Vaccine for Prevention of Mild and Moderate-to-Severe Influenza in Children

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ORIGINAL ARTICLE

Vaccine for Prevention of Mild and Moderate-to-Severe Influenza in Children

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ABSTRACT

BACKGROUND

Commonly used trivalent vaccines contain one influenza B virus lineage and may be ineffective against viruses of the other B lineage. We evaluated the efficacy of a candidate inactivated quadrivalent influenza vaccine (QIV) containing both B lineages.

METHODS

In this multinational, phase 3, observer-blinded study, we randomly assigned children 3 to 8 years of age, in a 1:1 ratio, to receive the QIV or a hepatitis A vaccine (control). The primary end point was influenza A or B confirmed by real-time polymerase chain reaction (rt-PCR). Secondary end points were rt-PCR–confirmed, moderate-to-severe influenza and rt-PCR–positive, culture-confirmed influenza. The vaccine efficacy and the effect of vaccination on daily activities and utilization of health care resources were assessed in the total vaccinated cohort (2584 children in each group) and the per-protocol cohort (2379 children in the QIV group and 2398 in the control group).

RESULTS

In the total vaccinated cohort, 62 children in the QIV group (2.40%) and 148 in the control group (5.73%) had rt-PCR–confirmed influenza, representing a QIV efficacy of 59.3% (95% confidence interval [CI], 45.2 to 69.7), with efficacy against culture-confirmed influenza of 59.1% (97.5% CI, 41.2 to 71.5). For moderate-to-severe rt-PCR–confirmed influenza, the attack rate was 0.62% (16 cases) in the QIV group and 2.36% (61 cases) in the control group, representing a QIV efficacy of 74.2% (97.5% CI, 51.5 to 86.2). In the per-protocol cohort, the QIV efficacy was 55.4% (95% CI, 39.1 to 67.3), and the efficacy against culture-confirmed influenza 55.9% (97.5% CI, 35.4 to 69.9); the efficacy among children with moderate-to-severe influenza was 73.1% (97.5% CI, 47.1 to 86.3). The QIV was associated with reduced risks of a body temperature above 39°C and lower respiratory tract illness, as compared with the control vaccine, in the per-protocol cohort (relative risk, 0.29 [95% CI, 0.16 to 0.56] and 0.20 [95% CI, 0.04 to 0.92], respectively). The QIV was immunogenic against all four strains. Serious adverse events occurred in 36 children in the QIV group (1.4%) and in 24 children in the control group (0.9%).

CONCLUSIONS

The QIV was efficacious in preventing influenza in children. (Funded by GlaxoSmithKline Biologicals; ClinicalTrials.gov number, NCT01218308.)

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The incidence of influenza among children is high, and the illness is associated with substantial increases in outpatient visits and hospitalizations during the influenza season.[1-4] Routine vaccination of children against influenza is recommended in the United States[5] and some other countries, despite limited evidence of the efficacy of inactivated influenza vaccine from randomized, controlled trials involving children.[6]

When trivalent influenza vaccines (TIVs) are used, there is a possibility of a mismatch between circulating and vaccine B strains, which results in inadequate protection from the vaccine.[7-10] A quadrivalent vaccine containing both B lineages would eliminate B-lineage mismatch. This may be particularly important in children, in whom TIVs elicit weak cross-reactive antibody responses and have reduced efficacy against influenza B caused by strains of the lineage that are not contained in the vaccine.[10-12]

In the current study, we evaluated the efficacy of a candidate inactivated quadrivalent influenza vaccine (QIV) for the prevention of influenza A or B in children 3 to 8 years of age, using a conventional end point (any influenza) and an additional end point (moderate-to-severe influenza) that captures the more clinically significant outcomes of influenza.

Methods

Study Oversight

We conducted a phase 3, randomized, controlled, observer-blind study with funding from GlaxoSmithKline Biologicals. The sponsor was involved in all stages of the study conduct and analysis of the data. The aim of the study was to assess the efficacy of the QIV for prevention of influenza A or B in children 3 to 8 years of age. The presence of influenza virus was to be confirmed by means of a real-time polymerase-chain-reaction (rt-PCR) assay. The trial received ethical approval at each participating center (see the Supplementary Appendix, available with the full text of this article at NEJM.org). Parents or legal representatives provided written informed consent, and children 7 years of age or older provided written assent, according to local standards. The sponsor donated the vaccines. Members of the core writing team (see the Supplementary Appendix) collaborated on writing all drafts of the manuscript, with assistance from a professional medical writer, who was paid by the sponsor. All the authors assume responsibility for the accuracy and completeness of the data and for the fidelity of the study to the protocol, which is available at NEJM.org.

