RESEARCH PAPER

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Booster vaccination of pre-school children with reduced-antigen-content diphtheria-tetanus-acellular pertussis-inactivated poliovirus vaccine co-administered with measles-mumps-rubella-varicella vaccine

A randomized, controlled trial in children primed according to a 2 + 1 schedule in infancy

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Keywords: pre-school, MMRV, diphtheria-tetanus-acellular pertussis-inactivated poliovirus vaccine, Italy, 2 + 1 schedule

Abbreviations: ATP, according to protocol; CI, confidence interval; DTPa-IPV, diphtheria-tetanus-acellular pertussis-inactivated poliovirus vaccine; EL.U/ml, ELISA units per milliliter; FHA, filamentous haemagglutinin; GMC, geometric mean antibody concentration; GMT, geometric mean antibody titer; IPV, inactivated poliovirus vaccine; IU/ml, international units per milliliter; mIU/m, milli-international units per milliliter; MMRV, measles-mumps-rubella-varicella vaccine; PRN, pertactin; PT, pertussis toxoid; SAE, serious adverse event

Background: Pertussis occurs in older children, adolescents and adults due to waning immunity after primary vaccination. Booster vaccination for pre-school children has been recommended in Italy since 1999. In this study (NCT00871000), the immunogenicity, safety and reactogenicity of a booster dose of reduced-antigen content diphtheria-tetanus-acellular pertussis-inactivated poliovirus vaccine (dTpa-IPV; GSK Biologicals *Boostrix*™-Polio; 3-component pertussis) vs. full-strength DTPa-IPV vaccine (sanofi-pasteur—MSD *Tetravac*™; 2-component pertussis) was evaluated in pre-school Italian children.

Methods: Healthy children aged 5–6 y primed in a routine vaccination setting with three doses of DTPa-based vaccines were enrolled and randomized (1:1) in this phase IIIb, booster study to receive a single dose of dTpa-IPV or DTPa-IPV; the MMRV vaccine was co-administered. Antibody concentrations/titers against diphtheria, tetanus, pertussis and poliovirus 1–3 were measured before and one month post-booster. Reactogenicity and safety was assessed.

Results: Three-hundred and five subjects were enrolled of whom 303 (dTpa-IPV = 151; DTPa-IPV = 152) received booster vaccination. One month post-booster, all subjects were seroprotected/seropositive for anti-diphtheria, anti-tetanus, anti-PT, anti-FHA and anti-poliovirus 1–3; 99.3% of dTpa-IPV and 60.4% of DTPa-IPV subjects were seropositive for anti-PRN; 98–100% of subjects were seropositive against MMRV antigens post-booster. Pain at the injection site (dTpa-IPV: 63.6%; DTPa-IPV: 63.2%) and fatigue (dTpa-IPV: 26.5%; DTPa-IPV: 23.7%) were the most commonly reported solicited local and general symptoms, during the 4-d follow-up period. No SAEs or fatalities were reported.

Conclusions: The reduced-antigen-content dTpa-IPV vaccine was non-inferior to full-strength DTPa-IPV vaccine with respect to immunogenicity. The vaccine was well-tolerated and can be confidently used as a booster dose in pre-school children.

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Introduction

Pertussis is a life-threatening childhood disease which accounts for 200,000−400,000 deaths worldwide each year.[1] The introduction of widespread immunization programs against pertussis in the 1940s has led to large reductions in the occurrence of this disease.[2] Primary pertussis vaccination is recommended for infants in Italy according to a 3-5-11 mo schedule (2 primary and 1 booster doses). Booster vaccination for pre-school children has been recommended since 1999.[3] National vaccination coverage against pertussis has increased gradually from below 40% in the early 1990s to approximately 96% in 2009.[3,4] Indeed, the lower reactogenicity associated with acellular pertussis vaccine has helped to increase the acceptance and compliance to vaccination among the general public.[3]

Universal diphtheria-tetanus-pertussis (DTP) vaccine programs led to a decreased incidence of pertussis in infants, but an increase in older children, adolescents and adults has been observed.[3] Pertussis in adults and adolescents not only increases the risk of disease transmission to susceptible infants, but may be an important cause of persistent cough in these populations.[5] Recent outbreaks of pertussis have been recorded in California,[6] Ireland[7] and Australia,[8] where although the majority of cases were observed in infants less than six months old, some were also reported in older individuals. These recent outbreaks of pertussis may be attributable to waning of vaccine-acquired immunity,[9-11] leaving the older population susceptible to the disease and then acting as disease reservoirs for young infants.

