Adverse events were evaluated from parental reports. Geometric mean concentrations (GMCs) or titers (GMTs) decreased from post-primary to pre-booster vaccination; however, at least 94.4% of children had protective levels of anti-tetanus (≥0.01 IU/ml), anti-poliovirus (≥8 1/dil) and anti-PRP (Hib, ≥0.15 µg/ml) antibodies prior to the booster. Anti-diphtheria antibody titers ≥0.01 IU/ml were also observed in the majority of children pre-booster. One month after the booster, seroprotection rates were 99.4% for PRP (≥1.0 µg/ml), 95.0% for diphtheria (≥0.10 IU/ml) and 100% for tetanus (≥0.1 IU/ml) and poliovirus types 1, 2, 3 (≥8 1/dil). At least 93.1% of subjects had 4 fold post-booster increases in anti-pertussis antibody titers. GMCs increased from 14.0 to 307.3 EU/ml and from 13.9 to 271.9 EU/ml for anti-PT and anti-FHA, respectively. Anti-PRP GMC increased from 1.2 to 62.2 µg/ml. The booster was well tolerated. A booster dose during the second year of life was safe and induced a strong immune response, indicative of long-term protection.

Keywords: pentavalent combined vaccine, acellular pertussis, booster vaccination, inactivated polio vaccine, Hib-conjugate vaccine, safety, immunogenicity

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INTRODUCTION

The Expanded Program of Immunuzation (EPI) originally included vaccinations against six diseases: tuberculosis, diphtheria, neonatal tetanus, whooping

cough, poliomyelitis and measles. Since then, other vaccines have been added including hepatitis B (Hep B) (WHO, 1992) and *Haemophilus influenzae* type b (Hib) (WHO, 1998). In Thailand, a diphtheria, tetanus and whole-cell pertussis combined vaccine (DTwP) is included in the national immunization schedule at 2, 4 and 6 months of age with boosters at 18-24 months and 5-6 years of age (WHO, 2007). Acellular pertussis vaccines (aP) contain well-characterized, purified, *Bordetella pertussis* antigens, have improved safety compared to wP-containing vaccines, and retain high immunogenicity (Hewlett and Cherry, 1997; Edwards and Decker, 2008). They have been widely adopted because of their reduced side-effect profile compared to wP vaccines, and the WHO now recommends aP vaccines for national childhood immunization programs for either the booster dose only or for the entire vaccination series (WHO, 2010a). According to the WHO, all available aP-containing vaccines have demonstrated high effectiveness in preventing pertussis (WHO, 2005, 2010a).

Continuing reductions in the incidence of childhood infections are major public health goals that can be achieved only with high vaccination coverage in target groups. Combination vaccines, typically including aP, inactivated poliovirus vaccine (IPV), *Haemophilus influenzae* type b and/or hepatitis B antigens in a single injection have been widely implemented over the past 10 years, and are used in the national immunization programs in North America, most European and a number of Asian countries, Mexico, Turkey, South Africa, Australia and New Zealand (WHO, 2010b). Combination vaccines reduce the number of injections required, and generally the overall cost is lower than administering each vaccine separately. The IPV in combination vaccines contains killed virus, eliminating the risk of vaccine associated paralytic poliomyelitis (VAPP) and polio outbreaks caused by circulation of oral polio vaccine-derived viruses (cVDPV).

Because the duration of protection against common childhood infectious diseases, such as pertussis and Hib, wanes over time; further reductions in the incidence of these diseases worldwide require additional doses of vaccine (WHO, 1997). Booster vaccinations against childhood diseases are recommended and practiced in many countries. A booster given during the second year of life at least 6 months after completing the primary series is expected to protect against pertussis for at least 6 years (WHO, 2010a).

Sanofi Pasteur has developed a liquid DTaP-IPV combination vaccine used to reconstitute lyophilized *Haemophilus influenzae* type b (Hib), polyribosyl ribitol phosphate capsular polysaccharide conjugated to tetanus protein (PRP~T) just before administration. This DTaP-IPV//PRP~T vaccine is licensed as Pentaxim® or Pentavac® in more than 100 countries worldwide, including Thailand. The IPV and Hib vaccines are licensed as Imovax® Polio and ActHIB®, respectively, and are WHO pre-qualified stand-alone vaccines (WHO, 2010c). The approved indication for the combined vaccine is as a three-dose primary series during the first year of life and/or booster vaccination during the second year of life.

