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The seroprevalence, waning rate, and protective duration of anti-diphtheria toxoid IgG antibody in Nha Trang, Vietnam

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ABSTRACT

Background: Diphtheria cases reported in Central Vietnam since 2013 were mainly in children aged 6-15 years, which may reflect an immunity gap. There is little information on population immunity against diphtheria in countries without a school-entry booster dose. We aimed to measure the age-stratified seroprevalence of anti-diphtheria toxoid antibodies, quantify the change in antibody levels in individuals over time, and estimate the length of protective immunity after vaccination in well-vaccinated communities in Vietnam.

Methods: An age-stratified seroprevalence survey among individuals aged 0-55 years was conducted at Nha Trang, Vietnam. The same participants were followed up after two years to quantify the change in antibody levels. IgG was measured using ELISA. The length of protective immunity after vaccination was estimated using a mixed-effect linear regression model with random intercept.

Results: Overall seroprevalence was 26% (95%CI:20-32%). Age-stratified seroprevalence was 68% (95%CI:4-11%), 7% (95%CI:4-11%), 12% (95%CI:7-19%), 33% (95%CI:27-40%), and 28% (95%CI:17-43%) among those aged ≤5, 6-15,16-25, 26-35, and 36-55 years, respectively. The antibody levels declined by 47% (95%CI:31-59%) over two years, and the predicted duration of vaccine-derived protective immunity after receiving four doses was 4.3 years (95%CI:3.5–5.3) among participants aged six years or younger.

Conclusion: Given the low seroprevalence and short period of vaccine protection, a school-entry booster dose (5-7 years) is recommended in Vietnam.

Background

Diphtheria is an acute infectious disease typically affecting the upper respiratory tract, which is mainly caused by toxin-producing *Corynebacterium diphtheriae*. Diphtheria used to be a major cause of child mortality, but introducing a highly effective toxoid vaccine decreased the disease burden. Recently, large-scale outbreaks were observed in Venezuela, Yemen, and the Rohingya population in Bangladesh, where the routine infant vaccination program was disrupted. Diphtheria remains endemic in many parts of the world. The number of cases increased, in particular, in South and South-

Abbreviations: DTP, the diphtheria-tetanus-pertussis; GMC, geometric mean concentration; ELISA, enzyme-linked immunosorbent assay; WHO, the World Health Organization.

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east Asia in the latter half of the 2010s compared with the earlier half (WHO 2017, Clarke et al. 2019).

Currently, WHO recommends that a three-dose primary series of diphtheria toxoid-containing vaccine should be administered in early infancy, a first booster dose in the second year of life, and a second and a third booster at the age of school entry and school leaving, respectively (WHO 2017). Nevertheless, many low-income countries have not introduced any booster dose yet. There is little information on the waning and the duration of immunity among children who receive three or four doses.

In Vietnam, the diphtheria-tetanus-pertussis (DTP) vaccine was introduced in 1981 and was replaced by the Pentavalent vaccine (DTP-Hib-HepB) in 2011 (MOH Vietnam 2012). The primary dose series was given at 2-3-4 months, and in 2012, a booster dose at 18 months was introduced (MOH Vietnam 2012). Although the number of reported cases decreased sharply from 3500 per year in 1983 to nearly zero in 2010 (Jit et al. 2015), several outbreaks have been observed since 2013.

The 76 confirmed diphtheria incidences in the Central and Highland region of Vietnam between 2014 and 2019 were in patients aged between 1 and 55 years, with 66% of the cases being children aged between 6-15 years (data in the Pasteur Institute in Nha Trang and the Institute of Hygiene and Epidemiology of Tay Nguyen, Vietnam). The age distribution in diphtheria cases reflects the pattern of population immunity and, thus, the history of DTP vaccination schedule and coverage (Galazka and Keja 1988, Galazka and Robertson 1995, Zakrzewska et al. 1997, Clarke et al. 2019). The population immunity has changed over time since the introduction of vaccine (Galazka 2000). In addition, it is known that the age at infection in diphtheria cases typically shifts to older ages after the introduction of vaccine (Galazka and Keja 1988). This raises the question of how population immunity against diphtheria is in Vietnam currently.

This study aimed to measure the age-stratified seroprevalence and geometric mean concentration (GMC) of anti-diphtheria toxoid antibodies in a study population in Nha Trang city, Vietnam, where high vaccination coverage was reported in most of the last ten years. The study also quantified the change in anti-diphtheria toxoid antibody levels as a function of age, baseline IgG level, and number of DTP vaccine doses to investigate the antibody waning pattern. Finally, the study estimated the length of time that protective immunity of >0.1 IU/ml was maintained after vaccination.