Participants

We recruited healthy children from 15 centers in Bangladesh, the Dominican Republic, Honduras, Lebanon, Panama, the Philippines, Thailand, and Turkey (see the Supplementary Appendix). Children were randomly assigned, in a 1:1 ratio, to receive the QIV (0.5-ml dose) or hepatitis A vaccine (Havrix, 0.5-ml dose), as a control (see the Supplementary Appendix). Both vaccines were manufactured by GlaxoSmithKline Vaccines. The QIV contained 15 μg of hemagglutinin antigen from each of four strains: A/California/7/2009 (H1N1), A/Victoria/210/2009 (H3N2), B/Brisbane/60/2008 (Victoria), and B/Florida/4/2006 (Yamagata). Children received one or two vaccine doses depending on their vaccine priming status (Fig. S1 in the Supplementary Appendix; see the Methods section in the Supplementary Appendix for the criteria used to define priming).

Study Design

The first children were enrolled in December 2010; the exact date varied among countries. Active and passive surveillance for influenza-like illness were conducted for at least 6 months (Fig. S1 in the Supplementary Appendix), continuing until the end of October 2011. An influenza-like illness was defined as a temperature of 37.8°C or higher, with at least one of the following: cough, sore throat, runny nose, or nasal congestion. The parents of children with an influenza-like illness completed a daily diary for 14 days starting from the onset of the illness (see the Supplementary Appendix).

Detection of Influenza Virus

The presence of influenza A or B virus in nasal or throat swabs from children with an influenza-like illness was confirmed by means of an rt-PCR assay,[13] and positive samples were further tested by means of cell culturing.[14] Influenza A subtyping was performed with the use of a nested reverse-transcriptase (RT)–PCR assay on clinical specimens, with primers targeting the hemagglutinin gene

for H1 and H3 (Table S1 in the Supplementary Appendix). Classification of influenza B as Yamagata or Victoria lineage was also performed by means of a nested RT-PCR assay with primers targeting hemagglutinin (Table S1 in the Supplementary Appendix), followed by sequencing analysis. Antigenic matching was performed on cultured isolates and was defined as a difference by less than a factor of 8 in hemagglutination inhibition relative to reference serum and vaccine antigen.

STUDY END POINTS

Efficacy

The primary end point was influenza A or B of any severity that was confirmed by means of rt-PCR assay. Secondary end points were rt-PCR–confirmed, moderate-to-severe influenza A or B and culture-confirmed influenza A or B caused by seasonal strains antigenically matching the vaccine strains or by any seasonal strain. Moderate-to-severe disease was defined as a body temperature higher than 39°C, physician-confirmed acute otitis media, lower respiratory tract illness (shortness of breath, pulmonary congestion, pneumonia, bronchiolitis, bronchitis, wheezing, or croup), or serious extrapulmonary complications such as myositis, encephalitis, seizure, or myocarditis. Exploratory end points were the incidence of influenza caused by individual A subtypes or B lineages, the incidence of influenza in two age subgroups (3 to 4 years and 5 to 8 years), clinical manifestations of moderate-to-severe influenza, and the effect of vaccination on daily activities and utilization of health care resources.

Immunogenicity and Safety

The hemagglutination-inhibition antibody titer against each vaccine strain was measured with the use of standard methods.[15] The immune response was assessed in a randomly chosen immunogenicity subgroup that comprised 544 children in the QIV group and 163 in the control group in the total vaccinated cohort and 457 children in the QIV group and 122 in the control group in the per-protocol cohort. The following safety end points were evaluated by means of a review of the diary card: injection-site and systemic symptoms (solicited information), assessed during the 7-day period after the vaccination; spontaneously reported (unsolicited) symptoms, assessed during the 28-day period after vaccination; and serious adverse events and adverse events for which medical care was sought, assessed over the course of the whole study period.

STATISTICAL ANALYSIS

The principal analysis was an analysis of efficacy in the per-protocol cohort (children who met all eligibility criteria, were successfully contacted at least once after vaccination, and adhered to the protocol); efficacy was also analyzed in the total vaccinated cohort. Immunogenicity was evaluated in a subgroup of the per-protocol cohort, and safety was evaluated in the total vaccinated cohort (Fig. S2 in the Supplementary Appendix).

The efficacy of the QIV was assessed with the use of time-to-event methods by means of a Cox regression model adjusted for age, region, and priming status, with 95% confidence intervals for the primary end point and exploratory end points and 97.5% confidence intervals for secondary end points (see the Supplementary Appendix). The criterion for declaring efficacy was a lower limit of the confidence interval of more than 30% for the primary end point (with the criterion affirmed by the Food and Drug Administration) and more than 0% for secondary end points. The principal analysis included cases identified from 14 days after vaccination to the end of the study; a complementary analysis included cases identified from day 0.