The safety and immunogenicity of the DTaP-IPV//PRP~T vaccine has been assessed in numerous clinical studies (Carlsson *et al*, 1998; Lagos *et al*, 1998; Kanra *et al*, 2000; Mallet *et al*, 2000; Carlsson *et al*, 2002; Capeding *et al*, 2008; Dutta *et al*, 2009; Thisyakorn *et al*, 2010). More than 2,000

infants received this vaccine as a three-dose primary vaccination in trials initiated by Sanofi Pasteur (Vidor and Plotkin, 2008). We report here antibody persistence and the results of booster vaccination with the DTaP-IPV//PRP~T vaccine at 18-19 months of age in a group of Thai children who had previously received a primary vaccination series with the same vaccine at 2, 4 and 6 months of age (Thisyakorn *et al*, 2010).

MATERIALS AND METHODS

Study design and subjects

This open, Phase IV study was conducted at King Chulalongkorn Memorial Hospital and Queen Sirikit National Institute of Child Health, Bangkok, Thailand. The ethical review board at each center approved the protocol. The study was conducted in compliance with Good Clinical Practice (GCP) and local regulations (Guidelines for Good Practice in the Conduct of Clinical Trials in Human Participants in Thailand). Written informed consent was obtained from parents or legal representatives before participation.

Healthy full-term (>37 weeks estimated gestational age) infants weighing ≥2.5 kg at birth had previously been enrolled and given the DTaP-IPV//PRP~T vaccine at 2, 4, 6 months of age. They had also received a hepatitis B vaccine at birth, 2 and 6 months of age according to the Thai national immunization schedule. Subjects who completed the primary series were eligible to receive a booster dose of the same vaccine at 18-19 months of age. The objectives of the booster phase of the study were to measure antibody persistence prior to booster administration and to assess seroprotection (SP) rates (diphtheria, tetanus, poliovirus, PRP), seroconversion (SC) rates (PT, FHA) and safety of the DTaP-IPV//PRP~T combined vaccine before and 1 month after the booster dose.

Vaccine

The combined vaccine (Pentaxim®, batch A2094-1) was produced and supplied by Sanofi Pasteur, Lyon, France. Its composition is described elsewhere (Thisyakorn *et al*, 2010). The lyophilized PRP~T component was reconstituted with the liquid DTaP–IPV vaccine immediately before intramuscular injection (0.5 ml) into the anterior upper right thigh.

Serology

Blood samples for antibody determination were taken just before and 1 month after the booster dose. The immunological assays were performed at Sanofi Pasteur's Global Clinical Immunology laboratory in Swiftwater, Pennsylvania, USA. Anti-tetanus toxoid antibody concentrations were measured by enzyme linked immunosorbent assay (ELISA, LLOQ 0.01 IU/ml) and compared to the WHO TE3 human standard. Anti-diphtheria antibody titers were assessed by a micrometabolic inhibition test in Vero cell culture (LLOQ 0.005 IU/ml) and compared to a WHO equine antitoxin standard. Anti-FHA and anti-PT antibody concentrations were measured by ELISA (LLOQ 2 EU/ml) and compared to Sanofi Pasteur reference standards. Anti-poliovirus antibody titers were assessed by microneutralization (LLOQ 4 1/dil) following a modified WHO-standardized procedure (WHO/EPI/GEN 93.9) using Vero cell culture and wild-type polioviruses. Anti-PRP~T antibody concentrations were assessed by a Farr-type RIA method (LLOQ 0.06 µg/ml) and compared to an American Food and Drug Administration (FDA) human reference serum. Anti-HBs antibody concentrations were measured

by quantitative RIA (LLOQ 0.5 mIU/ml, AUSAB®, Abbott Labs, North Chicago, IL). Pre-defined SP rates were: anti-PRP ≥0.15 and 1.0 μg/ml; anti-polio ≥8 (1/dil); anti-diphtheria and anti-tetanus ≥0.01 and 0.1 IU/ml. Seroconversion rates to pertussis antigens were assessed using ≥4-fold increases in antibody concentration from pre- to post-booster vaccination.