Materials and methods

Study population and area

Nha Trang city, the capital of Khanh Hoa Province in Central Vietnam, has a population of >300,000 (Yoshida et al. 2014). The two study communes, Vinh Hai and Vinh Phuoc communes, are adjacent to each other in the urban area of Nha Trang. According to a census conducted in 2015, the population size of the two communes was 42,397. Each commune has one health center, which provides essential health services, including vaccination.

Study design and sampling method

A cohort study was conducted between 2017 and 2019. The sample population was originally selected for a cross-sectional seroprevalence survey for antibodies against dengue in June 2017 (Biggs et al. 2020). The original target sample size was 500, and the participants were over-recruited, considering potential non-respondents. Finally, a total of 510 samples were collected in 2017. Age-stratified simple random sampling was conducted in each commune on the basis of census data. The five age strata were 0-5 years, 6-15 years, 16-25 years, 26-35 years, and 36-55 years in 2017, and samples of 50 were drawn randomly from each age stratum from the population data of each of the two communes. These data were available for this study, and all participants in the first cross-sectional survey were invited to join a follow-up survey in May 2019.

Study teams visited participants at their homes. Written informed consent was obtained from each participant or guardian if the participant was younger than 16 years. The survey teams interviewed each participant using a standardized questionnaire collecting information on sex, date of birth, and oral vaccination history. In addition, venous blood samples were collected from participants at the commune health centers. Ethical approval was obtained from the Vietnamese Ministry of Health and the London School of Hygiene and Tropical Medicine ethical review boards (IRB-VN01057-27/2015, LSHTM Ethics ref: 17518/17913).

Vaccination record of individuals and vaccination coverage

We obtained written vaccination records from each individual's vaccination card, in addition to the oral information, during the survey in 2019. If the vaccination card was not available, we searched the vaccination registration book, which recorded individual-based data at the local commune health center. We also collected administrative coverage data from the Department of Preventive Medicine in Nha Trang on the completed three-primary dose series of DTP vaccine (DTP3) and the fourth dose (DTP4) between 2011 to 2017 in the two study communes (Data at the Department of Preventive Medicine, Nha Trang). Local administrative coverage data before 2011 were not accessible. Thus we collected national administrative coverage data between 1983 and 2017 in Vietnam, obtained from the WHO data repository (WHO 2021).

Serological assay

Collected sera were stored at -80°C in the Pasteur Institute in Nha Trang until testing. Serum anti-diphtheria toxoid IgG level was measured using a commercially available ELISA kit (IBL, Germany) following the manufacturer's protocol. An IgG level of > 0.1 IU/ml in ELISA, the international standard cut-off value for the requirement of a booster dose, was considered seropositive (European Center for Disease Prevention and Control 2014, von Hunolstein. C et al. 2014). The seroprevalence was defined as the proportion of seropositive samples in the total number of samples assessed. We excluded the samples that had errors in sample processing or testing.

Statistical Analyses

Age-stratified seroprevalence and geometric mean concentration (GMC)

Age-stratified seroprevalence and GMC were summarized with 95% confidence intervals using the cross-sectional survey data collected in 2017. To do this, seroprevalence and GMC were weighted by population size in the ten age-sex strata in each commune. The sampling weights were the inverse probability of sampling in that age-sex stratum. The difference in weighted seroprevalence and weighted GMC between the sexes was examined by logistic regression and linear regression, respectively, by overall age and each age stratum. Because an initial inspection of data revealed a large difference in immunity between two younger age groups, the seroprevalence and GMC were calculated by each year of age.

Change in anti-diphtheria toxoid IgG antibody levels

The change in anti-diphtheria toxoid IgG level was examined in participants who participated in both the 2017 (IgG1) and 2019

(IgG2) surveys. Owing to the introduction of the fourth dose of the DTP vaccine in the national immunization program in 2012, many participants who were born after 2011, aged six or younger in 2017, had received four doses after this change. On the contrary, participants aged seven years or older in 2017 had received three doses or less. In addition, observation of plotted IgG values showed that the IgG level declined most when participants were of the age six or younger, whereas it often increased if participants were older than ten years. We, therefore, stratified age by three groups at 0-6, 7-10, and 11-55 years old for our analysis. We also categorized IgG1 into four groups, >0.1, 0.05-0.1, 0.03-0.05, and ≤0.03 IU/ml determined by observation of plotted IgG values, to quantify the change in IgG by IgG1 level.

Change in IgG level over two years was evaluated using a change in natural log-transform IgG values (log(IgG2) – log(IgG1)). IgG levels were logarithmically transformed in statistical analyses to reduce the skewness of the data. Back-transformation of log-transformed changes gives ratios of geometric means. We, therefore, defined the percentage decrease of IgG as (1 - geometric mean of (IgG2/IgG1)) x 100 percent.