Assuming a QIV efficacy of 60%, we estimated that approximately 194 cases of rt-PCR–confirmed influenza A and B would need to be documented in order to show a lower limit of the 95% confidence interval for the vaccine efficacy that was more than 30% at a 5% level of significance. On the basis of an estimated attack rate of 6%, and assuming that we would not be able to evaluate approximately 10% of the children, we calculated that we would need to recruit approximately 5200 children. The statistical analysis was performed with the use of SAS software, version 9.2 (SAS Institute).

RESULTS

PARTICIPANTS

A total of 5220 children were enrolled in the study: 5168 children in the total vaccinated cohort (2584 in the QIV group and 2584 in the control group) and 4777 in the per-protocol efficacy

cohort (2379 in the QIV group and 2398 in the control group) (Fig. S2 in the Supplementary Appendix). The mean age was 5.4 years in both the QIV and control groups, with approximately equal numbers of boys and girls (Table S2 in the Supplementary Appendix). Five children who had not undergone influenza vaccine priming and who had received one dose of vaccine during the study did not receive the second vaccine dose because of illness at the scheduled time of the second dose (2 children), unavailability during the scheduled window for vaccination defined by the protocol (1 child), parental decision with no reason given (1 child), and investigator decision owing to vaccination history (1 child) (Table S3 in the Supplementary Appendix).

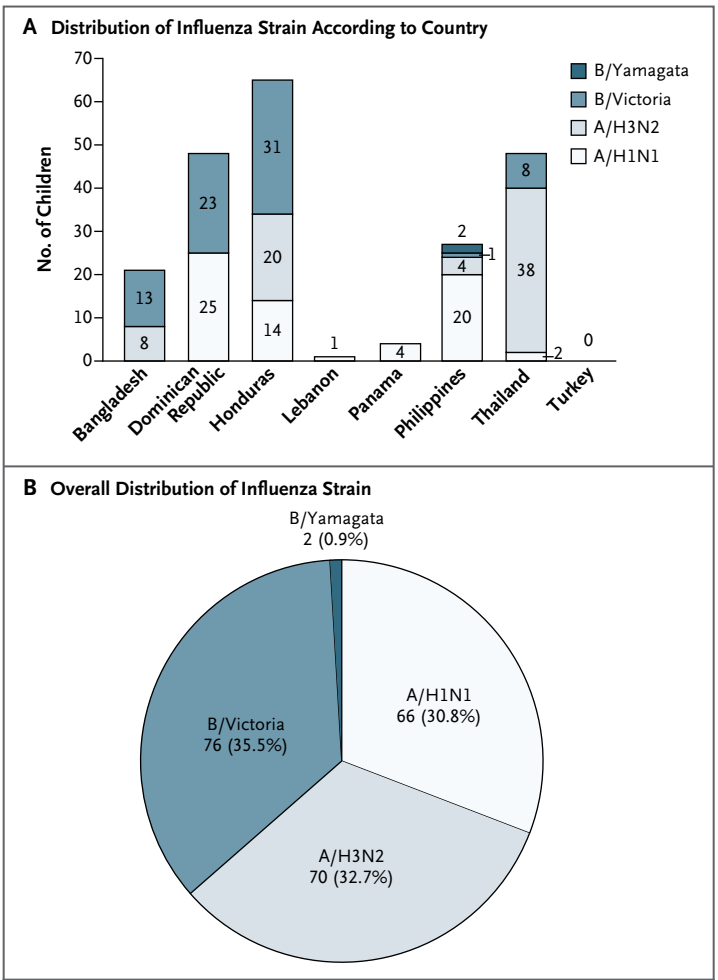

Figure 1. Distribution of Influenza A Subtype and Influenza B Lineage.

The distribution of four strains of influenza confirmed by means of a real-time polymerase-chain-reaction (rt-PCR) assay is shown according to country (Panel A) and overall (Panel B), with data from the total vaccinated cohort during the period from day 0 after vaccination until the end of the study. A total of 210 children had 216 cases of rt-PCR–confirmed influenza; 204 children had a single case and 6 children had two cases each. In 4 of these 6 children, the two cases were associated with different influenza strains, and in 2 children only one case could be typed. The graph does not include the two cases that could not be typed, and therefore shows 214 cases.

