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Immunogenicity of a Monovalent 2009 Influenza A(H1N1) Vaccine in Infants and Children
A Randomized Trial

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T HE DISEASE BURDEN OF SEA-sonal influenza in the pediatric population is generally attributed to a combination of immunologic naivety, prolonged virus shedding, and enhanced transmission opportunity in child-care and educational institutions.[1] Consistent with this experience, initial reports of the introduction and indigenous transmission of 2009 influenza A(H1N1) infection in many countries have largely involved children,[2] often attending day or boarding schools.[3-5]

Serosurveys have demonstrated little or no cross-reactivity of the pediatric sera sample to the new virus strain, under-

For editorial comment see p 73.

Context In the ongoing influenza pandemic, a safe and effective vaccine against 2009 influenza A(H1N1) is needed for infants and children.

Objective To assess the immunogenicity and safety of a 2009 influenza A(H1N1) vaccine in children.

Design, Setting, and Participants Randomized, observer-blind, age-stratified, parallel group study assessing 2 doses of an inactivated, split-virus 2009 influenza A(H1N1) vaccine in 370 healthy infants and children aged 6 months to less than 9 years living in Australia.

Intervention Intramuscular injection of 15 µg or 30 µg of hemagglutinin antigen dose of monovalent, unadjuvanted 2009 influenza A(H1N1) vaccine in a 2-dose regimen, administered 21 days apart.

Main Outcome Measures Hemagglutination inhibition assay to estimate the proportion of participants with antibody titers of 1:40 or greater, seroconversion, or a significant antibody titer increase, and factor increase in geometric mean titer. Assessments of solicited adverse events during 7 days and unsolicited adverse events for 21 days after each vaccination.

Results Following the first dose of vaccine, antibody titers of 1:40 or greater were observed in 161 of 174 infants and children in the 15-µg group (92.5%; 95% confidence interval [CI], 87.6%-95.6%) and in 168 of 172 infants and children in the 30-µg group (97.7%; 95% CI, 94.2%-99.1%). Corresponding seroconversion rates were 86.8% (95% CI, 80.9%-91.0%) and 94.2% (95% CI, 89.6%-96.8%), and factor increases in geometric mean titer were 13.6 (95% CI, 11.8-15.6) and 18.3 (95% CI, 15.7-21.4). All participants demonstrated antibody titers of 1:40 or greater after the second vaccine dose. Immune responses were robust regardless of age, baseline serostatus, or seasonal influenza vaccination status. The majority of adverse events were mild to moderate in severity.

Conclusion One 15-µg dose of vaccine was immunogenic in infants and children starting at 6 months of age and vaccine-associated reactions were mild to moderate in severity.

Trial Registration clinicaltrials.gov Identifier: NCT00940108

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lining the susceptibility of young children, which was further evidenced in the high hospitalization rates of those younger than 5 years in the current pandemic.[6] The effectiveness of influenza vaccination in children to reduce their infection rate, as well as transmission among household members and the community, has been well demonstrated.[7-10] Several modeling analyses indicate that targeted mass immunization of children will contribute to optimal control of the H1N1 influenza pandemic.[11-13] The US Advisory Committee on Immunization Practices includes individuals aged 6 months to 24 years among the 5 initial target groups for H1N1 immunization.[14]

The Advisory Committee on Immunization Practices also currently recommends that infants and children aged 9 years or younger receive 2 doses of H1N1 influenza vaccine at least 21 days apart, based on existing experience with seasonal trivalent influenza vaccines in this age group.[14] Given the novelty of this new pandemic strain and the uncertainty of the immune response of children to it, it is possible that a higher antigen dose is required to elicit an adequate response.

We conducted a randomized clinical trial of an inactivated, split-virus 2009 influenza A(H1N1) vaccine given as a 2-injection regimen 21 days apart in doses of either 15 µg or 30 µg of hemagglutinin antigen in infants and children between the ages of 6 months and younger than 9 years.

METHODS

This phase 2, multicenter, randomized, observer-blind, parallel-group pediatric study was conducted at 5 Australian centers (Murdoch Children's Research Institute, Melbourne; Princess Margaret Hospital for Children, Perth; Royal Children's Hospital, Brisbane; Children's Hospital at Westmead, Sydney; and Women's and Children's Hospital, Adelaide). The purpose of this study was to evaluate the immunogenicity and safety of 2 different doses of H1N1 vaccine administered as a 2-dose regimen in 2 cohorts of healthy infants and children (aged 6 months to <3 years and aged 3 years to <9 years).

The randomization code was prepared by a statistician (employed by CSL Limited, Parkville, Australia), using SAS software version 9.1.3 (SAS Institute Inc, Cary, North Carolina) and JMP version 8.0.1 (SAS Institute Inc) and a permuted-block randomization within strata. The randomization code was provided to the unblinded vaccine administrators in sealed envelopes. All participants, investigators, and other site personnel involved in clinical assessments were blinded to treatment allocations.

The study was conducted in accordance with the principles of the Declaration of Helsinki, Good Clinical Practice (as defined by the International Conference on Harmonisation), and Australian regulatory requirements. The human research ethics committee at each study center approved the study and written informed consent was obtained from the parent or guardian of the infant or child.

The H1N1 vaccine, a monovalent, unadjuvanted, thimerosal-free, inactivated, split-virion vaccine was produced by CSL Biotherapies (Parkville, Australia) as previously described (but without the addition of thimerosal).[15] The vaccine was presented in 0.5-mL prefilled syringes containing 30 µg of hemagglutinin antigen per 0.5-mL dose. The 2 doses were 15 µg of hemagglutinin antigen per 0.25-mL dose and 30 µg of hemagglutinin antigen per 0.5-mL dose.

Healthy infants and children aged 6 months to younger than 9 years were eligible for enrollment if they were born at or after 36 weeks' gestation (applying only to children <3 years of age) and able to provide venous blood samples. Children with confirmed or suspected 2009 influenza A(H1N1) infection or those who had received an experimental vaccine during the preceding 6 months were excluded from the study. Race was included as a variable in this study to permit judgment of external validity (generalizability).