Safety

Infants were monitored for 30 minutes after injection for immediate local or systemic reactions. During the week following vaccination, parents or legal guardians recorded the onset, duration and grade of solicited injection site reactions (tenderness, redness and swelling) and systemic reactions [fever (axillary temperature ≥37.4ºC), drowsiness, irritability, abnormal crying, loss of appetite, and vomiting]. Tenderness was severe if the infant cried when the limb was moved or if it prevented normal activity. For erythema and swelling, a diameter <2.5 cm was mild, 2.5 to <5 cm was moderate and >5 cm was severe. Severe crying was defined as continuing for more than 3 hours; severe irritability was described as "inconsolable"; severe loss of appetite required missing ≥3 meals or refusing most food; severe drowsiness was defined as sleeping most of the time or being difficult to wake up; severe fever was defined as an axillary temperature ≥39ºC. Unsolicited adverse events were recorded for 30 days after vaccination. Serious adverse events (SAEs) were assessed and reported throughout the trial.

Statistical analysis

Seroprotection and SC rates were calculated with 95% confidence intervals (CI) using the exact binomial method (Newcombe, 1998a). Geometric mean concentrations (GMCs: anti-PRP, anti-D, anti-T, anti-PT and anti-FHA) and titers (GMTs: anti-polio 1, 2 and 3) were calculated with 95% CIs using the normal approximation (Newcombe, 1998b), and post- *versus* pre-booster GMT or GMC ratios (GMTRs and GMCRs) were calculated. Reverse Cumulative Distribution Curves (RCDCs) were derived for antibody titers before and after booster vaccination.

All subjects who received the booster vaccine and had at least one available safety record were included in the safety analyses. The number and percentage (with 95% CI) of subjects reporting a given symptom after vaccination were calculated. All subjects given the booster vaccination; and with serum obtained according to the protocol were included in immunogenicity analyses. Statistical analysis was descriptive; no hypothesis was tested.

RESULTS

Subjects

A total of 186 infants were enrolled in the primary series vaccination, with 175 received all three primary series doses (Thisyakorn *et al*, 2010). Of these, eight subjects were lost to follow-up, and the remainder (167 subjects) received the booster vaccination. Two of the 167 subjects who were given the booster vaccine were lost to follow-up and two were withdrawn because of protocol violations, leaving 163 subjects who completed the booster vaccination phase. Two subjects who completed the study were excluded from the immunogenicity analysis set (N=161) because of protocol violations; 164 subjects had at least one available safety record and were included in the safety analysis.

Immunogenicity

The seroprotection and SC rates,

Table 1
Seroprotection and seroconversion rates at post-primary, pre-booster and post-booster vaccination.

Criteria	Post-primary	Pre-booster	Post-booster
Anti-PRP ≥0.15 µg/ml	100.0 (97.7-100.0)	94.4 (89.7-97.4)	100.0 (97.7-100.0)
Anti-PRP ≥1.0 µg/ml	96.3 (92.1-98.6)	57.1 (49.1-64.9)	99.4 (96.6-100.0)
Anti-Diphtheria ≥0.01 IU/ml	99.4 (96.8-100.0)	72.7 (65.1-79.4)	100.0 (97.7-100.0)
Anti-Diphtheria ≥0.10 IU/ml	53.8 (46.0-61.40	12.4 (7.8-18.5)	95.0 (90.4-97.8)
Anti-Tetanus ≥0.01 IU/ml	100.0 (97.7-100.0)	100.0 (97.7-100.0)	100.0 (97.7-100.0)
Anti-Tetanus ≥0.10 IU/ml	100.0 (97.7-100.0)	88.8 (82.9-93.2)	100.0 (97.7-100.0)
Anti-Polio 1 ≥8 1/dil	100.0 (97.7-100.0)	97.5 (93.8-99.3)	100.0 (97.7-100.0)
Anti-Polio 2 ≥8 1/dil	100.0 (97.7-100.0)	99.4 (96.6-100.0)	100.0 (97.7-100.0)
Anti-Polio 3 ≥8 1/dil	100.0 (97.7-100.0)	95.0 (90.4-97.8)	100.0 (97.7-100.0)
Anti-PT ≥4-fold increase	94.9 (90.3-97.8)[a]	NA	96.3 (92.1-98.6)[b]
Anti-FHA ≥4-fold increase	93.8 (88.8-97.0)[a]	NA	93.1 (88.0-96.5)[b]