The percentage decrease in geometric mean levels over two years was reported for overall and exposure age groups 0-6, 7-10, and 11-55 years, and for IgG1 levels >0.1, 0.05-0.1, 0.03-0.05, and ≤0.03 IU/ml. The percentage decrease in geometric mean levels among age group 0-6 years was reported according to whether three or four DTP vaccine doses were received before the measurement of IgG1 levels. The number of doses received that were confirmed on written vaccination records was used for stratification. The paired t-test was conducted to examine whether the change in natural log-transformed IgG in each group was statistically significant.

Duration of vaccine-derived protective immunity

The duration of vaccine-derived protective immunity was defined as the time since the last vaccination that IgG level was maintained at >0.1 IU/ml. All available paired log-transformed IgG values (log(IgG1) and log(IgG2)) for children aged six or younger in 2017, who were born after 2011, with a history of three or four doses of DTP vaccine before the measurement of IgG1 levels were included in the analysis. Their vaccination histories were confirmed through written vaccination records. We assumed that the immunity of children aged six or younger was attributable to vaccination.

A mixed-effect linear regression model with random intercept incorporating the between-individual variability was used to analyze antibody decay over time for anti-diphtheria toxoid IgG, whose decay pattern was assumed exponential, as reported in previously published studies (Laird and Ware 1982, Renard et al. 2001, Bates et al. 2015, Hammarlund et al. 2016, Antia et al. 2018). The time at which the geometric mean level of IgG declined to 0.1 IU/ml was estimated from the line of best fit. This predicted time can be considered to correspond to the duration of protective immunity. A 95% confidence interval around this duration of protective immunity was calculated by the Delta method (Oehlert 1992, Rice 1994).

Analyses were conducted in the STATA 15 software (StataCorp 2017). R software was used for data visualization (R Core Team (2020)).

Results

A total of 510 participants aged 0-55 years were recruited to the survey in 2017; 221 participants (43%) were male. A total of 306 participants (61%) were followed up in 2019. A written vaccination record was found for 108 participants aged 0-28 years. Ninety-four percent of the participants aged 0-5 years had written vaccination records, and 71% of the participants had received four doses. The vaccination records of 60% of the participants aged 6-15 years were confirmed, and 54% of the participants had received three doses (Table 1).

Vaccination coverage of DTP3 among participants who were born between 2007 and 2017 (aged 0-10 in 2017) in each year varied between 78% and 100% on the basis of oral information and 33% to 100% on the basis of vaccination records. The local and national administrative DTP3 coverage in the same period was between 92% and 100%, except 54% for the year 2013 when the Pentavalent vaccine was suspended in Vietnam during the investigation of one severe adverse event (WHO 2021).

A total of 510 samples collected in 2017 were included to measure seroprevalence for the five age strata. Overall weighted seroprevalence was 26% (95%CI:20%-32%). In this population, the highest seroprevalence was 68% (95%CI:67%-69%) in age group 0-5 years, and the lowest was 7% (95%CI:4%-11%) in age group 6-15 years. Seroprevalence levels in age groups 16-25, 26-35, 36-55 years were 12% (95%CI:7%-19%), 33% (95%CI:27%-40%), and 28% (95%CI:17%-43%), respectively. There was no statistical difference by sex in overall and age-stratified seroprevalence (Table 2).

Seroprevalence constantly decreased from 85% in participants aged one year to 0% in participants aged nine years. It stayed at 0% until the age of twelve and gradually increased afterward (Figure 1A). The highest GMC was 0.75 IU/ml at age one year, and it declined to the lowest GMC of 0.02 IU/ml at age ten years. The decreasing and increasing trend of GMC over age was similar to that of seroprevalence (Figure 1B).

Among 306 samples collected during the follow-up survey in 2019, two samples had errors in sample processing or testing. Therefore, 304 paired samples were available to examine the change in IgG level between 2017 and 2019. Of 304 participants, 50 had a record of three doses, and 49 had received four doses. None of these individuals received DTP vaccine or was diagnosed with diphtheria between 2017 and 2019.

All paired IgG values were plotted by age and number of DTP doses (Figure 2A).The IgG levels declined rapidly in two years among children aged six or younger. Although no consistent pattern of IgG change was observed in children aged seven or older, there was a particular pattern of IgG changes by IgG1 level (Figure 2B).

The percentage decrease of IgG in two years was calculated in different age groups by number of DTP doses and IgG1 levels. The percentage decrease of IgG was 41% (95%CI:28%,52%) over two years among those aged six or younger. IgG levels did not decline significantly among those who were older than six. The percentage decrease constantly diminished in accordance with the decline in IgG1 level. Among those who were younger than seven years, the percentage decreases of IgG after receiving three and four doses of DTP were 34% (95%CI:-8%,59%) and 47% (95%CI:30%,59%), respectively (Figure 2B and Table 3).