Eligible children were stratified by age at the time of the first study vaccination to 1 of 2 groups for infants and children aged 6 months to younger than 3 years and those aged 3 years to younger than 9 years (**FIGURE 1**). After stratification, children were randomized in a 1:1 ratio to receive either the 15-µg or 30-µg dose. Vaccines were administered by intramuscular injection into either the anterolateral aspect of the thigh (participants aged <12 months) or the deltoid muscle (participants aged ≥12 months). Because the injection volume differed between the 2 study doses, those administering the vaccine had no further involvement in the study.

Solicited local and systemic adverse events were recorded by the child's parent or guardian using a 7-day diary card, and unsolicited adverse events were recorded using a 21-day diary card. All solicited local adverse events were considered related to the H1N1 vaccine, whereas the investigator determined the causality of solicited systemic adverse events and unsolicited adverse events. Irritability and appetite loss were assessed only in infants and children younger than 3 years; while headache, malaise, and myalgia were collected only from children aged 3 years or older. Because of the novelty of the H1N1 strain, we prospectively collected data relating to the occurrence of several adverse events of special interest, including nervous system disorders (eg, Bell palsy, Guillain-Barré syndrome), immune system disorders, and other disorders.

An independent data and safety monitoring board monitored the safety of the study. Stopping rules were in place during the 7 days after vaccination.

Nasal and throat swabs were collected from participants reporting an influenzalike illness. An influenzalike illness was defined as an axillary temperature of more than 37.5°C or a clear history of fever or chills, at least 1 influenzalike symptom, and an onset of symptoms at least 72 hours after vaccination.

Hemagglutination inhibition antibody titers to the H1N1 antigen in the study vaccine were measured at enrollment and 21 to 25 days after vaccination, using methods previously described.[15] Virological testing of nasal-swab and throat-swab specimens was performed using methods previously described.[15] Focus Diagnostics Incorporated (Cypress, California) performed all laboratory assays.

The 3 coprimary immunogenicity end points were based on the international criteria used to evaluate influenza vaccines in adults aged 18 to 60 years.[16] These end points were the proportion of participants with hemagglutination inhibition antibody titers of 1:40 or greater[17]; the proportion of participants achieving either seroconversion (a prevaccination titer of <1:10 with a postvaccination titer of ≥1:40) or a sig-

nificant increase (by a factor of ≥4) in antibody titer; and the factor increase in geometric mean titer. Secondary (safety) end points were the frequency, duration, and intensity of solicited adverse events during the 7 days after vaccination; unsolicited adverse events during the 21 days after vaccination; and the incidence of adverse events deemed serious or of special interest reported during the study period.

Figure 1. Participant Disposition
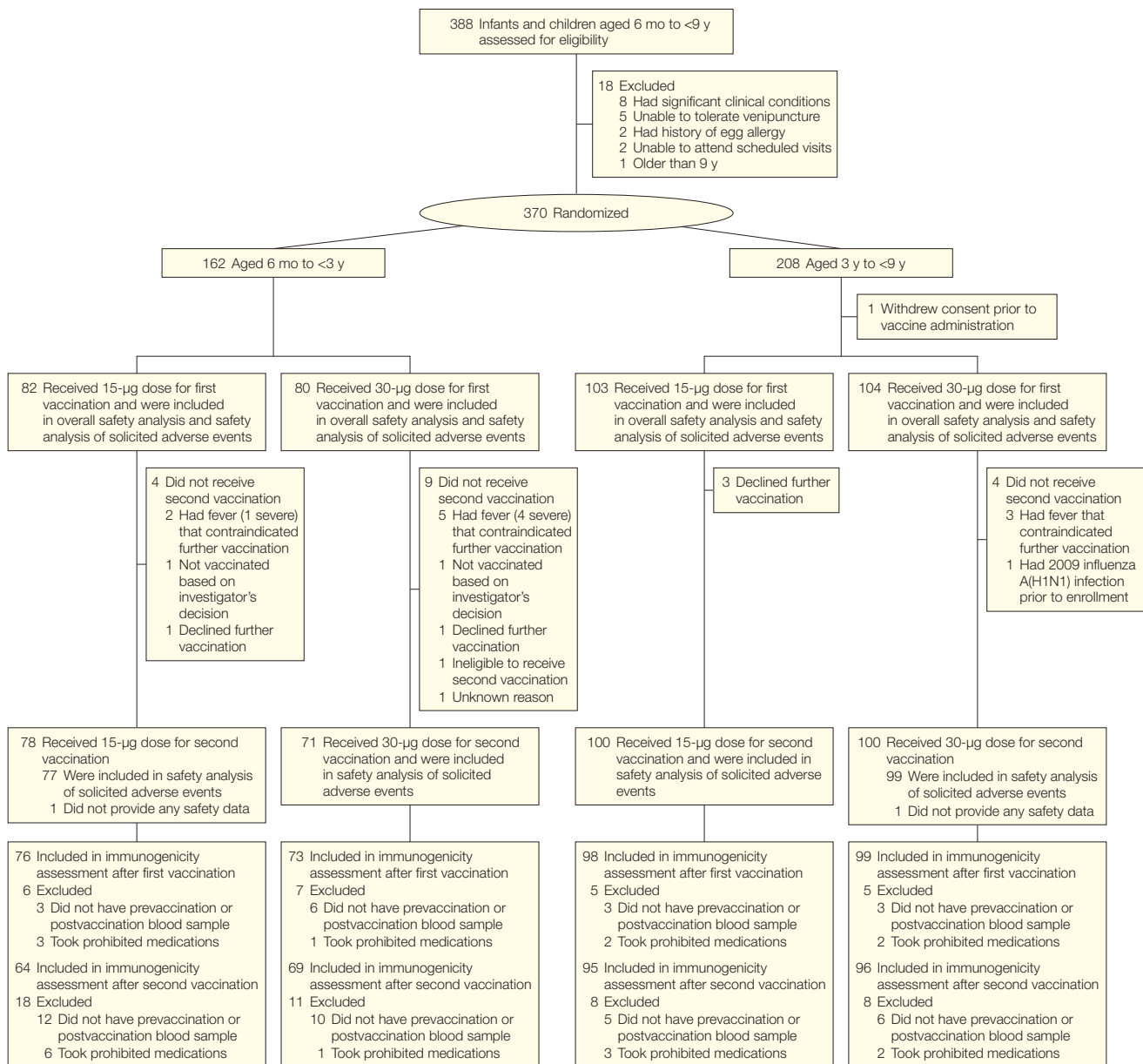
A sample size of 100 participants per study group (400 total) was estimated to provide sufficient power to compare each of the 3 coprimary end points between the 2 dose groups. A between-group difference in seroprotection or seroconversion rates in the order of 20% or a factor increase in geometric mean titers of 1.75 to 1.90 could be distinguished with between 80% and 90% power.