Despite complete primary and booster vaccination courses of DTPa, children can become susceptible to pertussis a few years later. As a result, recommendations for a pre-school pertussis booster against diphtheria, tetanus, pertussis and poliomyelitis are widely adopted.

The combined reduced-antigen-content dTpa which contains three-pertussis components and inactivated poliovirus vaccine (dTpa-IPV; *Boostrix*™ Polio, GSK Biologicals; **Table 1**) is indicated for booster immunization in adults, adolescents and children from the age of four years onwards. The safety and immunogenicity of this dTpa-IPV vaccine has already been established.[2,12]

Available data suggest that reduced-antigen-content dTpa vaccines are particularly suitable for boosting pre-school children,[13,14] as repeat doses of full-strength DTPa containing vaccines have been associated with increased reactogenicity.[15] Furthermore, the dTpa vaccine induces high immunogenicity to all antigens,[16] although a previous study in Italy reported that post-booster anti-diphtheria geometric mean concentrations (GMCs) were significantly lower after the reduced-antigen-content dT vaccine compared with the full-strength DT vaccine.[17] Since post-vaccination anti-diphtheria titers relate to duration of immunity, it was assumed at that time that reduced-antigen-content dT vaccines would offer less prolonged protection when compared with full-strength DT vaccines.[14] Consequently, until recently, reduced-antigen content dT vaccines were not recommended in Italy. However, a mathematical model has predicted that following reduced-antigen-content dTpa vaccines, protective levels of anti-diphtheria antibodies comparable to full-strength DTPa can be maintained for 10 y.[14] This is further substantiated by data from long-term follow up studies where persistence of immunity was measured up to 10 y after dTpa booster dose.[16,18,19]

In the present paper the immunogenicity, safety and reactogenicity of a booster dose of reduced-antigen content dTpa-IPV vs. full-strength DTPa-IPV was evaluated in Italian children aged 5−6 y. Since a second dose of measles-mumps-rubella-varicella (MMRV) vaccine is also recommended for pre-school children in Italy,[20] the immune response to MMRV vaccine when co-administered with the booster dose of dTpa-IPV/DTPa-IPV was also assessed.

Results

Demography. A total of 305 subjects were enrolled, 303 of whom were vaccinated with either dTpa-IPV (n = 151) or DTPa-IPV

Table 1. Vaccine composition

Antigens	dTpa-IPV (*Boostrix*™ Polio) (0.5 ml)	DTPa-IPV (*Tetravac*™) (0.5 ml)	MMRV (*Priorix Tetra*™) (0.5 ml)
Diphtheria toxoid	≥ 2 IU	≥ 30 IU	-
Tetanus toxoid	≥ 20 IU	≥ 40 IU	-
Pertussis toxoid	8 μg	25 μg	-
Filamentous Haemagglutinin	8 μg	25 μg	-
Pertactin	2.5 μg	-	-
Poliovirus type 1 (Mahoney strain)	40 D Ag unit	40 D Ag unit	-
Poliovirus type 2 (MEF-1 strain)	8 D Ag unit	8 D Ag unit	-
Poliovirus type 3 (Saukett strain)	32 D Ag unit	32 D Ag unit	-
Measles (Schwarz strain)	-	-	$\geq 10^{3.0}$ $CCID_{50}$ of live attenuated measles virus
Mumps (RIT 4385 strain, derived from Jeryl Lynn strain)	-	-	$\geq 10^{4.4}$ $CCID_{50}$ of live attenuated mumps virus
Rubella (Wistar RA 27/3 strain)	-	-	$\geq 10^{3.0}$ $CCID_{50}$ of live attenuated rubella virus
Varicella (OKA strain)	-	-	$\geq 10^{3.3}$ PFU of live attenuated varicella virus

(n = 152); 285 subjects (dTpa-IPV = 139; DTPa-IPV = 146) were included in the according-to-protocol (ATP) cohort for immunogenicity (**Fig. 1**). The mean age of subjects (total vaccinated cohort) in both groups was 5 ± 0.14 y (range: 4–6 y). The distribution of males (50.8%) and females (49.2%) was similar and the majority (97%) of subjects were white/caucasians.