INFLUENZA-LIKE ILLNESS

In the total vaccinated cohort, 563 influenza-like illnesses occurred in 422 children in the QIV group and 657 influenza-like illnesses occurred in 507 children in the control group during the period from day 0 after vaccination until the end of the study. A swab specimen was obtained in 96% of the cases of influenza-like illness and was obtained within 7 days after the onset of illness in all but 1 case. A total of 62 children in the QIV group (2.4%) and 148 in the control group (5.7%) had rt-PCR–confirmed influenza (Fig. 1). In the per-protocol efficacy cohort, 462 influenza-like illnesses occurred in 352 children in the QIV group and 531 occurred in 416 children in the control group during the period from 14 days after vaccination until the end of the study; 58 children (2.4%) and 128 (5.3%), in the two groups, respectively, had rt-PCR–confirmed influenza. Three children in each group in the per-protocol cohort had two rt-PCR–confirmed infections each; the second infections were not included in the analysis of vaccine efficacy.

EFFICACY

The study met its primary objective by showing that the efficacy of the QIV against any case of rt-PCR–confirmed influenza was 59.3% (95% confidence interval [CI], 45.2 to 69.7) in the total vaccinated cohort and 55.4% (95% CI, 39.1 to 67.3) in the per-protocol cohort (Table 1, and Fig. S3a and S4a in the Supplementary Appendix). The attack rate was 2.40% (62 cases) with the QIV and 5.73% (148 cases) with the control vaccine in the total vaccinated cohort; the corresponding rates in the per-protocol cohort were 2.44% (58 cases) and 5.34% (128 cases) (Table 1). Our intention in evaluating the end point of moderate-to-severe influenza was to dichotomize cases into categories of mild illness versus illnesses that are associated with the most clinically significant aspects of influenza. We observed higher efficacy of the QIV against moderate-to-severe influenza than against influenza of any severity. The efficacy against moderate-to-severe influenza in the

Table 1. Vaccine Efficacy against rt-PCR–Confirmed and Culture-Confirmed Influenza A or B According to Age and A Subtype and B Lineage.*

Cohort and Influenza Variable	QIV Group		Control Group		QIV Efficacy
	Cases	Attack Rate	Cases	Attack Rate	
	no.	%	*no.*	%	% *(95% CI)*
Total vaccinated cohort					
rt-PCR–confirmed influenza, any severity	62	2.40	148	5.73	59.3 (45.2 to 69.7)
rt-PCR–confirmed influenza, moderate-to-severe	16	0.62	61	2.36	74.2 (51.5 to 86.2)†
Culture-confirmed, rt-PCR–confirmed influenza, any severity, any seasonal strain	54	2.09	129	4.99	59.1 (41.2 to 71.5)†
Culture-confirmed, rt-PCR–confirmed influenza, any severity, vaccine-matched strain	35	1.35	66	2.55	47.7 (16.4 to 67.3)†
Per-protocol efficacy cohort					
rt-PCR–confirmed influenza, any severity	58	2.44	128	5.34	55.4 (39.1 to 67.3)
rt-PCR–confirmed influenza, moderate-to-severe	14	0.59	52	2.17	73.1 (47.1 to 86.3)†
Culture-confirmed, rt-PCR–confirmed influenza, any severity, any seasonal strain	50	2.10	112	4.67	55.9 (35.4 to 69.9)†
Culture-confirmed, rt-PCR–confirmed influenza, any severity, vaccine-matched strain	31	1.30	56	2.34	45.1 (9.3 to 66.8)†
Per-protocol efficacy cohort stratified by age‡					
rt-PCR–confirmed influenza, any severity					
Children 3–4 yr of age	32	3.78	48	5.69	35.3 (–1.3 to 58.6)
Children 5–8 yr of age	26	1.70	80	5.15	67.7 (49.7 to 79.2)
rt-PCR–confirmed influenza, moderate-to-severe					
Children 3–4 yr of age	6	0.71	18	2.13	67.5 (18.0 to 87.1)
Children 5–8 yr of age	8	0.52	34	2.19	76.2 (48.5 to 89.0)
Per-protocol efficacy cohort stratified by strain§					
rt-PCR–confirmed influenza, any severity					
Influenza A	37	1.56	85	3.54	56.8 (36.4 to 70.6)
A/H1N1	17	0.71	38	1.58	55.6 (21.3 to 74.9)
A/H3N2	20	0.84	47	1.96	57.6 (28.5 to 74.9)
Influenza B	23	0.97	45	1.88	49.5 (16.6 to 69.5)
B/Victoria	23	0.97	43	1.79	47.2 (12.4 to 68.2)
B/Yamagata	0	0.00	2	0.08	100.0 (— to 100.0)
rt-PCR–confirmed influenza, moderate-to-severe					
Influenza A	8	0.34	40	1.67	79.9 (57.1 to 90.6)
A/H1N1	4	0.17	17	0.71	76.5 (30.3 to 92.1)
A/H3N2	4	0.17	23	0.96	82.4 (49.1 to 93.9)
Influenza B	7	0.29	13	0.54	46.5 (34.1 to 78.7)
B/Victoria	7	0.29	12	0.50	42.1 (47.1 to 77.2)
B/Yamagata	0	0.00	1	0.04	100.0 (— to 100.0)

* The total vaccinated cohort included 2584 children in the quadrivalent influenza vaccine (QIV) group and 2584 in the control group. The incidence of influenza was assessed from day 0 after vaccination to the end of the study. The per-protocol cohort included 2379 children in the QIV group and 2398 in the control group. The incidence of influenza was assessed from day 14 after vaccination to the end of the study. Rt-PCR denotes real-time polymerase chain reaction.