The primary and secondary end point analyses were descriptive with calculation of 2-sided 95% confidence intervals (CIs), assuming binomial distributions for dichotomous variables and log normal distribution for hemagglu-tination inhibition titers. For categorical variables, statistical summaries included counts and percentages relative to the appropriate dose group and age cohort and these were compared using the Fisher exact test. All tests were 2-sided with an α level of .05.

The safety population comprised all randomized participants who received a dose of study vaccine and provided safety data. The evaluable population comprised all vaccinated participants who provided baseline and postvaccination sera samples and did not take a prohibited medication or have a laboratory-confirmed 2009 influenza A(H1N1) infection during the study period. The intention-to-treat (ITT) population included all infants and children who were randomized. The primary analysis was based on the evaluable population. To verify the robustness of these results, a secondary analysis was performed on the ITT population, in which missing hemagglutination inhibition titers were imputed using last observation carried forward if baseline titers were available or the lower limit of detection (hemagglutination inhibition titer of 1:5) of the assay if baseline titers also were missing.

Table 1. Participant Demographics

Characteristic	15-µg Vaccine Dose (n = 185)			30-µg Vaccine Dose (n = 184)			All Participants (N = 369)		
	Age <3 y (n = 82)	Age ≥3 y (n = 103)	Total (n = 185)	Age <3 y (n = 80)	Age ≥3 y (n = 104)	Total (n = 184)	Age <3 y (n = 162)	Age ≥3 y (n = 207)	Total (N = 369)
Age, mean (SD) [median], y	1.7 (0.7) [1.6]	5.8 (1.7) [5.7]	4 (2.4) [3.6]	1.7 (0.7) [1.8]	5.7 (1.7) [5.6]	4 (2.4) [3.4]	1.7 (0.7) [1.7]	5.7 (1.7) [5.7]	4 (2.4) [3.5]
Sex, No. (%)									
Male	41 (50)	50 (49)	91 (49)	39 (49)	55 (53)	94 (51)	80 (49)	105 (51)	185 (50)
Female	41 (50)	53 (51)	94 (51)	41 (51)	49 (47)	90 (49)	82 (51)	102 (49)	184 (50)
Race, No. (%)									
White	72 (88)	97 (94)	169 (91)	74 (93)	97 (93)	171 (93)	146 (90)	194 (94)	340 (92)
Other[a]	10 (12)	6 (6)	16 (9)	6 (7)	7 (7)	13 (7)	16 (10)	13 (6)	29 (8)
Received seasonal influenza vaccine, No. (%)[b]	35 (43)	47 (46)	82 (44)	27 (34)	39 (38)	66 (36)	62 (38)	86 (42)	148 (40)

[a] Defined as Asian, African, black, or other.
[b] Indicates participants who received the 2009 Southern Hemisphere seasonal trivalent influenza vaccine, which contained 15 µg of hemagglutinin antigen of each of the following strains: A/Brisbane/59/2007 (H1N1), A/Uruguay/716/2007 (H3N2), and B/Florida/4/2006 (B).

Table 2. Immune Responses After the First and Second Vaccinations With 2009 Influenza A(H1N1) Vaccine as Measured by the Hemagglutination Inhibition (HI) Assay

	HI Titer ≥1:40 No./Total; % (95% CI)			Postvaccination Seroconversion or Increase, No./Total; % (95% CI)		Geometric Mean Titer, (95% CI)			Postvaccination Geometric Mean Fold Increase, (95% CI)	
		Postvaccination					Postvaccination			
	Baseline[a]	First	Second	First	Second	Baseline[a]	First	Second	First	Second
Age <3 y										
15 µg	7/76; 9.2 (4.5-17.8)	70/76; 92.1 (83.8-96.3)	64/64; 100 (94.3-100)	67/76; 88.2 (79.0-93.6)	62/64; 96.9 (89.3-99.1)	8.1 (6.2-10.6)	113.1 (87.1-146.9)	517.2 (413.0-647.8)	14.0 (11.4-17.1)	57.6 (42.9-77.5)
30 µg	10/73; 13.7 (7.6-23.4)	73/73; 100 (95.0-100)[b]	69/69; 100 (94.7-100)	71/73; 97.3 (90.5-99.2)	68/69; 98.6 (92.2-99.7)	8.1 (6.1-10.6)	178.7 (139.1-229.7)[b]	610.7 (496.9-750.6)	22.2 (17.9-27.5)[b]	72.9 (56.1-94.8)
Age ≥3 y										
15 µg	27/98; 27.6 (19.7-37.1)	91/98; 92.9 (86.0-96.5)	95/95; 100 (96.1-100)	84/98; 85.7 (77.4-91.3)	93/95; 97.9 (92.6-99.4)	15.2 (11.3-20.3)	201.1 (155.8-259.6)	546.4 (450.0-663.5)	13.2 (10.8-16.2)	37.5 (28.3-49.6)
30 µg	33/99; 33.3 (24.8-43.1)	95/99; 96.0 (90.1-98.4)	96/96; 100 (96.2-100)	91/99; 91.9 (84.9-95.8)	93/96; 96.9 (91.2-98.9)	16.8 (12.7-22.4)	268.0 (204.6-351.0)	652.4 (542.9-784.0)	15.9 (12.9-19.7)	37.1 (28.8-47.7)
All										
15 µg	34/174; 19.5 (14.3-26.1)	161/174; 92.5 (87.6-95.6)	159/159; 100 (97.6-100)	151/174; 86.8 (80.9-91.0)	155/159; 97.5 (93.7-99.0)	11.5 (9.4-14.2)	156.4 (129.8-188.5)	534.5 (462.0-618.3)	13.6 (11.8-15.6)	44.6 (36.3-54.8)
30 µg	43/172; 25.0 (19.1-32.0)	168/172; 97.7 (94.2-99.1)[b]	165/165; 100 (97.7-100)	162/172; 94.2 (89.6-96.8)[b]	161/165; 97.6 (93.9-99.1)	12.3 (10.0-15.1)	225.7 (186.8-272.6)[b]	634.6 (553.9-727.1)	18.3 (15.7-21.4)[b]	49.2 (40.7-59.4)
Total	77/346; 22.3 (18.2-26.9)	329/346; 95.1 (92.3-96.9)	324/324; 100 (98.8-100)	313/346; 90.5 (86.9-93.1)	316/324; 97.5 (95.2-98.7)	11.9 (10.3-13.8)	187.7 (164.2-214.5)	583.3 (528.1-644.3)	15.7 (14.2-17.5)	46.9 (40.8-53.8)