Immunogenicity. Non-inferiority of dTpa-IPV vs. DTPa-IPV in terms of seroprotection rates for anti-diphtheria, anti-tetanus and anti-polio 1, 2 and 3 one month after booster vaccination was demonstrated in accordance with the pre-defined criteria [upper limit of the standardized asymptotic 95% CI for group difference (DTPa-IPV minus dTpa-IPV) was ≤ 10%; **Table 2**].

Pre-booster serological status. Prior to booster vaccination, the percentage of subjects who remained seroprotected/seropositive was at least 88% for anti-diphtheria, 86% for anti-tetanus, 18% for anti-pertussis toxoid (anti-PT), 92% for anti-filamentous haemagglutinin (anti-FHA), 59.3% for anti-pertactin (anti-PRN) and 95%, 97% and 89% for anti-poliovirus 1, 2 and 3, respectively. Geometric mean concentrations (GMCs) for anti-diphtheria and anti-tetanus at pre-booster time point were at least 0.289 IU/ml in both groups. Anti-PT, anti-FHA and anti-PRN GMCs were not less than 3.5 EL.U/ml, 42.9 EL.U/ml and 7.9 EL.U/ml, respectively. Minimum anti-poliovirus 1, 2 and 3 GMTs were 89.9, 84.5 and 76.1, respectively.

Post-booster immune response. One month after the booster dose, all subjects were seroprotected/seropositive for anti-diphtheria, anti-tetanus, anti-PT, anti-FHA and all three anti-poliovirus antibodies. The composition with regards to the PRN content differs between the two vaccines and therefore, as expected, 99.3% of dTpa-IPV recipients were seropositive for anti-PRN compared with 59.3% (pre-booster level) of DTPa-IPV subjects. In the dTpa-IPV group, the percentage of subjects with anti-diphtheria antibodies ≥ 1.0 IU/ml increased from 14.7% (pre-booster) to 99.3% (post-booster), while the percentage of subjects with anti-tetanus antibodies ≥ 1.0 IU/ml increased from 9.5% (pre-booster) to 98.6% (post-booster).

Booster responses among dTpa-IPV recipients for anti-diphtheria, anti-tetanus, anti-PT, anti-FHA, anti-PRN and anti-polio 1, 2 and 3 were 95.6%, 100%, 89.8%, 94.9%, 94.2%, 82.7%, 81.3% and 91.3%, respectively (**Fig. 2**).

Compared with the pre-booster time point, there was a marked increase in antibody concentration/titers against each of the vaccine antigens one month after the booster dose, in both groups (**Table 3**).

Immunogenicity of MMRV vaccine. At least 90.6% of subjects were seropositive for anti-measles, anti-mumps and anti-rubella antibodies before the booster dose; at least 71.9% of subjects were seropositive for anti-varicella. Seropositivity rates