Data are % (95% CI); NA, not applicable; [a]Increase from pre-primary; [b]Increase from pre-booster

Table 2
Geometric mean concentrations (GMCs) or titers (GMTs) for each antigen at 1 month post-primary, pre-booster and post-booster vaccination and post- to pre-booster ratios (GMCRs or GMTRs).

	Post-primary	Pre-booster	Post-booster	
	GMC/GMT[a] (95% CI)	GMC/GMT[a] (95% CI)	GMC/GMT[a] (95% CI)	GMCR/GMTR[b] (95% CI)
Anti-PRP µg/ml	9.62 (7.91-11.70)	1.21 (0.98-1.50)	62.23 (52.81-73.33)	51.23
Anti-Diphtheria IU/ml	0.12 (0.10-0.14)	0.02 (0.02-0.03)	2.67 (0.10-0.14)	128.55
Anti-Tetanus IU/ml	1.13 (1.03-1.23)	0.30 (0.26-0.35)	9.99 (8.97-11.13)	33.15
Anti-Polio 1 (1/dil)	1,267.23 (1,033.98-1,553.09)	166.46 (130.17-212.87)	4,620.75 (3,901.03-5,473.27)	27.76
Anti-Polio 2 (1/dil)	1,602.34 (1,311.06-1,958.34)	250.01 (198.31-315.18)	6,086.64 (5,179.62-7,152.49)	24.35
Anti-Polio 3 (1/dil)	3,078.57 (2,478.53-3,823.88)	156.07 (121.15-201.06)	5,596.51 (4,598.79-6,810.69)	35.86
Anti-PT EU/ml	181.49 (163.05-201.01)	14.01 (11.98-16.37)	307.35 (281.01-336.16)	21.95
Anti-FHA EU/ml	119.13 (108.12-131.26)	13.94 (11.70-16.60)	271.86 (246.91-299.32)	19.54

[a]GMT for anti-polio antibodies; [b]GMTR for anti-polio antibodies

GMTs and GMCs are summarized in Tables 1 and 2. At 7 months of age, 1 month after completing the primary series, 99.4 to 100% of subjects had seroprotection against diphtheria and tetanus (≥0.01 IU/ml), polio (≥8 1/dil) and *Haemophilus influenzae* type b (anti-PRP ≥0.15 µg/ml) and four-fold increases in anti-PT and FHA antibody concentrations were observed in 94.9 and 93.8% of subjects, respectively. At 18-19 months of age, approximately 1 year after priming, at least 94.4% of the children still had protective levels of antibodies to tetanus (≥0.01 IU/ml), the three poliovirus types (≥8 1/dil), and *Haemophilus influenzae* type b (PRP, ≥0.15 µg/ml). Protective anti-diphtheria antibody titers (≥0.01) were also observed in the majority of the children (72.7%). As expected, GMCs and GMTs decreased between completion of the primary series and booster vaccinations.

One month after the booster, seroprotection rates were 99.4% for PRP (≥1.0 mg/ml), 95.0% for diphtheria (≥0.10 IU/ml) and 100% for tetanus (≥0.10 IU/ml) and poliovirus types 1, 2, 3 (≥8 1/dil). Thresholds of 1.0 µg/ml for PRP and 0.1 IU/ml for diphtheria and tetanus are classically considered to indicate longer-term protection more applicable to a booster response than the thresholds that are used as correlates of seroprotection (0.15 µg/ml and 0.01 IU/ml, respectively) following primary vaccination (Plotkin, 2010; Plotkin *et al*, 2011). A ≥4-fold increase in antibody titer against pertussis (PT, FHA) antigens occurred in at least 93.1% of subjects. GMCs increased strongly following booster administration; from 14.01 to 307.35 EU/ml and from 13.94 to 271.86 EU/ml for anti-PT and anti-FHA, respectively. Similar increases in GMC were observed for diphtheria, tetanus and PRP, and in GMTs for polio antibodies (Table 2). Overall, GMTRs and GMCRs ranged from 19.54 for anti-FHA to 128.55 for anti-diphtheria (Table 2). Reverse cumulative distribution curves (RCDCs) show strong, linear increases in antibody titers/concentrations for anti-PT, anti FHA, anti-PRP, and anti-poliovirus (Fig 1) (pre-primary baseline concentrations are included for PT and FHA since seroprotective antibody levels have not been established).