The length of time that the geometric mean of IgG was maintained > 0.1 IU/ml was estimated by a mixed-effect linear regression with a random intercept model. The number of samples included in the analysis was 114 (57 individuals) overall, 22 (11 individuals) of whom received three doses only, and 92 (46 individuals) of whom received four doses. IgG was estimated to be maintained at >0.1 IU/ml for 4.7 years (95%CI:3.67-5.63) when the analysis was conducted combining those who received three or four doses (Figure 3). IgG was estimated to be maintained at >0.1IU/ml for 4.3 years (95%CI:3.46-5.26) after receiving four doses. Because of the limited number of participants who received only three doses, we could not estimate the duration of protective immunity among them accurately.

Table 1
The number of study participants by age group in 2017 and follow-up rate and vaccination history by participants' age group in 2019.
Vaccine record for more than 90% of the participants aged 0-5 were available; 71% received four doses. Sixty percent of participants aged 6-15 years had vaccine records, and 54% had a record of receiving three doses.

		Number of participants				F/U rate	Vaccination history among 306 participants in 2019											
year of birth	age (2017)	in 2017		in 2019			with record	(%)	number of vaccines received								no record	(%)
		n	(%)	n	(%)				4	(%)	3	(%)	2	(%)	1	(%)	0	(%)				
2012-2017	0-5yr	100	20%	63	21%	63%	59	94%	45	71%	9	14%	2	3%	1	2%	2	3%	4	6%
2002-2011	6-15yr	107	21%	65	21%	61%	39	60%	3	5%	35	54%	0	0%	1	2%	0	0%	26	40%
1992-2001	16-25yr	105	21%	52	17%	50%	7	13%	0	0%	6	12%	0	0%	0	0%	1	2%	45	87%
1982-1991	26-35yr	94	18%	59	19%	63%	3	5%	1	2%	1	2%	0	0%	0	0%	1	2%	56	95%
1962-1981	36-55yr	104	20%	67	22%	64%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	67	100%
	All	510	100%	306	100%	60%	108	35%	49	16%	51	17%	2	1%	2	1%	4	1%	198	65%

Ninety-four percent of the participants aged 0-5 years had a written vaccination record; 71% received four doses. A total of 60% of the participants aged 6-15 years had vaccine records, and 54% had a record of three doses. Difference in vaccine history of DTP3 and DTP4 in the youngest two age groups occurred because the national vaccination program introduced the fourth dose in 2012. Participants aged >36 years in 2017 were not vaccinated because DTP was not introduced in Vietnam until 1981.

Table 2
Seroprevalence (seropositive was defined as IgG value >0.1 IU/ml) and the geometric mean concentration (GMC) weighted by population size, age, and sex structures in two communes in Nha Trang city in 2017.

All		Seroprevalence					GMC		
age group	mean age, ±SD(year)	N	n	%	95% CI	*p-value* (Male vs Female)	IU/ml	95% CI	*p-value* (Male vs Female)
0-5yr	3.8 ±0.02	100	68	68%	(67%, 69%)		0.22	(0.21, 0.22)	
6-15yr	11.1 ±2.74	107	7	7%	(4%, 11%)		0.03	(0.02, 0.04)	
16-25yr	20.2 ±5.92	105	13	12%	(7%, 19%)		0.04	(0.03, 0.05)	
26-35yr	31.1 ±1.19	94	30	33%	(27%, 40%)		0.07	(0.06, 0.08)	
36-55yr	45.7 ±2.44	104	30	28%	(17%, 43%)		0.07	(0.04, 0.11)	
total	28.9 ±3.01	510	148	26%	(20%, 32%)		0.06	(0.05, 0.07)	
Male									
0-5yr	3.8 ±0.37	50	32	64%	(60%, 68%)		0.22	(0.15, 0.32)	
6-15yr	11.1 ±5.5	60	3	5%	(3%, 8%)		0.03	(0.02, 0.04)	
16-25yr	20 ±1.97	46	4	8%	(0%, 66%)		0.03	(0.02, 0.06)	
26-35yr	30.8 ±0.46	30	12	39%	(24%, 57%)		0.08	(0.06, 0.09)	
36-55yr	45.1 ±4.88	35	9	26%	(23%, 29%)		0.07	(0.05, 0.09)	
total	28.2 ±4.76	221	60	25%	(19%, 31%)		0.06	(0.05, 0.07)	
Female									
0-5yr	3.9 ±0.45	50	36	72%	(69%, 75%)	0.20	0.21	(0.14, 0.33)	0.90
6-15yr	11.1 ±1.73	47	4	9%	(3%, 24%)	0.52	0.03	(0.03, 0.04)	0.21
16-25yr	20.4 ±6.57	59	9	16%	(8%, 28%)	0.69	0.04	(0.04, 0.04)	0.50
26-35yr	31.4 ±1.45	64	18	28%	(27%, 29%)	0.35	0.06	(0.06, 0.06)	0.28
36-55yr	46.2 ±2.71	69	21	30%	(12%, 57%)	0.71	0.07	(0.04, 0.13)	0.58
total	29.7 ±2.34	289	88	27%	(18%, 37%)	0.66	0.06	(0.05, 0.08)	0.39