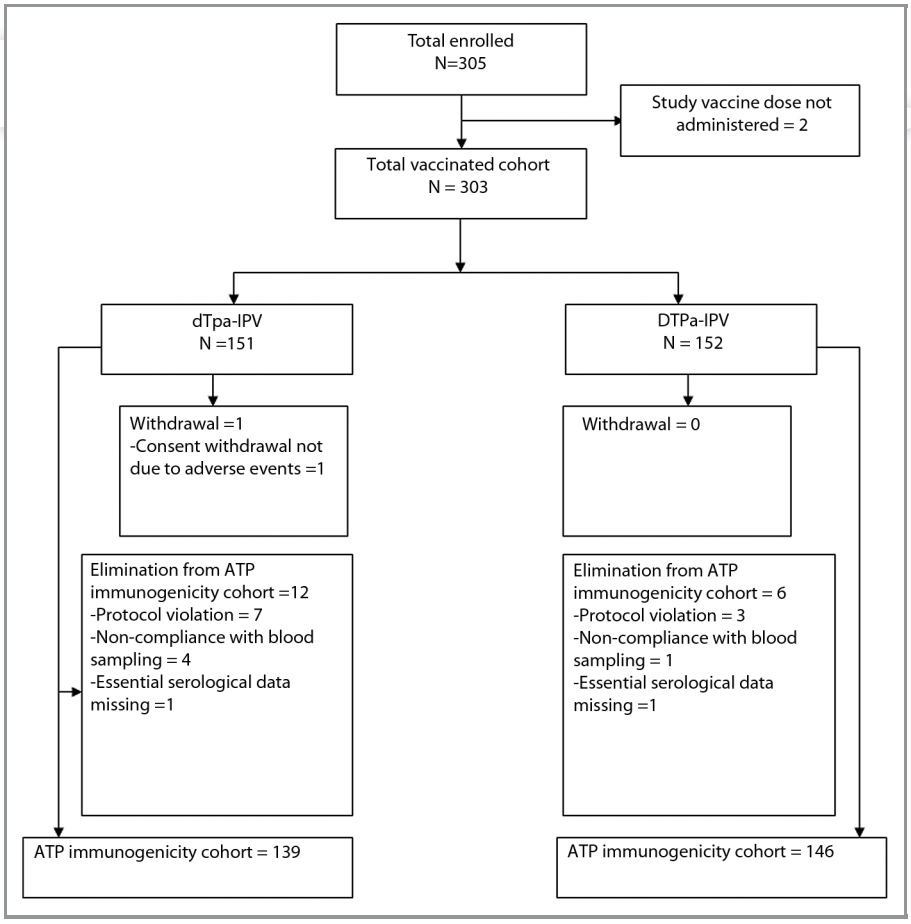

Figure 1. Trial profile.

Table 2. Assessment of non-inferiority: Difference between groups in terms of anti-diphtheria, anti-tetanus and anti-poliovirus types 1, 2 and 3 seroprotection rates one month after the booster dose (ATP cohort for immunogenicity)

	dTpa-IPV		DTPa-IPV		Difference in seroprotection rates (DTPa-IPV Group minus dTpa-IPV Group) [95% CI]
Antibody	**N**	**%**	**N**	**%**	
Anti-diphtheria	139	100	144	100	0.00 [-2.61; 2.70*]
Anti-tetanus	139	100	144	100	0.00 [-2.61; 2.70*]
Anti-poliovirus type 1	139	100	144	100	0.00 [-2.61; 2.70*]
Anti-poliovirus type 2	139	100	144	100	0.00 [-2.61; 2.70*]
Anti-poliovirus type 3	138	100	144	100	0.00 [-2.61; 2.72*]

N, number of subjects with available results; %, percentage of subjects with anti-diphtheria and anti-tetanus antibody concentrations ≥ 0.1 IU/ml and anti-poliovirus types 1, 2 and 3 antibody titers ≥ 8 ED_{50} one month after the booster dose; 95% CI, 95% confidence interval; *non-inferiority criterion met as the UL of the 95% CI was ≤ 10%.

ranged from 95.9–100% against all the MMRV antigens one month post-booster (**Table 4**).

Safety and reactogenicity. During the 4-d follow-up period, symptoms (solicited/unsolicited, local /general) were recorded in 78.8% of dTpa-IPV and 76.3% of DTPa-IPV subjects. Injection site pain and redness (any and Grade 3) were reported by fewer dTpa-IPV than DTPa-IPV recipients (data not shown). Pain at the injection site was the most commonly reported solicited local symptom in both groups (dTpa-IPV: 58.9%; DTPa-IPV: 61.2%; **Fig. 3**). Grade 3 swelling was reported in 5.3% and 3.3% of dTpa-IPV and DTPa-IPV subjects, respectively. Fatigue (dTpa-IPV: 26.5%; DTPa-IPV: 23.7%) and fever (dTpa-IPV: 21.2%; DTPa-IPV: 19.7%) were the most frequently reported solicited general symptoms. Grade 3 general symptoms were infrequent (**Fig. 3**).

At least one unsolicited symptom was reported in 23 dTpa-IPV subjects, [15.2% (95% CI: 9.9–22.0%)] and 20 DTPa-IPV subjects [13.2% (95% CI: 8.2–19.6%)]. Pyrexia was the most frequently recorded unsolicited symptom and occurred in five dTpa-IPV [3.3% (95% CI: 1.1–7.6%)] and four DTPa-IPV [2.6% (95% CI: 0.7–6.6%)] subjects.

