

# Safety and Immunogenicity of 2009 pH1N1 Vaccination in HIV-Infected Pregnant Women

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**Background.** Pregnant women infected with human immunodeficiency virus (HIV) may have particular vulnerability to 2009 pandemic H1N1 influenza (pH1N1) infection. The safety and immunogenicity of pH1N1 vaccination in HIV-infected pregnant women are unknown.

**Methods.** HIV-infected women 18–39 years of age and 14–34 weeks' gestation on antiretroviral therapy received two 30- $\mu$ g doses of unadjuvanted, inactivated pH1N1 vaccine 21 days apart. Hemagglutination inhibition titers were measured at entry, 21 days after dose 1, and 10 and 21 days after dose 2, and, in mothers and infants, at delivery and 3 and 6 months postdelivery.

**Results.** No severe vaccine-related adverse events were observed among 127 subjects. At entry, 21% had seroprotective ( $\geq 1:40$ ) titers. Seroprotection and seroresponse ( $\geq 4$ -fold rise) occurred in 73% and 66% after dose 1 and 80% and 72% after dose 2, respectively. Of women lacking seroprotection at entry, 66% attained seroprotection after dose 1 and 75% after dose 2. Seroprotective titers were present in 67% of mothers and 65% of infants at delivery (median 66 days after dose 2), 60% of mothers and 26% of infants at 3 months postdelivery, and 59% of mothers and 12% of infants at 6 months postdelivery.

**Conclusions.** Two 30- $\mu$ g doses were moderately immunogenic in HIV-infected pregnant women. No concerning vaccine-related safety signals were observed. Seroprotection persisted in most women postpartum. Efficient transplacental antibody transfer occurred, but seroprotection in infants waned rapidly. Vaccination to protect HIV-infected pregnant women and their newborns from new influenza strains is feasible, but more immunogenic platforms should be evaluated.

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Early in the 2009 pH1N1 pandemic, it was recognized that pregnant women were at high risk, with excess hospital and intensive care unit admissions and an estimated 5-fold relative risk of death. Risk was particularly associated with infection in the second and third trimesters, extending to the second week postpartum [1–6]. Similar patterns observed in previous influenza pandemics and epidemics were thought to reflect immunological and physiological changes associated with pregnancy [2]. Additionally, maternal infection with pH1N1 or seasonal influenza has been associated with preterm labor, preterm delivery, and adverse fetal/neonatal outcomes, and infection in young

infants with an increased risk of severe illness and death [2, 3, 5, 7, 8].

A second group that had increased risk of hospitalization, intensive care unit admission, and death in the 2009 pH1N1 pandemic was patients with medical comorbidities, including immunocompromising conditions, which conferred an estimated 16-fold relative risk of fatality [4, 9]. Although some reports did not link human immunodeficiency virus (HIV) infection to excess pH1N1 morbidity or death [10], others demonstrated increased hospital admission rates among HIV-infected populations [11, 12].

Women who are pregnant and HIV infected lie at the intersection of 2 high-risk conditions that, in combination, may pose particular vulnerability to pH1N1 infection. This group represents a high priority for vaccination to prevent maternal morbidity and mortality, avert pregnancy complications, and provide protection to infants in the first months of life before they are eligible for vaccination. Although information is available on the immune response to pH1N1 vaccine in pregnancy [5, 7, 8], none exists for HIV-infected pregnant women. Study P1086 of the International Maternal Pediatric Adolescent AIDS Clinical Trials (IMPAACT) group was designed to evaluate safety and immunogenicity of a pH1N1 vaccination regimen in HIV-infected pregnant women.