Safety

After the booster vaccination, 128 (78%) subjects reported at least one solicited reaction; 105 (64%) had at least one injection site reaction and 88 (53.7%) had at least one systemic reaction. The most common injection site reaction reported after booster injection was tenderness (55.5% of subjects), and the most common systemic reactions reported were abnormal crying (34.1% of subjects), irritability, and fever (Table 3). Most reactions were mild-to-moderate, occurred within 3 days of vaccination, and resolved without treatment. Severe reactions were infrequent. Overall, six subjects (3.7%) reported a severe injection site reaction and seven (4.3%) reported a severe solicited systemic reaction.

Unsolicited events were reported by 62 subjects (37.8%). The most frequent were upper respiratory tract infections in 30 subjects, pharyngitis in 11 subjects, and pyrexia in 8 subjects. One subject experienced an injection site reaction that was considered by the investigator to be related to the vaccination - a bruise of mild severity. Three SAEs occurred, none of which was determined to be vaccination-related: an asthma attack 1 day post-booster, viral pneumonia, and febrile convulsions (both 10 days post-booster). No deaths occurred during the study.

Table 3
Incidence of solicited local and systemic reactions within 8 days after a booster dose of DTaP-IPV//PRP~T combined vaccine given at 18-19 months of age.

		DTaP-IPV//PRP~T *N*=164	
		n	% (95% CI)
Local reactions:			
Tenderness	Any	91	55.5 (47.5-63.2)
	Severe	5	3.0 (1.3-6.9)
Redness	Any	60	36.6 (29.2-44.5)
	Severe	2	1.2 (0.3-4.3)
Swelling	Any	36	22.0 (15.9-29.1)
	Severe	2	1.2 (0.3-4.3)
Systemic reactions:			
Fever	Any	42	25.6 (19.1-33.0)
	Severe	3	1.8 (0.6-5.2)
Vomiting	Any	23	14.0 (9.1-20.3)
	Severe	1	0.6 (0.1-3.3)
Abnormal crying	Any	56	34.1 (26.9-41.9)
	Severe	4	2.4 (0.9-6.1)
Drowsiness	Any	24	14.6 (9.6-21.0)
	Severe	1	0.6 (0.1-3.3)
Loss of appetite	Any	36	22.0 (15.9-29.1)
	Severe	4	2.4 (0.9-6.1)
Irritability	Any	48	29.3 (22.4-36.9)
	Severe	4	2.4 (0.9-6.1)

N, number of injected subjects with available safety data; *n*, number of subjects with a specific adverse event; %, percentage of subjects with a specific adverse event.

DISCUSSION

Booster vaccinations during the second year of life are recommended in many countries, including Thailand, with the aim to further reduce the burden of childhood infectious diseases. This study evaluated the immunogenicity and safety of a DTaP-IPV//PRP~T combination vaccine booster at 18-19 months of age. All study subjects had completed a primary series with the same vaccine at 2, 4 and 6 months of age, and the immunogenicity and safety of the primary vaccination was consistent with previous studies of this vaccine using different three-dose primary vaccination schedules (Thisyakorn *et al*, 2010).