There were no SAEs or fatalities reported in this study.

Discussion

The recommended age for a pertussis booster and the choice of either full-strength DTPa vs. reduced-antigen-content dTpa, varies from country-to-country. This paper presents data on the immune response induced by reduced-antigen-content dTpa-IPV vaccine vs. the full-strength DTPa-IPV vaccine when

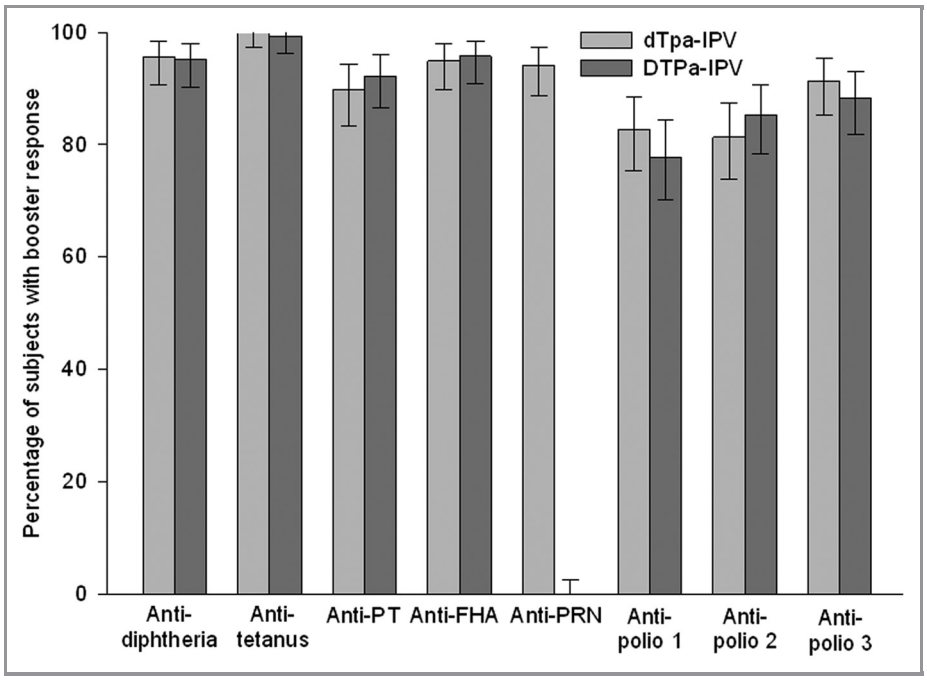

Figure 2. Booster responses one month post-booster vaccination (ATP cohort for immunogenicity).

Table 3. GMC/Ts one month post-booster vaccination (ATP cohort for immunogenicity)

	dTpa-IPV		DTPa-IPV	
	N	GMC/T (95% CI)	N	GMC/T (95% CI)
Anti-Diphtheria	139	9.207 (8.057–10.522)	144	21.393 (19.165–23.880)
Anti-Tetanus	139	12.527 (10.957–14.323)	144	11.070 (9.872–12.413)
Anti-PT	139	59.8 (52.2–68.5)	144	75.9 (65.7–87.7)
Anti-FHA	139	556.2 (491.4–629.5)	144	613.5 (547.0–688.2)
Anti-PRN	139	354.8 (280.2–449.4)	144	7.8 (6.5–9.2)
Anti-Polio1	139	1145.6 (978.7–1340.9)	144	948.0 (817.5–1099.4)
Anti-Polio2	139	1076.4 (908.7–1274.9)	144	1315.3 (1123.1–1540.3)
Anti-Polio3	138	1937.8 (1631.4–2301.8)	144	1657.3 (1385.5–1982.6)

N, number of subjects with available results; GMC/T, geometric mean concentration/titer; 95% CI, 95% confidence interval; dTpa-IPV group, subjects who received a booster dose of reduced-antigen-content dTpa-IPV vaccine co-administered with MMRV vaccine; DTPa-IPV group, Subjects who received a booster dose of full-strength DTPa-IPV vaccine co-administered with MMRV vaccine (this vaccine does not contain the PRN component).