† The confidence interval in this category is a 97.5% confidence interval.

‡ In the QIV group, 846 children were 3 or 4 years of age, and 1533 were 5 to 8 years of age; in the control group, 844 children were 3 or 4 years of age, and 1554 were 5 to 8 years of age. The analysis of vaccine efficacy according to age was a protocol-specified exploratory analysis.

§ The analysis of vaccine efficacy according to A subtype and B lineage was a post hoc exploratory analysis.

total vaccinated cohort was 74.2% (97.5% CI, 51.5 to 86.2), with attack rates of 0.62% (16 cases) in the QIV group and 2.36% (61 cases) in the control group (Table 1, and Fig. S3b in the Supplementary Appendix). The corresponding efficacy in the per-protocol cohort was 73.1% (97.5% CI, 47.1 to 86.3), with an attack rate of 0.59% (14 cases) in the QIV group and 2.17% (52 cases) in the control group (Table 1, and Fig. S4b in the Supplementary Appendix). The QIV vaccine was also effective against rt-PCR–positive, culture-confirmed disease caused by any seasonal or vaccine-matched strains (Table 1), with higher efficacy against moderate-to-severe disease than against influenza of any severity. However, most H3N2 isolates could not be antigenically typed for matching to the QIV (Table S4 in the Supplementary Appendix).

In an exploratory analysis, we assessed QIV efficacy according to influenza A subtype and influenza B lineage. The QIV was most effective against moderate-to-severe influenza A (Table 1). Only two cases of influenza B caused by Yamagata lineage viruses were observed, both in the control group. Efficacy against influenza of any severity appeared to be lower among children 3 to 4 years of age than among children 5 to 8 years of age in the per-protocol cohort (35.3% [95% CI, –1.3 to 58.6] vs. 67.7% [95% CI, 49.7 to 79.2]) (Table 1). However, efficacy against moderate-to-severe disease appeared to be similar among children in the two age groups (67.5% [95% CI, 18.0 to 87.1] and 76.2% [95% CI, 48.5 to 89.0], respectively). An unadjusted per-protocol analysis and analysis of the total vaccinated cohort showed similar levels of efficacy (Tables S5 and S6 and Fig. S5 in the Supplementary Appendix).

High body temperature and lower respiratory tract illness were the most common symptoms associated with moderate-to-severe influenza; both symptoms were reported in fewer children in the QIV group than in the control group (Table 2, and Table S7 in the Supplementary Appendix). Few children had acute otitis media or pneumonia (one and three cases, respectively, in the control group), and none had an extrapulmonary complication of influenza.

The QIV reduced the effect of illness on daily activities, parental time away from work, and utilization of health care resources, particularly in cases of moderate-to-severe disease. In the per-protocol cohort, of 66 children with rt-PCR–confirmed, moderate-to-severe influenza, 8 children in the QIV group and 35 in the control group missed school (relative risk with QIV, 0.23 [95% CI, 0.11 to 0.49]), and 1 child in the QIV group and 4 in the control group were hospitalized (relative risk, 0.25 [95% CI, 0.03 to 2.25]) (Table 2, and Table S8 in the Supplementary Appendix). The results were similar in the total vaccinated cohort (Tables S7 and S9 in the Supplementary Appendix).

IMMUNE RESPONSE

In the immunogenicity subgroup of the per-protocol cohort, the geometric mean titer after administration of the QIV was 10 to 20 times the prevaccination titer against all four vaccine strains (Fig. 2). After vaccination, the seroprotection rate against each strain was more than 95% (Fig. S6 in the Supplementary Appendix). Immunogenicity was similar in the two age groups. At 6 months and later after vaccination, the seroprotection rate against the A/H3N2 and B/Yamagata strains was more than 90%, and the seroprotection rate against the A/H1N1 and B/Victoria strains was more than 80% (Fig. S6 in the Supplementary Appendix). The findings were similar in the immunogenicity subgroup of the total vaccinated cohort.

SAFETY

There were no notable differences between the QIV group and the control group with respect to safety end points, except that pain at the injection site was reported more frequently in the QIV group (Table 3, and Table S10 in the Supplementary Appendix). Serious adverse events occurred in 36 children in the QIV group (1.4%) and in 24 children in the control group (0.9%) (Table 3, and Table S11 in the Supplementary Appendix).