The increases in antibody titers and concentrations observed after the booster dose, and the high SP rates against all vaccine antigens, indicate strong anamnestic immune responses and imply long-term protection. The booster responses seen here are comparable to other reports in which the study vaccine was administered during the second year of life to children

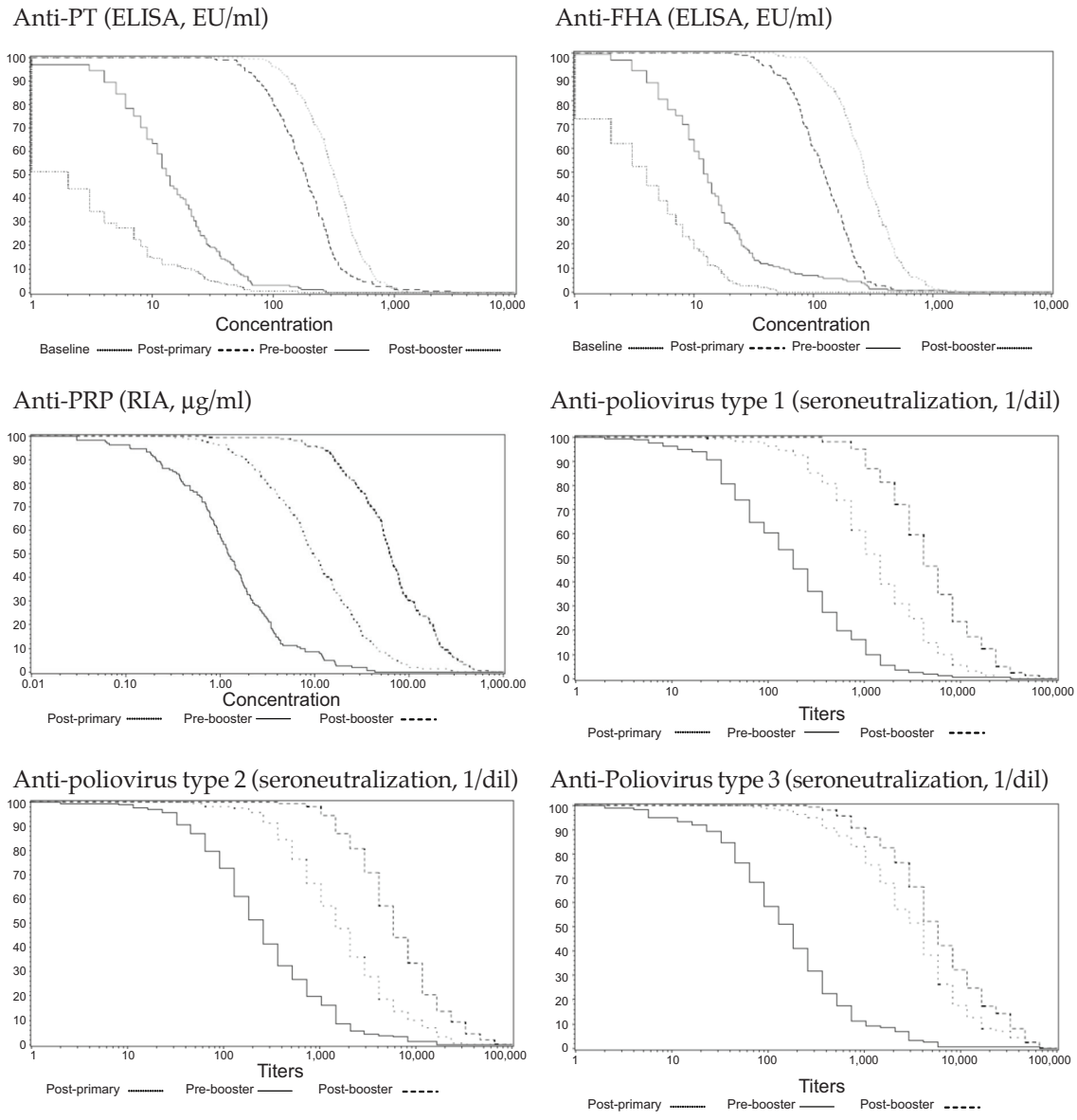

Fig 1–Reverse cumulative distribution curves for anti-PT, anti-FHA, anti-PRP, and anti-polio antibodies, including baseline (pre-primary for anti-PT and -FHA only), post primary, pre- and post-booster antibody levels

who had been primed with the same or other DTaP- or DTwP-based combination vaccines (Carlsson *et al*, 1998, 2002; Mallet *et al*, 2000; Langue *et al*, 2004; Dutta *et al*, 2009; Thisyakorn *et al*, 2009; Li *et al*, 2011; Madhi *et al*, 2011a).