administered as a booster dose in pre-school Italian children. The data demonstrated that the reduced-antigen-content dTpa-IPV vaccine was non-inferior to the full-strength DTPa-IPV vaccine in terms of seroprotection rates against diphtheria, tetanus and poliovirus 1 to 3, one month after booster dosing in pre-school children 5−6 y of age, who were initially primed with DTPa vaccine in a routine setting. A recent study from France has also shown that seroprotection after a reduced diphtheria content dT polio vaccine was non-inferior to the full-strength DT polio vaccine when administered as a booster to children at 6 y of age.[21]

Before booster vaccination, at least 86% of subjects had anti-diphtheria, anti-tetanus, anti-FHA and anti-poliovirus 1 to 3 antibodies above the seroprotective/seropositive cut-off. Approximately 60% of subjects were seropositive for anti-PRN antibodies and seropositivity rates of at least 17.5% were observed for anti-PT.

One month after booster vaccination, all subjects in the dTpa-IPV group were seroprotected or seropositive against diphtheria, tetanus, PT, FHA, PRN and poliovirus 1–3 with the exception of one subject who presented with inadequate PRN. The immune responses to the co-administered MMRV vaccine were unaffected by the co-administration of the dTpa-IPV vaccine. One month after the booster dose, several fold increases (range: 9–43-fold) in GMC/T were observed against diphtheria, tetanus, PT, FHA, PRN, poliovirus 1 to 3 as compared with the pre-booster time point.

As expected, anti-diphtheria GMC was almost twice as high in the DTPa-IPV group compared with the dTpa-IPV group, attributable to the higher amount of diphtheria toxoid in the full-strength DTPa-IPV formulation. However, there is unlikely to be any immediately relevant clinical impact of this observation as antibody levels against diphtheria were well above the correlate of seroprotection of 0.1 IU/ml. Anti-diphtheria seroprotection rates induced by the low-strength dTpa-IPV vaccine persisted up to 10 y post-dTpa booster in long-term follow-up studies involving children, adolescents and adults.[16,18,19]

The overall occurrence of solicited local and general symptoms was similar in both study groups, although local injection site reactions were lower at the dTpa-IPV compared with the DTPa-IPV injection sites. Although a lower reactogenicity would be expected after dTpa vaccine administration due to the reduced-antigen-content, this study was not powered to detect a statistically significant difference between the full-strength DTPa and reduced-antigen-content dTpa vaccines in terms of reactogenicity. Previous studies in different age groups have shown that reductions in the antigen content of diphtheria, tetanus and pertussis vaccines can be associated with fewer injection site reactions.[2,13]

Table 4. Seropositivity rates to the MMRV antigens one month after booster vaccination (ATP cohort for immunogenicity)

	dTpa-IPV			DTPa-IPV		
	N	Seropositivity % (95% CI)	GMC Value (95% CI)	N	Seropositivity % (95% CI)	GMC Value (95% CI)
Anti-Measles ≥ 150 mIU/ml	139	100 (97.4–100)	2743.9 (2411.4–3122.2)	146	100 (97.5–100)	2863 (2534.6–3233.9)
Anti-Mumps ≥ 231 U/ml	139	100 (97.4–100)	4141.3 (3590.5–4776.5)	146	98.6 (95.1–99.8)	3837.6 (3275.1–4496.7)
Anti-Rubella ≥ 4 IU/ml	139	100 (97.4–100)	154.5 (141.3–168.9)	146	99.3 (96.2–100)	162.5 (145.8–181.0)
Anti-varicella ≥ 50 mIU/ml	139	97.1 (92.8–99.2)	856.7 (671.8–1092.4)	146	95.9 (91.3–98.5)	909.9 (721–1148.2)

N, number of subjects with available results; %, percentage of subjects with titer within the specified range; 95% CI = 95% confidence interval; GMC, geometric mean antibody concentration calculated on all subjects; Pre, Pre-vaccination blood sampling time-point; Post, Post-vaccination blood sampling time-point; dTpa-IPV group, Subjects who received a booster dose of reduced-antigen-content dTpa-IPV vaccine co-administered with MMRV vaccine; DTPa-IPV group, subjects who received a booster dose of full-strength DTPa-IPV vaccine co-administered with MMRV vaccine. The pertussis antigen of this vaccine does not contain the PRN component.