DISCUSSION

There is limited evidence from randomized trials to support the administration of inactivated influenza vaccine in healthy children.[6] This randomized study provides additional evidence of the efficacy of QIV against influenza (as confirmed by means of rt-PCR assay). The study was performed to provide direct evidence of the clinical benefit of the vaccine, especially in the prevention of moderate-to-severe influenza, an end point that captures the most clinically significant

Table 2. Clinical Characteristics of rt-PCR–Confirmed Influenza and Effect on Daily Activities.*

Variable	Moderate-to-Severe Disease			Mild Disease		
	QIV Group	Control Group	Relative Risk with QIV (95% CI)	QIV Group	Control Group	Relative Risk with QIV (95% CI)
Clinical characteristics						
No. of rt-PCR–confirmed cases	14	52				
Temperature >39°C with no other defining symptom (no. of patients)	12	41	0.29 (0.16–0.56)			
Acute otitis media with no other defining symptom (no. of patients)	0	1	—			
Lower respiratory tract illness (no. of patients)†	2	10	0.20 (0.04–0.92)			
With pneumonia‡	0	3	—			
Without pneumonia	2	7	0.29 (0.06–1.38)			
Effect on daily activities and utilization of health care resources						
No. of rt-PCR–confirmed cases§	14	52		45	76	
Absence from school						
No. of children affected	8	35	0.23 (0.11–0.49)¶	23	41	0.56 (0.34–0.94)¶
No. of school days missed per illness	4.1±3.1	5.0±3.7	—	3.6±2.5	3.4±1.9	—
Parental absence from work to take care of the child						
No. of parents affected	7	18	0.39 (0.16–0.93)¶	18	18	1.01 (0.52–1.93)¶
No. of work days missed per illness	3.1±2.0	4.2±3.4	—	2.5±1.6	4.8±4.6	—
Visit to doctor or other medical person						
No. of children affected	14	45	0.31 (0.17–0.57)	35	65	0.54 (0.36–0.81)
No. of doctor visits per illness	2.3±1.8	1.8±1.7	—	1.4±1.1	1.4±1.0	—
Hospitalization						
No. of children affected	1	4	0.25 (0.03–2.25)	0	1	—
No. of days in hospital per illness	5.0	4.8±2.9	—	—	4.0	—

* Plus–minus values are means ±SD. Data are from the per-protocol efficacy cohort, which comprised 2379 children in the QIV group and 2398 in the control group. A total of 12 children (3 in the QIV group and 9 in the control group) were excluded from the analysis because they received a medication or vaccine that was not allowed by the protocol. The analysis of clinical characteristics of the illness was a post hoc exploratory analysis of data from day 14 after vaccination until the end of the study. The analysis of the effect on daily activities and utilization of health care resources was a protocol-specified exploratory analysis of data from day 0 to day 14 of the illness.

† Lower respiratory tract illness included bronchitis, wheezing, shortness of breath, pneumonia, and pulmonary congestion.

‡ Two of the three cases of pneumonia were diagnosed by means of chest radiography, but no causative organism was identified. No information is available for the third case, which was a spontaneously reported adverse event, with no explanation of the method of diagnosis and no description of symptoms.

§ One child had one episode of rt-PCR–confirmed moderate-to-severe influenza and one episode of rt-PCR–confirmed mild influenza and is included in both categories.

¶ Data were included only when both parents worked outside the home.

outcomes leading to health care consultations or hospitalization. The greatest value of vaccination is in the prevention of clinically significant disease rather than mild upper respiratory tract illness. Parents are most likely to seek medical help for children with a respiratory tract infection who have symptoms of lower respiratory tract disease, high temperature, or earache.[16] Studies that do not differentiate these manifestations of influenza from mild illness cannot assess the effectiveness of the vaccine in attenuating illness and therefore may undervalue its benefit.

Our study showed QIV efficacy of 55% against influenza of any severity. This is similar to the

