Table 2 Adverse Reactions in Children Two to Five Years Old Who Received Two Injections of Vi-rEPA Conjugate

	First injection		Second injection	
	Vi-rEPA	Placebo	Vi-rEPA	Placebo
N	5991	6017	5525	5566
Temp. >37.5°C	81 (1.35%)	32 (0.53%)	109 (1.97%)	25 (0.45%)
> 39.0°C	17 (0.28%)	5 (0.08 %)	1 (0.02%)	1 (0.02%)
Swelling >5 cm	0	0	20 (0.36%)	1 (0.02%)
Erythema >5 cm	0	0	2 (0.04%)	0

Table 3 Efficacy of Vi-rEPA in Children Two- to Five-Years Old Over 46 Months of Follow-Up, Including 27 Months of Active Surveillance Followed by 19 Months of Passive Surveillance

Variable	Vaccine group	Placebo group	Vaccine efficacy (95% CI)
No. fully immunized	5466	5506	89.0% (76.0–96.9)
No. of typhoid cases	8	73	
No. of single dose[a]	388	383	87.7% (50.1–94.8)
No. of typhoid cases	1	8	

[a]Children received only one injection but participated in the surveillances.

children had a ≥8-fold rise in serum anti-Vi IgG. There was a booster response after the second dose.

Phase III Clinical Trial

The safety, immunogenicity, and efficacy of the Vi-rEPA conjugate vaccine were evaluated in a double-blind, randomized trial in children two to five years old in 16 communes in Dong Thap province, Vietnam (10). In 1998, 11,091 two to five-year old children received two injections, six weeks apart, of either Vi-rEPA or a saline placebo. Less than 2% of children had adverse reactions, none considered serious (Table 2). Cases of typhoid, confirmed by the isolation of *S. typhi* from blood cultures after three or more days of fever, were identified by active surveillance over a period of 27 months, and passive surveillance during an additional 19 months after the vaccine code was opened (9).

Over 27 months of follow-up, *S. typhi* was isolated from 4 of the 5525 children who were fully vaccinated with Vi-rEPA versus from 47 of the 5566 children who received both injections of placebo (91.5% efficacy, 95% CI, 77.1–96.6) (Table 3).

During the 19 months of passive surveillance, typhoid was detected in 3 vaccinees and in 17 placebo recipients (82.4% efficacy; 95% CI, 22.3–99.1). Over the entire 46-month period, the vaccine efficacy was 89.0% (95% CI, 76.0–96.9) (Table 3) (10). Among children who received only one injection ($n = 771$), there was one case of typhoid in 388 children in the vaccine group and 8 cases in 383 children in the placebo group; thus, the estimated efficacy with only 1 injection of Vi-rEPA was 87.7% (95% CI, 50.1–94.8).

Throughout the surveillance period, blood samples were collected monthly from four randomly selected participants of each commune for assessing IgG anti-Vi (10). The persistence of serum IgG anti-Vi level was examined as the geometric means (GMs) for each half-year. There was an age dependence in IgG anti-Vi response, but the difference of GM levels between the younger (2–3 years old) and older (4–5 years old) was not statistically significant. We estimated the protective level of IgG anti-Vi to be 3.52 EU (~0.11 μg/mL IgG) based on the GM of the younger age group at 46 months, since there was no statistically significant difference between the efficacies in the two stratified age groups (10). At 42 months after vaccination, the GM level of IgG anti-Vi decreased from 22.50 to 3.66 EU in the vaccine group, and increased from 0.65 to 0.80 EU in the placebo group (3.66 vs. 0.8, $p < 0.001$). The slight increase in the placebo group could reflect the environmental stimulation during the study period.

Long-Term Follow-up of Phase III Study

Since our ultimate goal for Vi-rEPA trials is to incorporate this vaccine into immunization programs, data on long-term protection and antibody persistence are essential. A follow-up study is underway, which will provide information on the duration of protection in adults and school-age children, and should determine whether a booster dose of Vi-rEPA is necessary. The preliminary data show that in adults, the anti-Vi IgG level remained more than sevenfold higher than the prevaccination baseline 10 years after vaccination.

The phase III study populations provide a unique opportunity to assess protection eight years after one or two injections of Vi-rEPA were administered at two to five years of age, and four years after one injection at five to eight years. All children in the phase III trial are now 10 to 13 years old. They represent ~7% of the total population of the trial district of this age group (5–15 years old), the peak age incidence of typhoid in Vietnam. The reduction in typhoid fever in this age cohort might have reduced transmission of *S. typhi* to families and contacts of other age groups. Our review of hospitalized cases of typhoid in all ages from the trial district and nontrial district will provide data on the effects of vaccination on the community.

Protection will be assessed by comparing the rates of hospitalized typhoid fever among children in the trials to that in unvaccinated children of similar age in the adjacent district/town, that is, Binh Thanh district and Cao Lanh town. The persistence of antibody will be assessed by a serological survey in the twice-injected and once-injected children compared with the levels in unvaccinated children of similar age. Serological comparison will also be evaluated for the persistence of IgG anti-Vi in the vaccinated children and compared with the levels in unvaccinated children of similar age.

Dosage Study

Dosage-related immunogenicity has been observed for other polysaccharide conjugates such as Hib and pneumococcal vaccines. A dosage-immunogenicity study of Vi-rEPA was evaluated in 241 children two to five years old in Phu Tho province, Vietnam. Children were divided randomly into three groups, each received two injections, six weeks apart, containing 25 μg (full dosage), 12.5 μg, or 5 μg of Vi-rEPA. At 10 weeks after the first injection, all children responded with greater than estimated protective level of anti-Vi IgG (>3.52 EU/mL) (Table 4) (34). There was a direct correlation between the dosage and the