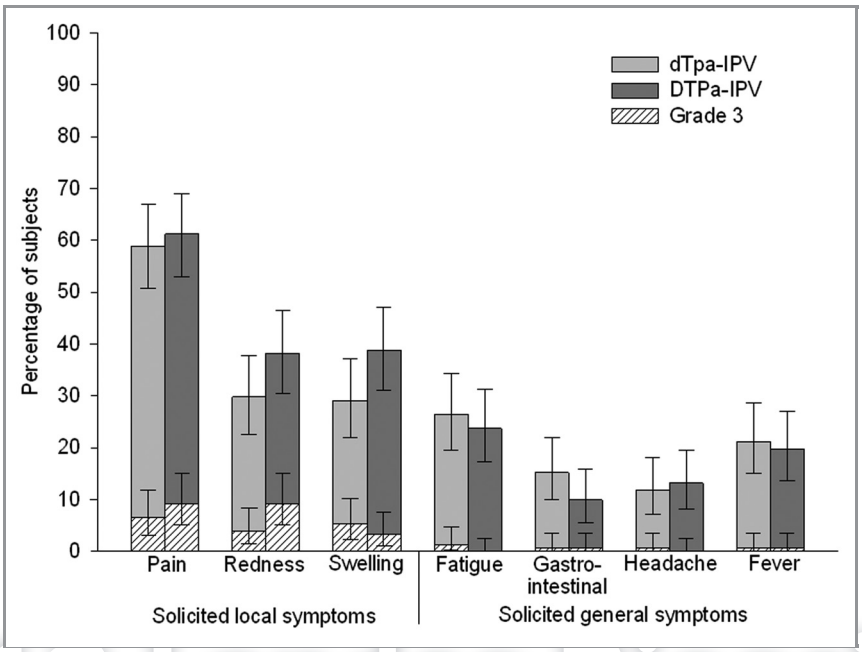

Figure 3. Incidence of any and grade 3 solicited local and general symptoms reported during the 4-d post-booster follow-up period (total vaccinated cohort). Grade 3 symptoms: injection site diameter > 50 mm (redness/swelling); > 39.0°C axillary temperature (fever); prevented normal daily activities (pain, fatigue, gastrointestinal and headache).

A study performed in adolescents in Germany (primed with reduced antigen dTpa-IPV vaccines as pre-school children) demonstrated consistent (98.2%) protection against diphtheria at the pre-adolescent booster timepoint and 100% seroprotection after the booster dose in adolescence.[16] Pertussis antibody titers measured after vaccination with the reduced antigen dTpa-IPV vaccine were higher than those observed in household contact efficacy trials of pertussis prevention.[16,22]

The nature of studies undertaken with subjects who had received previous vaccination in a routine setting introduces uncertainty with respect to the primary vaccination schedules and precise dosage of DTPa vaccines administered. The resulting serology, by design, thus reflects everyday practice but requires caution to be exercised when comparing these persistence data (pre-booster time point) with previously published studies of antibody persistence.

Conclusion

The reduced-antigen-content dTpa-IPV vaccine is non inferior to full-strength DTPa-IPV vaccine in terms of immunogenicity profile and the co-administration of the MMRV vaccine had no negative effect on the immune response. In conclusion, the study contributes to the rationale for using reduced-antigen-content dTpa-IPV vaccines as boosters for pre-school children.

Materials and Methods

Subjects and study design. This phase IIIb, open, multi-center study was conducted in Italy between April and November 2009 (NCT00871000). Healthy Italian children aged 5–6 y, primed with three doses of DTPa-based vaccine in a routine setting were randomized (1:1) to receive a pre-school booster dose with either reduced-antigen-content dTpa-IPV [*Boostrix*™ Polio, GlaxoSmithKline (GSK) Biologicals] or full-strength DTPa-IPV (*Tetravac*™, Sanofi pasteur) vaccines; both were co-administered with MMRV vaccine (*Priorix Tetra*™, GSK Biologicals).

Subjects were excluded if they had received any investigational drug/vaccine in the month preceding booster vaccination, had a clinical history of natural infection against which the study vaccines were targeted or had received previous booster vaccination against diphtheria, tetanus, pertussis or polio.