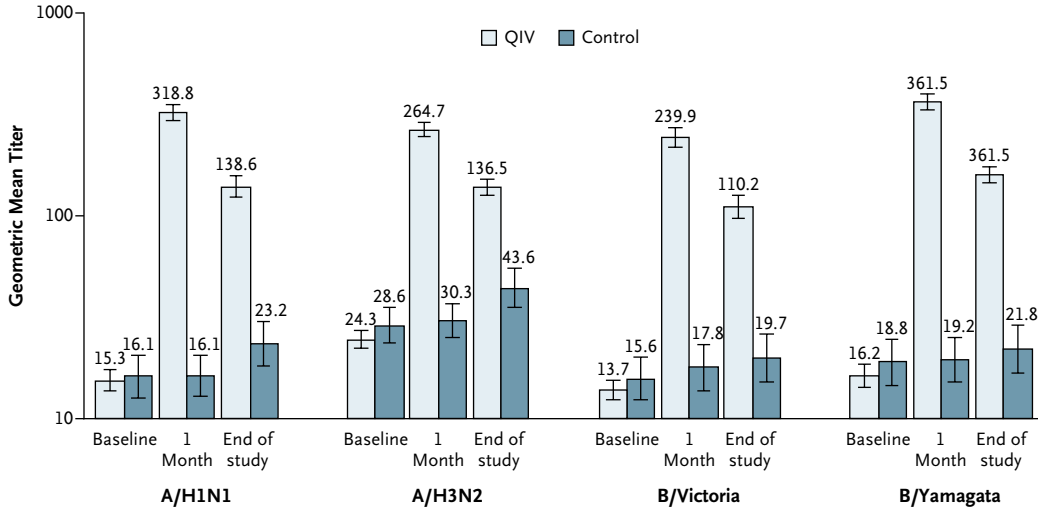

Figure 2. Immunogenicity According to Strain.

Hemagglutination-inhibition antibody titers against influenza A subtype and B lineage in the per-protocol cohort are shown for the period before vaccination, 1 month after administration of a single vaccine dose in children who had influenza priming or 1 month after the second dose in children who did not have influenza priming, and at the end of the study (6 to 8 months after first dose). (For the definition of priming in the study, see the Supplementary Appendix.) Data are expressed as geometric mean titers (the antilog of the arithmetic mean of the $\log_{10}$-transformed titers). Data were available before vaccination and 1 month after vaccination for 457 children in the group that received the quadrivalent influenza vaccine (QIV) and 122 in the group that received the control vaccine (a hepatitis A vaccine) and were available at the end of the study for 444 children in the QIV group and 115 in the control group. Children with a hemagglutination-inhibition antibody titer of less than 1:10 (assay cutoff value) were considered to be seronegative. I bars indicate 95% confidence intervals.

estimates of the efficacy of TIV in other randomized trials: 43% (among children 6 months to 6 years of age),[17] 51% (among children 18 months to 6 years of age),[18] and 56% (among children 3 to 9 years of age, against H1N1 disease),[19] as well as an estimate of 48% from a meta-analysis of data for children of all ages.[20] In our study, the efficacy of QIV was higher against moderate-to-severe disease (approximately 70% overall and in each of the two age groups). Most breakthrough cases in the QIV group were of mild severity. In a post hoc analysis, the QIV was associated with an 80% reduction in the rate of lower respiratory tract illness (the most common serious outcome of influenza) and a 70% reduction in the rate of body temperature above 39°C, as compared with the control vaccine.

Our study provided evidence that the QIV prevents influenza associated with the A/H1N1, A/H3N2, and B Victoria strains individually. Only two rt-PCR–confirmed cases associated with the B Yamagata virus were seen (both in the control group), precluding a meaningful estimate of the vaccine efficacy against that strain. However, the immune response to B Yamagata was as high as the response to the other strains, suggesting that the efficacy of the QIV against B Yamagata may be similar to the efficacy against the other strains. Vaccine efficacy is substantially reduced when the vaccine B strain is mismatched to the circulating strain.[7-10] Only in the Philippines were both B lineages detected, reflecting the unpredictable geographic variability of influenza virus circulation. Introduction of the QIV is expected to result in a modestly lower incidence of influenza-related outcomes than that seen with TIV, with the net effect varying from one season to another.[21] The true value of the QIV will be seen in years when the two B lineages are cocirculating or if there is a shift from one lineage to another between the time the vaccine is developed and the beginning of the influenza season.

Influenza in children results in increased out-

patient visits, hospitalizations, and days missed from school.[1-3,16,22,23] Among children with moderate-to-severe disease, the QIV, as compared with the control vaccine, was associated with 69% fewer medical visits, 75% fewer hospitalizations, 77% fewer absences from school, and 61% fewer parental absences from work. In all the study countries, the school year includes most of the peak influenza season. The age at which children begin day care or school varies considerably among countries, ranging from 3 to 7 years of age. The proportion of parents working outside the home is also variable. The estimate of the effect of vaccination with the QIV on school and workplace absenteeism is thus a broad approximation. In countries outside the study where children begin attending day care or school at an earlier age, a higher proportion of parents work outside the home, or both, the effect of vaccination on absenteeism may be greater than is indicated here.