## METHODS

### Study Population

HIV-infected women 18–39 years of age and between 14–34 weeks of gestation were enrolled at 31 US IMPAACT sites in October–November 2009. Subjects were required to be on antiretroviral therapy prior to or concomitantly with the first visit. Exclusion criteria included allergy to eggs or other vaccine components; history of severe reaction to seasonal inactivated influenza vaccine; previous polymerase chain reaction (PCR)–documented pH1N1 infection or positive influenza test between June 2009 and screening; previous pH1N1 vaccination; any recent vaccination, nonlicensed medication, or blood product; acute illness or fever; neoplastic or immunosuppressive disease (other than HIV infection); immunosuppressive treatment including corticosteroids in the preceding 3 months; personal or family history of Guillain-Barré syndrome; and onset of neurologic disease in the preceding 6 months. Informed consent was obtained in accordance with the guidelines of the US Department of Health and Human Services and participating institutions.

### Vaccine Protocol

Unadjuvanted, inactivated pH1N1 monovalent vaccine (Fluvirin, Novartis, Basel, Switzerland), 30 µg/dose (2 standard 15 µg/0.5 mL injections, intramuscularly, 1 per upper extremity

or at least 2 inches apart in the same upper extremity), was given at entry. Subjects who did not experience a grade  $\geq 3$  adverse event or develop any condition that would have led to study exclusion received a second 30-µg dose on day 21 (+7 days). A 2 double-dose regimen was chosen to optimize immunogenicity. The second dose was temporarily deferred for acute illness, recent infection, or steroid treatment. Subjects who delivered prior to the second dose received the second dose after delivery. Inactivated seasonal influenza vaccine (not containing pH1N1) was permitted  $\geq 14$  days prior to the first dose or  $\geq 21$  days after the second dose of pH1N1 vaccine. Adverse events were assessed by a symptom diary and telephone contact or clinic visits on days 2, 10, and 21 after each vaccination; at delivery; and at 3 and 6 months after delivery. Clinic visits occurred within 72 hours of fever or symptoms of an influenza-like illness and within 24 hours of onset of lower extremity weakness, hand/feet tingling, or difficulty walking. Respiratory specimens were obtained at fever/influenza-like illness visits.

### Immunogenicity Assessments

Maternal pH1N1 hemagglutination inhibition (HAI) titers were measured as previously described [13], at entry, 21 days after dose 1; 10 and 21 days after dose 2; at delivery; and at 3 months after delivery. Infant HAI titers were measured at birth (cord blood or infant blood obtained within 7 days of delivery) and at 3 months. Maternal and infant titers were measured at 6 months after delivery in a subset of mother–infant pairs who received both doses of vaccine and had complete sets of samples prior to delivery. HAI titers were expressed as the reciprocal of the endpoint titer. Titers  $< 10$  were considered undetectable and were assigned values of 5. Seroprotection, defined as titer  $\geq 40$ ; geometric mean titers (GMTs); seroresponse, defined as a  $\geq 4$ -fold rise in titer from baseline following vaccination; and complete response, defined as attaining seroprotection and seroresponse, were characterized.

### Sample Size and Statistical Analysis

The accrual target was 130 subjects to obtain at least 100 mothers who received 2 vaccine doses and had complete sets of specimens prior to delivery, which would provide  $\geq 95\%$  probability of detecting an adverse event with a population incidence of  $\geq 3\%$  and confidence intervals of  $\pm 9\%$  or less for vaccine response rates. Antibody responses after the first vaccination were analyzed for subjects with available data who received the first vaccine dose and had the day 21 postvaccine dose 1 visit prior to delivery. Responses 10 and 21 days after the second vaccination were analyzed for women with available data who received both doses of vaccine and had the respective visits prior to delivery. Exact McNemar test was used

for comparisons of rates of seroprotection, seroresponse, and complete response among timepoints. Changes from baseline in maternal log<sub>10</sub> HAI titers and from delivery in infant titers and between subsequent timepoints were analyzed with the sign test. Wilcoxon signed-rank test was used for comparisons of titers among groups. Correlations between maternal and infant titers were done with the Spearman correlation test. Univariate and multivariable logistic regression analyses were performed to identify predictors of complete response.