The study was conducted following Good Clinical Practice guidelines and the Declaration of Helsinki. The study protocol, informed consent and other relevant documents were approved by the independent ethics committee of the study centers. Parents/guardians of children participating in the study provided written informed consent before the start of any study-related procedures.

Vaccines. The reduced-antigen-content dTpa-IPV (*Boostrix*™ Polio) and MMRV (*Priorix Tetra*™) vaccines were manufactured by GSK Biologicals, while the full-strength DTPa-IPV vaccine with two pertussis components (*Tetravac*™) was manufactured by sanofi pasteur-MSD. The compositions of the vaccines are presented in **Table 1**.

Assessment of immunogenicity parameters. Blood samples were collected from all subjects immediately before and one month post-booster (range: 30–48 d). Antibodies against diphtheria, tetanus, PT, FHA, PRN, were measured using in-house enzyme-linked immunosorbent assay (ELISA). Seroprotection was defined as antibodies ≥ 1.0 IU/ml for diphtheria and tetanus;[23] seropositivity was defined as: ≥ 5 EL.U/ml (PT,

FHA and PRN).[24-26] Antibodies against the three poliovirus types were measured using a virus micro-neutralization assay. Seroprotection was defined as ≥ 1:8 dilution for all three poliovirus serotypes.[27] Antibodies against measles, mumps, rubella and varicella were measured using an ELISA kit manufactured by Dade Behring; seropositivity was defined as ≥ 150 mIU/ml (measles); ≥ 231 U/ml (mumps) ≥ 4 IU/ml (rubella); ≥ 50 mIU/ml (varicella). As there is no established correlate of protection against pertussis, a booster response was defined as the appearance of antibodies in initially seronegative subjects (antibodies below the assay cut-off of 5 El.U/ml), or at least a 2–4-fold increase in antibody concentrations in subjects seropositive prior to vaccination.

Assessment of safety and reactogenicity. Local (pain, redness and swelling at injection site) and general (fever, headache, fatigue and gastrointestinal symptoms) symptoms were solicited for four days using diary cards; unsolicited and serious adverse events (SAEs) were recorded for the duration of the study. Solicited symptoms were graded 1–3, where Grade 3 was: injection site diameter > 50 mm (redness/swelling); > 39.0°C axillary temperature (fever); prevented normal daily activities (other symptoms).

Statistical analysis. The statistical analyses were performed using the SAS v9.2 and StatXact-8.1 procedure on SAS. Block randomization (1:1; SAS) using a central internet system facilitated balance between the treatment arms.

The primary endpoint (non-inferiority) on immunogenicity was performed on the ATP cohort, which included subjects who had received a booster dose of the dTpa-IPV/DTPa-IPV vaccine, complied with the protocol and for whom pre-and post-booster blood samples were available.

Reduced-antigen-content dTpa-IPV was non-inferior to full-strength DTPa-IPV if the upper limit of the standardized asymptotic 95% CI on the group difference (DTPa-IPV group minus dTpa-IPV group) in terms of seroprotection rates for anti-diphtheria, anti-tetanus and anti-poliovirus 1, 2 and 3 was ≤ 10% one month after booster vaccination (primary objective). With a sample size of 85 subjects in each group, the study had at least 95% power (Bonferroni adjustment for β) to conclude that the reduced-antigen-content dTpa-IPV was non-inferior to full-strength DTPa-IPV (primary objective). A sample size of 300 subjects was planned for enrolment for descriptive evaluation of safety of the vaccine.

The 95% CI for the seroprotection/seropositivity and geometric mean concentration/titers against each of the vaccine antigens were calculated one month post-booster.

Safety and reactogenicity analysis was performed on the total vaccinated cohort, which included all vaccinated subjects. The percentage of subjects reporting at least one local (solicited/unsolicited), general (solicited/unsolicited) symptom during the 4-d post-vaccination follow-up period, and unsolicited symptoms reported during the 31-d follow-up period was tabulated with exact 95% CI.

Disclosure of Potential Conflicts of Interest

F.M., S.K. and K.H. are employees of GSK Biologicals. M.M. is contracted to GSK Biologicals via Chiltern BV, a contract research organization. *Boostrix Polio* and *Priorix tetra* are trademarks of the GlaxoSmithKline group of companies. *Tetravac* is a trademark of Sanofi pasteur-MSD group of companies.

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