The safety profile of the QIV was similar to that of hepatitis A vaccine, which we used as a control, except that pain at the injection site was reported more frequently in the QIV group. The experience of injection-site pain with the QIV as compared with TIV has been variable, with some studies suggesting similar levels of pain with the QIV and TIV and others suggesting a modestly higher level of pain with the QIV.[24-27] A higher incidence of fever resulting from inclusion of a fourth strain was a theoretical concern because of the high incidence of fever observed with the TIV Fluvax (CSL) in Australia,[28] which is believed to be due to a larger number of viral components in that vaccine than in other TIVs. However, in the current study, the incidence of fever was similar in the QIV and control groups and similar to the rate observed in previous studies of TIVs in children (in which fever was defined as a body temperature of 37.5°C or higher or 38°C or higher).[15,29] In the current study involving children residing mainly in tropical countries, the immunogenicity of the QIV was high, similar to that in a study of the same vaccine in children 3 to 17 years of age residing in Canada, Mexico, and the United States.[24]

A limitation of the study is that it was conducted during only one season and provides only a snapshot of the situation in that season. For example, the findings are limited by the low circulation of the B Yamagata lineage, as noted above. A major strength is that this was an individually randomized trial of an inactivated influenza vaccine in children 3 to 8 years of age. The study was conducted in three global regions and used both active and passive surveillance to identify cases of influenza, with analysis of more than 95% of samples from children with suspected cases. Other important strengths were the end points we selected and the use of an rt-PCR assay to confirm the presence of influenza virus. Selection of appropriate end points is a

Table 3. Safety Outcomes in the Total Vaccinated Cohort.*

Outcome	QIV Group N=2584	Control Group N=2584
	no. of children (%)	
Injection-site symptom during 7-day postvaccination period†		
Pain	1215 (47.7)	888 (34.8)
Redness	17 (0.7)	5 (0.2)
Swelling	46 (1.8)	10 (0.4)
Symptom during 28-day postvaccination period‡		
Any	843 (32.6)	855 (33.1)
Related to vaccine	30 (1.2)	37 (1.4)
Medically attended event during entire study period§		
Any	792 (30.7)	749 (29.0)
Related to vaccine	6 (0.2)	13 (0.5)
Serious adverse event during entire study period¶		
Any	36 (1.4)	24 (0.9)
Related to vaccine‖	1 (<0.1)	0
Grade 3**	3 (0.1)	1 (<0.1)

* Safety end points were analyzed descriptively. No significant difference was observed between the incidence of adverse events after the first dose of vaccine and the incidence after the second dose.

† The percentages were calculated on the basis of 2546 children in the QIV group and 2551 children in the control group whose parents or guardians completed a symptom diary. Reports of injection-site symptoms were solicited (i.e., specific questions were included in the daily diary parents were to complete). All injection-site symptoms were considered to be related to vaccination.

‡ Symptoms during the 28-day period after vaccination were reported spontaneously (i.e., not in response to specific questions) in the daily diary.

§ Included were hospitalization, emergency department visit, and visit to physician, nurse practitioner, or other health care worker.

¶ No serious adverse event occurred at an incidence higher than 0.2% (see Table S11 in the Supplementary Appendix for more details).

‖ There was one serious case of bronchitis in the QIV group.

** There was one case of bronchitis and one case of convulsion in the QIV group and one case of drowning in each group.

major challenge for influenza vaccine trials. We chose rt-PCR–confirmed influenza as the primary end point because rt-PCR assay has been shown to be associated with higher detection rates than conventional methods such as serologic testing and cell culture.[13,30-33] As noted above, we included the prevention of moderate-to-severe influenza as an end point because we believe that the most important effect of influenza vaccination lies in the prevention of moderate-to-severe disease. Ideally, trials of vaccine efficacy would capture separately both the most severe cases (severe lower respiratory tract illness and serious complications) and cases that are less severe but nevertheless concern parents and have an effect on utilization of health care resources. Unfortunately, such trials would require the enrollment of impractically large numbers of participants. We therefore chose a dichotomous classification of influenza: mild and not mild (i.e., moderate-to-severe). We believe that this is a valid classification because it distinguishes between influenza that causes mild upper respiratory tract illness and low-grade fever from more severe illness that may have adverse clinical consequences and may increase the utilization of health care resources. Moreover, it is a practical classification with respect to the sample size needed for vaccine trials. Because of this relatively smaller sample size, however, the number of children in the study who had lower respiratory tract illness was small (10 children, all in the control group).

In conclusion, the QIV was shown to be efficacious in preventing influenza A and B in children 3 to 8 years of age. Given the problem of poor vaccine efficacy against mismatched B lineages, the QIV will be of greatest value during seasons in which both lineages are circulating or in the event that there is an unexpected shift from one lineage to another. The efficacy of the vaccine was higher against moderate-to-severe disease — a potentially important end point associated with the highest clinical, social, and economic burden — than against illness of any severity. These results highlight the potential clinical benefit of administering inactivated influenza vaccines in healthy children.

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APPENDIX

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