Although the GMTs and GMCs decreased during the year between the primary series and booster administration, antibody persistence was high. At least 94.4% of the children still had protective levels of antibodies to tetanus, the three

poliovirus types and PRP prior to booster vaccination. Anti-diphtheria antibody concentrations were also observed in the majority of children prior to the booster, although the SP rates were lower than for the other antigens and lower than expected based on results of previous trials of the study vaccine (Plotkin *et al*, 2011). Similar persistence of anti-diphtheria antibodies following primary vaccination has however been observed with other DTaP-combined vaccines (Tiru *et al*, 2000; Tichmann *et al*, 2006).

The waning of serum antibody responses to pertussis antigens following primary vaccination is well documented (Grimprel *et al*, 1996; Guiso *et al*, 2007; Edwards and Decker, 2008) and pertussis outbreaks have occurred in countries with schedules that included a three-dose primary series without subsequent booster vaccinations (Barret *et al*, 2010). The booster vaccination results in a T-cell response that is specific for *B. pertussis* (Ryan *et al*, 2000). Although schedules vary, national surveillance in Sweden, France and Austria shows that routine use of aP vaccines provides immunity for at least 6 years if primary vaccination and a booster during the second year of life are given (Bonmarin *et al*, 2007; Rendi-Wagner *et al*, 2007; Carlsson and Trollfors, 2009; Swedish Institute for Infectious Disease Control, 2009). We believe these surveillance data are applicable to the Thai population as the study vaccine had high immunogenicity, similar to previous studies that have included a booster during the second year of life (Mallet *et al*, 2000; Carlsson *et al*, 2002; Langue *et al*, 2004; Thisyakorn *et al*, 2009). The Swedish data show that protection remains high for 5 to 7 years after the second-year booster dose (Gustafsson *et al*, 2006; Swedish Institute for Infectious Disease Control, 2009), suggesting a need for an additional booster dose of aP-containing vaccines in children at about 6 years old. [This is recommended in Thailand (WHO, 2007) and by the WHO (WHO, 2010a)].

Children may also be at risk if a Hib vaccine booster is not given (Kelly *et al*, 2004; Ladhani *et al*, 2010). A fourth (booster) dose of the Hib vaccine is now recommended in the UK, Ireland and Chile, following resurgence of invasive Hib disease among young children when only a primary 3-dose series vaccination schedule was given (Fitzgerald *et al*, 2005; Cameron and Pebody, 2006; Cruces *et al*, 2006; Johnson *et al*, 2006). Our data confirm a strong increase in anti-PRP GMC following booster vaccination.

The high anti-IPV antibody persistence and strong IPV booster response in our study are particularly relevant given the expected cessation of the use of the OPV vaccine and the switch to IPV use in the post-polio eradication era (Rennels, 2009; WHO, 2010d). Athough IPV immunization schedules vary by country, all include 2 to 3 doses during the first year of life and at least one booster dose 6-12 months after completing the primary series. The possibility of exposure to either wild or vaccine-derived poliovirus continues to be a risk worldwide. Booster doses of IPV should be given until long-term antibody persistence is demonstrated without additional doses after the second year of life, or until better worldwide control is achieved. The inclusion of IPV in a DTaP combination vaccine should assure that polio vaccination coverage is as high as that for pertussis and avoid the risk for VAPP or poliomyelitis outbreaks caused by VDPVs.

As expected for DTaP-based vaccines (Eglund *et al*, 1994; Pichichero *et al*, 1997; Edwards and Decker, 2008), the overall

side-effect profile of the study vaccine booster was low. Severe solicited symptoms were reported 3% of the 164 subjects. No hypotonic-hyporesponsive episodes or seizures were reported and no subjects were withdrawn because of vaccination-related AE or SAE.

This study confirms that Pentaxim® administered as a booster at 18-19 months of age is well tolerated and induces high antibody responses to all vaccine antigens. The timing of the booster was appropriate since pre-booster antibody titers were still at satisfactory levels.

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