Abbreviation: CI, confidence interval.
[a] Baseline values are based on the evaluable population for the first vaccination.
[b] $P<.05$ compared with 15-µg dose.

RESULTS

Between August 3 and September 4, 2009, 370 participants were enrolled and randomized to treatment (**TABLE 1** and Figure 1). Recruitment was completed with numbers slightly less than originally planned because of slower than expected recruitment rates in younger children. Due to the public health urgency and need for dosing data in infants and children, enrollment was terminated with 162 infants and children younger than 3 years and 208 children aged 3 years or older (1 participant in the older age group withdrew consent prior to the first vaccination). Although fewer participants were recruited than planned, enrollment was sufficient to allow analysis with adequate precision of the immunogenicity and safety data generated.

Overall, 369 participants who received the 2009 influenza A(H1N1) vaccine comprised the safety population. Because 15 children did not provide either a baseline or postvaccination sera sample and 8 children took prohibited medications during the assessment period, the evaluable population after the first vaccination dose was 346 participants (<3 years: 149 participants; ≥3 years: 197 participants), and was 324 (<3 years: 133 participants; ≥3 years: 191 participants) after the second vaccination dose.

Overall, 148 of 369 vaccinated participants (40.1%) reported having received a 2009 Southern Hemisphere seasonal trivalent inactivated vaccine (TIV), with similar coverage observed in each of the age and dose cohorts (Table 1). Participants recruited from geographic locations within Australia with declining reporting rates for H1N1 influenza[18] accounted for 224 of 369 vaccinated participants (60.1%).

Immunogenicity

Baseline proportions of participants with antibody titers of 1:40 or greater were higher among children 3 years of age or older (27.6% and 33.3%) than those younger than 3 years (9.2% and 13.7%) in the 15-µg and 30-µg dose groups, respectively (**TABLE 2**). Vacci-

nation with the 2009 seasonal TIV was significantly negatively associated with having baseline antibody titers of 1:40 or greater (no 2009 TIV: 25.8% [95% CI, 20.4%-32.2%] vs 2009 TIV: 16.8% [95% CI, 11.5%-23.9%]; 2-tailed Fisher exact test, *P* = .049).

Overall, 329 of 346 participants (95.1%; 95% CI, 92.3%-96.9%) had antibody titers of 1:40 or greater after a single H1N1 vaccination (Table 2). Following the first dose of vaccine, antibody titers of 1:40 or greater were

observed in 161 of 174 infants and children in the 15-µg group (92.5%; 95% CI, 87.6%-95.6%) and in 168 of 172 infants and children in the 30-µg group (97.7%; 95% CI, 94.2%-99.1%). Corresponding seroconversion rates were 86.8% (95% CI, 80.9%-91.0%) and 94.2% (95% CI, 89.6%-96.8%), and factor increases in geometric mean titers were 13.6 (95% CI, 11.8-15.6) and 18.3 (95% CI, 15.7-21.4). Responses were similar in the younger age group compared with older

Figure 2. Reverse Cumulative Distribution Curves of Antibody Titers on Hemagglutination Inhibition Assay by Vaccine Dose
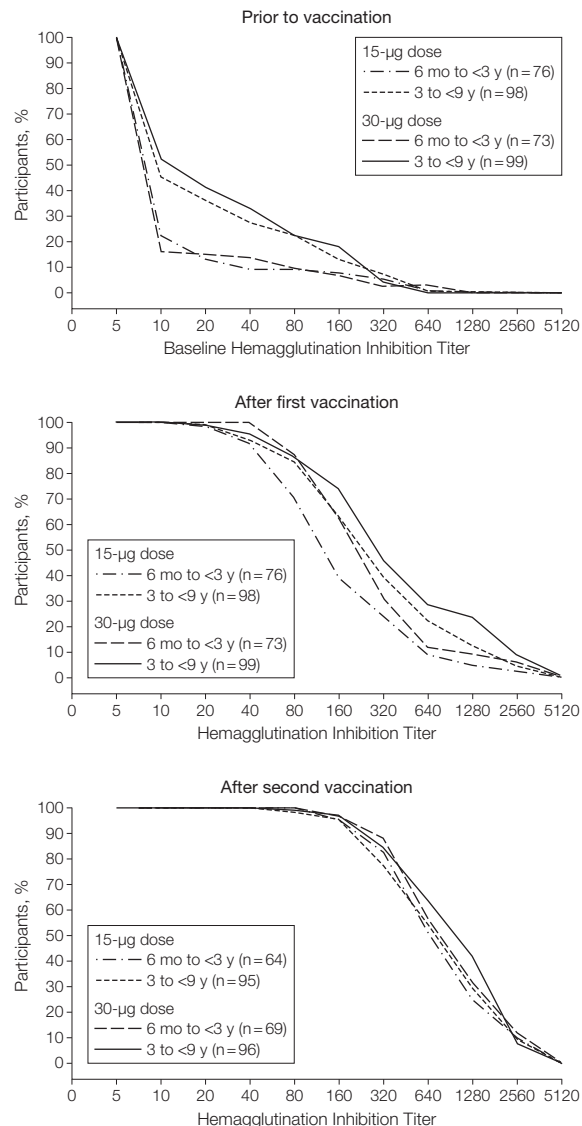
children; however, the factor increase in geometric mean titer was higher in the older children (Table 2 and **FIGURE 2**). Responses following the second vaccine dose were even more vigorous, with all participants in both age cohorts and dose groups achieving titers of 1:40 or greater (Table 2 and Figure 2).