Sex difference in seroprevalence was compared using logistic regression and sex difference in GMC was compared using linear regression.

Table 3
The percent reduction of IgG among four baseline IgG levels, five age groups, and number of doses of DTP.

		Percentage decrease of IgG in two years			paired t-test
		N	% decrease	95%CI	*p-value*
all paired samples		304	16%	(8%,24%)	<0.01
age groups	0-6 years	66	41%	(28%,52%)	<0.01
	7-10 years	27	-18%	(-40%,1%)	0.06
	11-55 years	211	11%	(0%,20%)	0.09
baseline IgG level in 2017	>0.1IU/ml	90	56%	(48%,63%)	<0.01
	0.05-0.1IU/ml	73	15%	(5%,24%)	<0.01
	0.03-0.05IU/ml	61	2%	(-12%,15%)	0.72
	<=0.03IU/ml	80	-50%	(-77%,-28%)	<0.01
age 0-6 years only	3 or 4 doses	57	44%	(30%,55%)	<0.01
	3 doses	11	34%	(-8%,59%)	0.65
	4 doses	46	47%	(31%,59%)	<0.01

Percentage decrease = (1- geometric mean of (IgG2/IgG1) *100 percent IgG1: IgG measured in 2017, IgG2: IgG measured in 2019

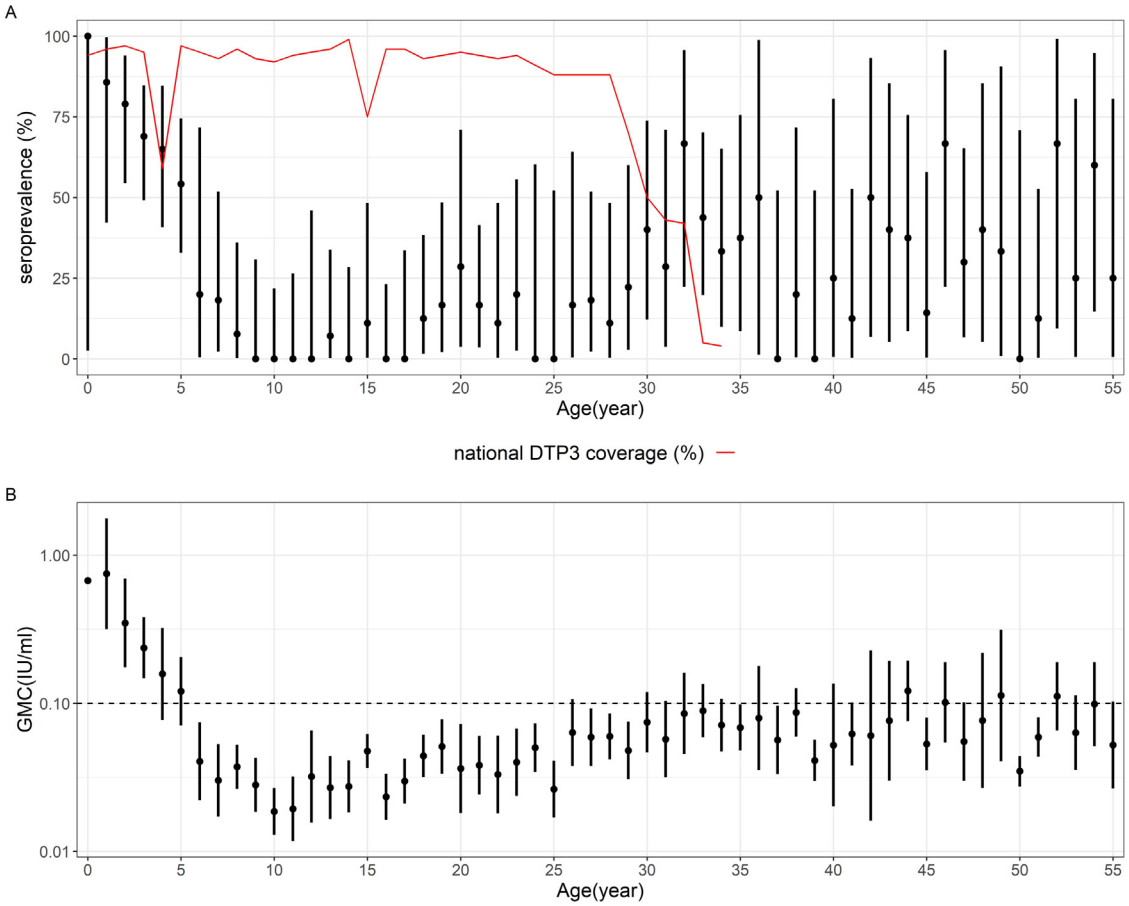

Figure 1. A. Seroprevalence with 95% CI and nationwide DTP3 coverage by single age strata. The low DTP3 coverage in those aged four and 15 years reflects the nationwide suspension of Pentavalent vaccines in 2013 and the stock out of DTP vaccines in 2017.
B. GMC with 95% CI. Dotted horizontal line represents the cut-off for protective immunity (0.1IU/ml).

Discussion

This study describes the age-stratified seroprevalence from infants to adults in well-vaccinated communities where no school-entry booster dose has been introduced. The study shows that 30 years of an immunization program, which provided three or four doses of DTP vaccines within the first two years of life, created a large immunity gap in the study population. Although the reported coverage was high, the seroprevalence was generally low among those aged 6-55 years. The age group in which the reported cases most frequently fell (6-15 years) and the broad age range of cases (1-55 years) in the last few years in Vietnam were consistent with observed seroprevalence pattern by age. The rapid waning of immunity against diphtheria after vaccination may explain the increase in diphtheria cases in Vietnam and other countries in the late 2010s (Wanlapakorn et al. 2014, Hughes et al. 2015, Sein et al. 2016, Sangal et al. 2017, WHO 2017).

Seroprevalence declined quickly from age one to nine years. This result differs from the report that showed that after three doses of DTP vaccine in Germany, antibodies did not wane for the first ten years (Hasselhorn et al. 1998). On the contrary, the pattern of declining seroprevalence in Sweden is similar to that observed in Vietnam (Edmunds et al. 2000). The difference in seroprevalence between populations may be due to the different vaccine components, schedule, and coverage, or the degree of natural infection, but none of them explain the difference clearly. Seroprevalence and GMC tended to increase after ten years of age, which suggests either ongoing transmission of toxigenic *C.diphtheriae* or residual immunity from the transmission in the past.

The seroprevalence among children younger than five years was reported to be 64% in Lao PDR in 2012, which was similar to ours (Nanthavong et al. 2015). The seroprevalence between age 15 and 24 years was 43% in another study in Lao in 2013, and that between age 6 and 25 years was 32% and 52%, respectively, in Kon Tum province in Vietnam in 2016 (Black et al. 2015, Le et al. 2017). The seroprevalence among individuals aged 6-15 and 16-25 years in Lao and Kon Tum was higher than our results, which might reflect natural infection of toxigenic *C. diphtheriae* because both study areas reported cases during the study period.

Although Nha Trang has had no reported diphtheria cases for at least the last ten years, seroprevalence among participants aged 0-55 years was as low as 26% (95%CI:20-32%). The absence of cases in Nha Trang may be attributable to the high DTP3 coverage among infants. Diphtheria cases have been reported where the infant DTP3 coverage was low, e.g., 23% in Rohingya refugee camps in 2017 (Rahman and Islam 2019) and 43% in Nigeria in 2011 (Besa et al. 2014). High immunity among young children may protect the community from large outbreaks although the immunity among older children and adults is low. There are several other potential explanations for the absence of cases despite the low seroprevalence. Our result might not accurately measure the protection level in individuals because we did not use a neutralization assay,