The ITT analyses (with imputation for missing values) confirmed the robustness of these results with 341 of 370 randomized children (92.2%;
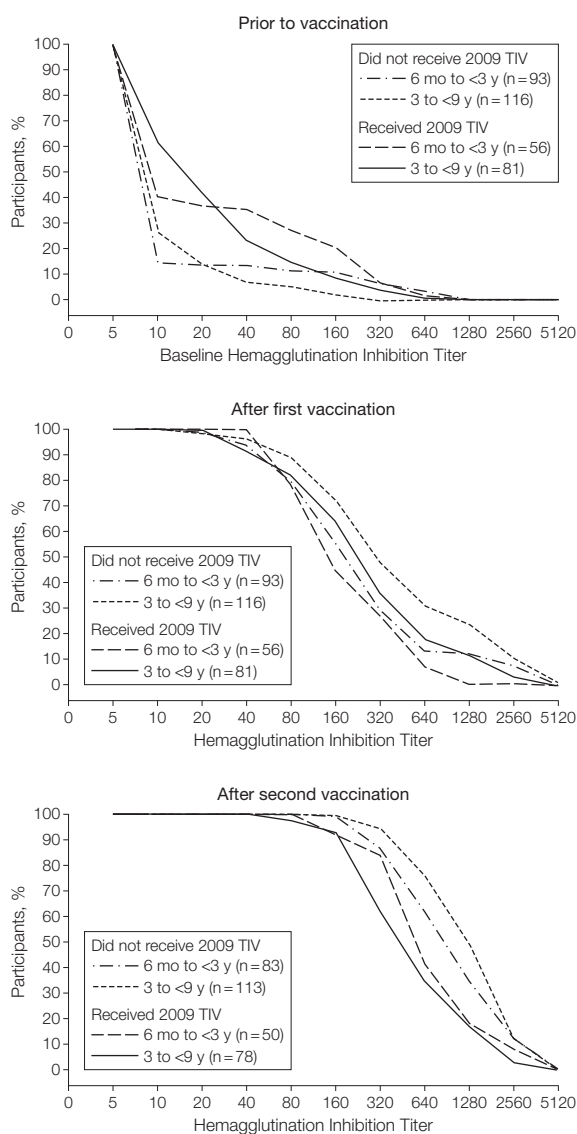
95% CI, 89%-94.5%) achieving antibody titers of 1:40 or greater after a single H1N1 vaccination and 321 of 370 of participants (86.8%; 95% CI, 82.9%-89.8%) achieving seroconversion or significant increase. The factor increase in geometric mean titer was 14.2 (95% CI, 12.6-15.9). For the second dose, the corresponding ITT results were 361 of 370 (97.6%; 95% CI, 95.4%-98.7%), 352 of 370 (95.1%; 95% CI, 92.4%-96.9%), and 39.8 (95% CI, 34.6-45.9).

Children who had baseline antibody titers of 1:40 or greater exhibited significantly smaller factor increases in titer following vaccination compared with those with baseline titer of less than 1:40. Of those who had baseline titers of less than 1:10, more than 92.7% (95% CI, 88.5%-95.5%) achieved seroconversion after 1 dose of vaccine.

Those who had received a 2009 TIV tended to have lower postvaccination titers (152.1; 95% CI, 126.4-183.0) than those not reporting 2009 TIV (215.4; 95% CI, 179.3-258.8; *P*=.01) after the first dose (**FIGURE 3**). The effect persisted after the second dose (Figure 3).

A total of 25 influenzalike illness episodes were reported and assessed during the period between receipt of the first and second vaccine dose, none of which was positive for influenza. A further 19 influenzalike illnesses were reported after the second dose and prior to the second postdose blood sample. None of these was positive for influenza.

Adverse Events

No deaths were reported. There were 2 serious adverse events. The first was a significant unexpected event of a 4-day episode of fluctuating fever (to 39.7°C) with onset within 24 hours of the first vaccination (30-µg dose) in an 8-year-old child, briefly halting enrollment. The investigator considered that this event was possibly related to vaccination. The data and safety monitoring board attributed the episode to a possible viral infection and the study was recommenced within 1 day of notification. This child made a full and uneventful recovery; laboratory evaluation did not identify a specific etiology. The second serious adverse event occurred in a 1-year-old child after the second vaccination (15-µg dose). This adverse event was diagnosed clinically as viral gastroenteritis and was considered unrelated to vaccination.