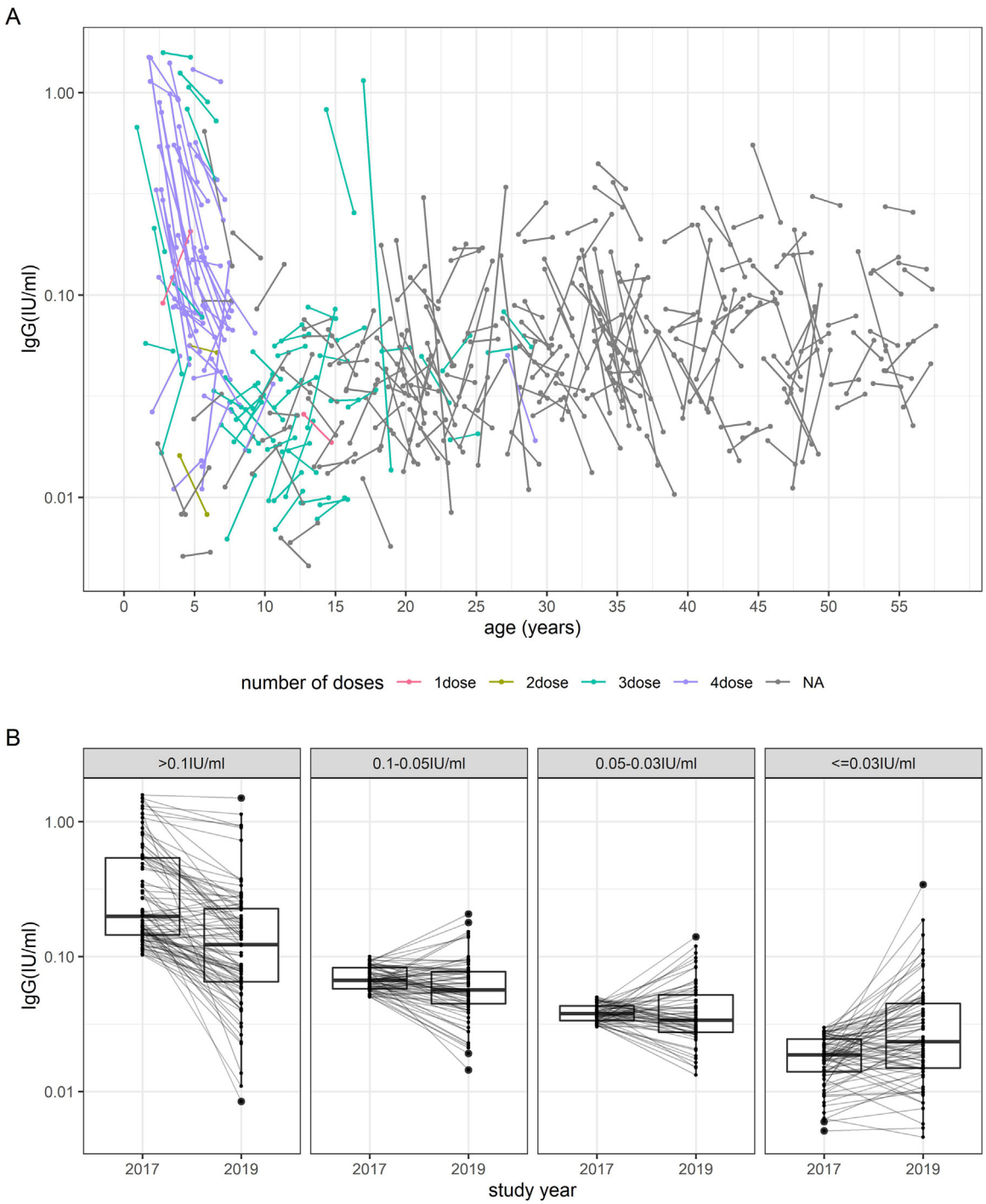

Figure 2. Change of antibody (IgG) levels in 304 paired sera between 2017 and 2019 plotted over age by different number of DTP doses (A) and plotted by four categories of baseline IgG levels with boxplot showing median and first and third quantile of IgG in 2017 and IgG in 2019 (B).

a gold-standard assay. Cellular immunity may persist after vaccination and protect individuals although antibodies are not detected (Gunatillake and Taylor 1968, Heyworth and Ropp 1973).

According to historical data in North America before the introduction of the vaccine, clinical diphtheria was more common in boys among children and in women among adults (Crum 1917). After vaccination was introduced, women were likely to have lower antibody levels than men (Edmunds et al. 2000, Galazka 2000, Plotkin et al. 2018). Biological differences, social role of women, or frequent opportunities of vaccination for men because of military services were discussed. But no clear reasons were identified. This study examined the sex difference in seroprevalence but found no significant difference.

This study shows no evidence of any difference in the waning rate of immunity between receiving three doses or four doses of DTP, although this may be because of the small sample size. The antibody level waned rapidly among individuals with high antibody levels and the antibody level increased among individuals with low antibody levels. This finding suggests that vaccine-derived immunity against diphtheria toxoid is not maintained for life. After it has waned, immunity may increase with subsequent subclinical or clinical infection, as previously reported