Two further adverse events of special interest considered unrelated to

Figure 3. Reverse Cumulative Distribution Curves of Antibody Titers on Hemagglutination Inhibition Assay by Prior Receipt of 2009 TIV

TIV indicates 2009 Southern Hemisphere seasonal trivalent inactivated vaccine.

the vaccine were reported. The first of these was a 2-year-old boy with persistent fever, with onset 4 days after the first immunization with the 30-µg dose. Differential diagnosis included Kawasaki disease and viral or bacterial infection. There was no coronary vascular involvement on the echocardiogram. The second case event was an 18-month-old boy with a single febrile convulsion in association with concurrent pneumonia with onset 20 days following receipt of the first dose of H1N1 vaccine (15-µg dose).

Solicited local adverse events reported within 7 days of vaccination are summarized in **TABLE 3**. Detailed data by age group and vaccine dose are available online (supplementary eTable 1 and eTable 2 are available at http://www.jama.com). Following the first vaccination, no local adverse event was reported by 42.5% of participants overall, with 45% reporting mild adverse events, 10% reporting moderate, and 2.4% reporting severe events. Pain at the injection site was the most commonly reported event.

Systemic adverse events following vaccination are shown in **TABLE 4**. The most common reactions in infants and younger children were fever and irritability. After the first vaccination, severe postimmunization fever (>39.5°C) was reported by 1 of 82 participants in those younger than 3 years (1.2%; 95% CI, 0.2%-6.6%) in the 15-µg dose group and 4 of 79 children in the same age group (5.1%; 95% CI, 2%-12.3%) who received the 30-µg dose (eTable 1). In children aged 3 years or older, none experienced severe fever in the 15-µg group compared with 3 of 104 in the 30-µg group (2.9%; 95% CI, 1.0%-8.1%). Irritability of a severe degree was reported in only 1 participant in each dose group in the younger group after the first vaccination, and severe nausea or vomiting in a further participant in the 30-µg group in the same age group.

There were no severe fevers following the second dose in the 15-µg group in either age group compared with 2 of 71 infants and children younger than 3 years in the 30-µg group (2.8%; 95% CI, 0.8%-9.7%). There were no febrile

convulsions after the second vaccination dose. Overall, the pattern of adverse events following the second vaccination dose was similar to that following the first dose. Reporting of unsolicited clinical events for 21 days after each vaccination dose produced no remarkable findings (data available at http://www.jama.com as supplementary eTable 3).

COMMENT

This study demonstrates that a single 15-µg dose of this monovalent 2009 influenza A(H1N1) vaccine is sufficient to induce hemagglutination inhibition antibody titers of 1:40 or greater in 92.5% of children, including those as young as 6 months. A second vaccine dose resulted in every child achieving a protective threshold, with substantial geometric mean titers and robust factor increases in antibody titers. Furthermore, vaccine tolerability was as expected and the reactogenicity profile is in line with that of seasonal trivalent influenza vaccines.[19]

These findings have important public health implications given that young

Table 3. Participants Reporting Solicited Adverse Events Within 7 Days After Each Vaccination

Solicited Local Adverse Events[a]	Maximum Intensity, No./Total; % (95% CI)							
	None		Mild		Moderate		Severe	
	15 µg	30 µg	15 µg	30 µg	15 µg	30 µg	15 µg	30 µg
Any								
First vaccination	74/185; 40 (33.2-47.2)	83/184; 45.1 (38.1-52.3)	91/185; 49.2 (42.1-56.3)	75/184; 40.8 (33.9-48.0)	19/185; 10.3 (6.7-15.5)	18/184; 9.8 (6.3-14.9)	1/185; 0.5 (0.1-3.0)	8/184; 4.3 (2.2-8.3)
Second vaccination	91/177; 51.4 (44.1-58.7)	77/170; 45.3 (38.0-52.8)	66/177; 37.3 (30.5-44.6)	68/170; 40 (32.9-47.5)	17/177; 9.6 (6.1-14.8)	22/170; 12.9 (8.7-18.8)	3/177; 1.7 (0.6-4.9)	3/170; 1.8 (0.6-5.1)
Pain								
First vaccination	101/185; 54.6 (47.4-61.6)	109/184; 59.2 (52.0-66.1)	78/185; 42.2 (35.3-49.4)	64/184; 34.8 (28.3-41.9)	5/185; 2.7 (1.2-6.2)	10/184; 5.4 (3.0-9.7)	1/185; 0.5 (0.1-3.0)	1/184; 0.5 (0.1-3.0)
Second vaccination	115/177; 65.0 (57.7-71.6)	97/170; 57.1 (49.5-64.3)	51/177; 28.8 (22.6-35.9)	60/170; 35.3 (28.5-42.7)	11/177; 6.2 (3.5-10.8)	12/170; 7.1 (4.1-11.9)	0/177	1/170; 0.6 (0.1-3.3)
Redness								
First vaccination	123/185; 66.5 (59.4-72.9)	134/184; 72.8 (66.0-78.7)	52/185; 28.1 (22.1-35.0)	38/184; 20.7 (15.4-27.1)	10/185; 5.4 (3.0-9.7)	9/184; 4.9 (2.6-9.0)	0/185	3/184; 1.6 (0.6-4.7)
Second vaccination	123/177; 69.5 (62.4-75.8)	125/170; 73.5 (66.4-79.6)	46/177; 26.0 (20.1-32.9)	32/170; 18.8 (13.7-25.4)	5/177; 2.8 (1.2-6.4)	12/170; 7.1 (4.1-11.9)	3/177; 1.7 (0.6-4.9)	1/170; 0.6 (0.1-3.3)
Swelling or lump								
First vaccination	154/185; 83.2 (77.2-87.9)	146/184; 79.3 (72.9-84.6)	23/185; 12.4 (8.4-18.0)	23/184; 12.5 (8.5-18.1)	8/185; 4.3 (2.2-8.3)	10/184; 5.4 (3.0-9.7)	0/185	5/184; 2.7 (1.2-6.2)
Second vaccination	141/177; 79.7 (73.1-84.9)	136/170; 80.0 (73.4-85.3)	31/177; 17.5 (12.6-23.8)	24/170; 14.1 (9.7-20.1)	5/177; 2.8 (1.2-6.4)	8/170; 4.7 (2.4-9.0)	0/177	2/170; 1.2 (0.3-4.2)

[a] Coded with 4 grades of intensity for erythema and induration across both age cohorts: none (0 mm), mild (<10 mm), moderate (≥10 mm to ≤30 mm), or severe (>30 mm). Pain for those younger than 3 years was graded as none (absent), mild (minor reaction on touch), moderate (cries or protests on touch), or severe (cries when limb is moved or spontaneously painful). Pain for those aged 3 years or older was graded as none (absent), mild (does not interfere with daily activities), moderate (interferes with daily activities), or severe (prevents daily activities).

children are at the highest risk for hospitalization and requirement for intensive care.[20] The results are of particular added clinical significance because of the unexpected finding of the possible adequacy of a single dose given that the US and UK governments recommend a 2-dose regimen in infants and young children.