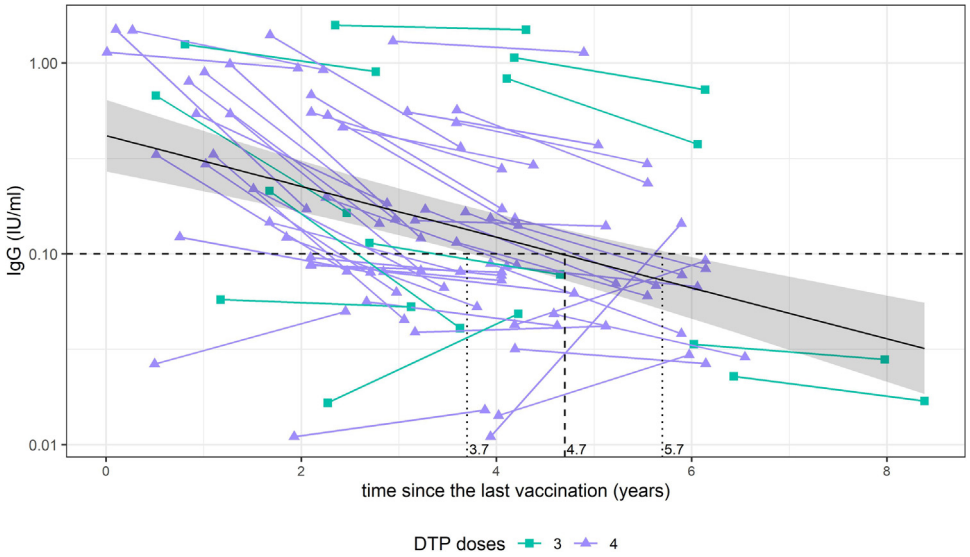

Figure 3. Predicted geometric mean of IgG since the last vaccination after three or four doses of DTP with 95% confidence interval.
Black line and gray area: Geometric mean of IgG concentration since the last vaccination and 95% confidence interval estimated by a mixed effect random intercept model.
Green squares and purple triangles: IgG concentration of each individual who received three doses and four doses of DTP, respectively.
Dotted line shows the protective threshold 0.1 IU/ml. After three or four doses of DTP were given, predicted log-transformed anti-diphtheria toxoid IgG declined linearly and crossed 0.1 IU/ml at 4.7 years after the last vaccination.

(Burnet 1972). On the contrary, regression to the mean could also explain that a high antibody level is likely to decline and a low level of antibody likely to increase (Barnett et al. 2004).

In this study, IgG was estimated to be maintained at a concentration of more than 0.1 IU/ml for 4.3 (95%CI:3.5-5.3) years after four doses of vaccine. Given that the fourth dose is currently scheduled at 18 months of age, 95% of children lose protection between ages of 5.0 and 6.8 years, and thus a booster dose should be administered at school entry, which is at age six in Vietnam. Because DTP vaccine was replaced in Vietnam by Pentavalent vaccine in 2011 and a booster dose at 18 months was introduced in 2012, most of the participants in this analysis had received vaccines according to the current vaccination program. Therefore, the recommendation drawn from this study results will be suitable for Vietnam. This is also compatible with the current WHO recommendation.

This study has several strengths. We used written records to confirm the participants' last vaccination date, which provided accurate vaccination history and time since the last vaccination in each individual. We used paired sera collected in the longitudinal study to estimate the waning of immunity over time.

There are several limitations to this study. It was conducted with a small sample size in a limited geographical area. We collected data only at two points at a fixed interval. The follow-up duration was short, and the results might be affected by random error as there were only two data points. The vaccination history could not be confirmed among older participants, and thus the long-term effect of vaccine-derived immunity could not be evaluated.

An exponential decay model was used to estimate the length of time of vaccine protection up to seven years of age in this study. On the contrary, a previous study used a power function decay model to analyze the waning of anti-diphtheria toxoid IgG (Swart et al. 2016). Another longitudinal study following up the same individuals for seven years showed a biphasic waning pattern of anti-diphtheria toxoid IgG (Nakayama et al. 2019). The exponential decay model may not best fit, especially when follow-up time is longer.

Conclusions

This study showed that the population aged six years or older was largely susceptible in the study community in Vietnam, where the DTP3 coverage was high in the last ten years except in 2013. If any cases were imported or DTP3 coverage declined for some reason, there is a potential risk of re-emergence of the disease. Considering these risks, a cost-effectiveness analysis for introducing an additional booster dose may be warranted. The high case fatality ratio of diphtheria would justify a booster dose at school entry to reduce preventable deaths. The study also showed that a school-entry booster dose would be required to maintain immunity against diphtheria in Vietnam.

Conflict of Interest

There is no conflict of interest to be declared.

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Ethical consideration

Ethical approval was obtained from the Vietnamese Ministry of Health and the London School of Hygiene and Tropical Medicine ethical review boards (IRB-VN01057-27/2015, LSHTM Ethics ref: 17518/17913).

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.ijid.2022.01.025.

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