Similar to that observed in a previously reported adult trial of this vaccine,[15] a higher than expected proportion of children exhibited baseline antibody titers greater than 1:40. This finding was particularly notable among children aged 3 years or older. While we excluded individuals with confirmed H1N1 infection, this finding may still represent previous subclinical exposure because nearly two-thirds of study participants were recruited from geographic areas in which 2009 influenza A(H1N1) infection notifications had already started to decline.[18] We found that there was a diminution of response in those with higher baseline titers, prior TIV exposure, or both, sug-

Table 4. Participants Reporting Solicited Systemic Adverse Events Within 7 Days After Each Vaccination

Solicited Systemic Adverse Events[a]	Maximum Intensity, No./Total; % (95% CI)							
	None		Mild		Moderate		Severe	
	15 µg	30 µg	15 µg	30 µg	15 µg	30 µg	15 µg	30 µg
Any								
First vaccination	89/185; 48.1 (41.0-55.3)	62/184; 33.7 (27.3-40.8)	59/185; 31.9 (25.6-38.9)	62/184; 33.7 (27.3-40.8)	35/185; 18.9 (13.9-25.2)	50/184; 27.2 (21.3-34.0)	2/185; 1.1 (0.3-3.9)	10/184; 5.4 (3.0-9.7)
Second vaccination	96/177; 54.2 (46.9-61.4)	96/170; 56.5 (49.0-63.7)	56/177; 31.6 (25.2-38.8)	55/170; 32.4 (25.8-39.7)	24/177; 13.6 (9.3-19.4)	17/170; 10.0 (6.3-15.4)	1/177; 0.6 (0.1-3.1)	2/170; 1.2 (0.3-4.2)
Diarrhea								
First vaccination	160/185; 86.5 (80.8-90.7)	154/184; 83.7 (77.7-88.3)	20/185; 10.8 (7.1-16.1)	25/184; 13.6 (9.4-19.3)	5/185; 2.7 (1.2-6.2)	5/184; 2.7 (1.2-6.2)	0/185	0/184
Second vaccination	164/177; 92.7 (87.8-95.7)	156/170; 91.8 (86.7-95.0)	8/177; 4.5 (2.3-8.7)	12/170; 7.1 (4.1-11.9)	4/177; 2.3 (0.9-5.7)	2/170; 1.2 (0.3-4.2)	1/177; 0.6 (0.1-3.1)	0/170
Fever[b]								
First vaccination	140/184; 76.1 (69.4-81.7)	108/183; 59.0 (51.8-65.9)	30/184; 16.3 (11.7-22.3)	42/183; 23.0 (17.5-29.6)	13/184; 7.1 (4.2-11.7)	26/183; 14.2 (9.9-20.0)	1/184; 0.5 (0.1-3.0)	7/183; 3.8 (1.9-7.7)
Second vaccination	144/176; 81.8 (75.5-86.8)	144/168; 85.7 (79.6-90.2)	26/176; 14.8 (10.3-20.8)	19/168; 11.3 (7.4-17.0)	6/176; 3.4 (1.6-7.2)	3/168; 1.8 (0.6-5.1)	0/176	2/168; 1.2 (0.3-4.2)
Nausea or vomiting								
First vaccination	169/185; 91.4 (86.4-94.6)	148/184; 80.4 (74.1-85.5)	13/185; 7.0 (4.2-11.7)	18/184; 9.8 (6.3-14.9)	3/185; 1.6 (0.6-4.7)	16/184; 8.7 (5.4-13.7)	0/185	2/184; 1.1 (0.3-3.9)
Second vaccination	165/177; 93.2 (88.5-96.1)	159/170; 93.5 (88.8-96.3)	6/177; 3.4 (1.6-7.2)	8/170; 4.7 (2.4-9.0)	5/177; 2.8 (1.2-6.4)	3/170; 1.8 (0.6-5.1)	1/177; 0.6 (0.1-3.1)	0/170
Irritability (age <3 y only)								
First vaccination	41/82; 50.0 (39.4-60.6)	30/80; 37.5 (27.7-48.5)	27/82; 32.9 (23.7-43.7)	28/80; 35.0 (25.5-45.9)	13/82; 15.9 (9.5-25.3)	21/80; 26.3 (17.9-36.8)	1/82; 1.2 (0.2-6.6)	1/80; 1.3 (0.2-6.7)
Second vaccination	46/77; 59.7 (48.6-70.0)	39/71; 54.9 (43.4-66.0)	17/77; 22.1 (14.3-32.5)	26/71; 36.6 (26.4-48.2)	13/77; 16.9 (10.1-26.8)	6/71; 8.5 (3.9-17.2)	1/77; 1.3 (0.2-7.0)	0/71
Loss of appetite (age <3 y only)								
First vaccination	59/82; 72.0 (61.4-80.5)	49/80; 61.3 (50.3-71.2)	17/82; 20.7 (13.4-30.7)	21/80; 26.3 (17.9-36.8)	6/82; 7.3 (3.4-15.1)	10/80; 12.5 (6.9-21.5)	0/82	0/80
Second vaccination	60/77; 77.9 (67.5-85.7)	54/71; 76.1 (65.0-84.5)	14/77; 18.2 (11.2-28.2)	14/71; 19.7 (12.1-30.4)	2/77; 2.6 (0.7-9.0)	3/71; 4.2 (1.4-11.7)	1/77; 1.3 (0.2-7.0)	0/71
Malaise (age ≥3 y only)								
First vaccination	89/103; 86.4 (78.5-91.7)	81/104; 77.9 (69.0-84.8)	8/103; 7.8 (4.0-14.6)	12/104; 11.5 (6.7-19.1)	6/103; 5.8 (2.7-12.1)	9/104; 8.7 (4.6-15.6)	0/103	2/104; 1.9 (0.5-6.7)
Second vaccination	91/100; 91.0 (83.8-95.2)	89/99; 89.9 (82.4-94.4)	7/100; 7.0 (3.4-13.7)	7/99; 7.1 (3.5-13.9)	2/100; 2.0 (0.6-7.0)	3/99; 3.0 (1.0-8.5)	0/100	0/99
Myalgia (age ≥3 y only)								
First vaccination	93/103; 90.3 (83.0-94.6)	85/104; 81.7 (73.2-88.0)	8/103; 7.8 (4.0-14.6)	14/104; 13.5 (8.2-21.3)	2/103; 1.9 (0.5-6.8)	4/104; 3.8 (1.5-9.5)	0/103	1/104; 1.0 (0.2-5.2)
Second vaccination	90/100; 90.0 (82.6-94.5)	92/99; 92.9 (86.1-96.5)	10/100; 10.0 (5.5-17.4)	6/99; 6.1 (2.8-12.6)	0/100	1/99; 1.0 (0.2-5.5)	0/100	0/99
Headache (age ≥3 y only)								
First vaccination	82/103; 79.6 (70.8-86.3)	83/104; 79.8 (71.1-86.4)	16/103; 15.5 (9.8-23.8)	11/104; 10.6 (6-18)	5/103; 4.9 (2.1-10.9)	9/104; 8.7 (4.6-15.6)	0/103	1/104; 1.0 (0.2-5.2)
Second vaccination	85/100; 85.0 (76.7-90.7)	89/99; 89.9 (82.4-94.4)	13/100; 13.0 (7.8-21.0)	8/99; 8.1 (4.2-15.1)	2/100; 2.0 (0.6-7.0)	2/99; 2.0 (0.6-7.1)	0/100	0/99

[a] Coding for mild, moderate, and severe was the same for all events in both age cohorts: mild (adverse event easily tolerated by the participant, causing minimal discomfort and does not interfere with everyday activities), moderate (adverse event sufficiently discomforting to interfere with everyday activities), or severe (adverse event prevents normal everyday activities and requires significant medical intervention). For fever, the grades were mild (≥37.5°C to ≤38.5°C), moderate (>38.5°C to ≤39.5°C), and severe (>39.5°C).
[b] One participant younger than 3 years in the 30-µg dose group and 1 participant aged 3 years or older in the 15-µg dose group did not provide data on temperature after the first vaccination. Three participants aged 3 years or older (1 in the 15-µg dose group and 2 in the 30-µg dose group) did not provide data on temperature after the second vaccination.

gesting that, if anything, the vaccine response may have been underestimated.

The immune response observed after a single vaccination differs from that of previous studies of seasonal H1 strains in vaccine-naive children and in H5N1 vaccination studies, in which 2 doses are required to induce protective responses.[19,21] The highly immunogenic nature of the 2009 influenza A(H1N1) hemagglutinin and the magnitude of the immune response after a single dose is consistent with that observed in adults,[15,22] and may be related to specific antigen characteristics of this novel virus, which have yet to be determined. Additionally, and particularly in younger children, the robust response may be due in part to twice the antigen strain dose being used compared with each strain in seasonal TIV.

Although we did not include a placebo control group, we are confident that the observed immune responses were the result of the vaccine rather than active infection because our rigorous surveillance for influenzalike illness did not detect a single case of H1N1 infection. Subclinical infection may have occurred, but it is unlikely to have significantly affected the study results.

The data suggest that children who had not received TIV before enrollment were more likely to have baseline hemagglutination inhibition antibody titers of 1:40 or greater, and higher hemagglutination inhibition titer responses to the vaccine strain than those children who had received TIV. This was not previously observed in adults,[15] but has been reported following H5N1 influenza vaccine in children.[21] Insufficient data are currently available to support a consistent effect of prior TIV on either H1N1 immunogenicity or disease risk because early reports are conflicting.[23-25]

This study does not address the immunogenicity of a lower antigen dose or the important issue of whether seroresponses are attenuated in children with chronic disease and immunocompromising conditions or

treatments. Studies exploring these issues are under way.

Both the US[26] and UK[27] governments have to date recommended a 2-dose schedule for pandemic influenza vaccines in children younger than 9 years of age. This requirement poses substantial logistic challenges for those involved in immunization delivery. Furthermore, if 2 doses are necessary for optimal protection, the delay to vaccine effect could substantially reduce likely herd immunity benefits of pediatric vaccination. In China, similar observations of monovalent influenza vaccine immunogenicity have spurred ongoing debate regarding the need for a second vaccine dose at any age.[28,29] Our findings suggest that a single 15-μg dose vaccine regimen may be effective and well tolerated in children, and may have positive implications for disease protection and reduced transmission of pandemic H1N1 in the wider population.

ticle using the methods described in the article. Repeat tabulations for other end points were performed. Results for the adverse events also were replicated. Results were comparable with those obtained by the sponsor, and the results of the independent analyses are the results reported in this article. While there were a few small discrepancies, all were resolved prior to submission of the manuscript for publication and none affected the inferences made in this article. No financial compensation was provided for Dr Carlin or Ms Vidmar. CSL Limited provided no funds to support the independent statistical analysis.

form). These companies were compensated through commercial arrangements with the study's sponsor, CSL Limited.

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