## RESULTS

### Study Population

Of the 127 women analyzed, 123 received both vaccinations, at a median interval of 22 days (range, 21–34 days; Figure 1; Table 1). One hundred sixteen women delivered subsequent

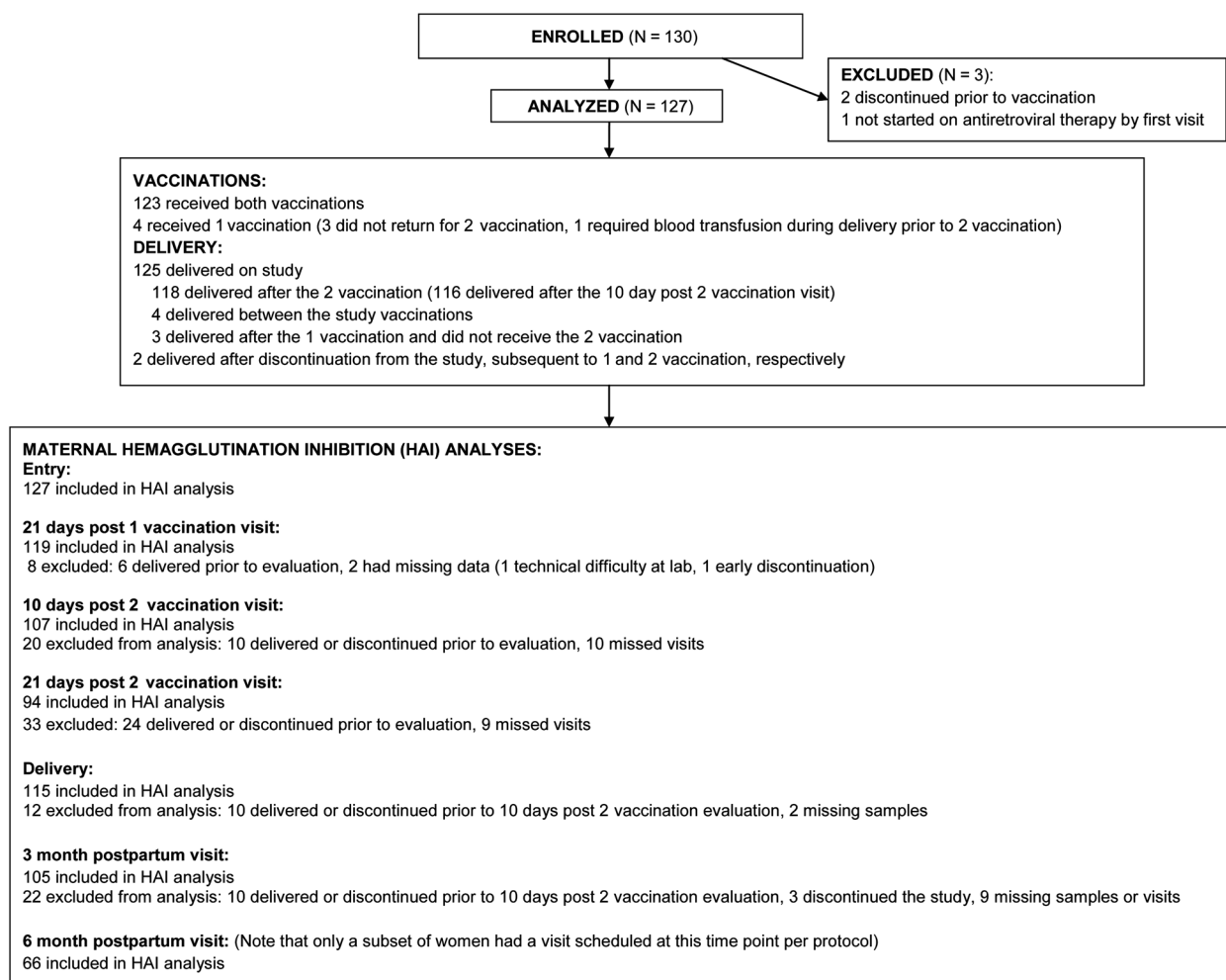
to the 10-day postvaccine dose 2 timepoint, at a median of 66 days (range, 9–155 days) following dose 2.

### Safety

There were no vaccine-related grade  $\geq 3$  events. Four subjects had grade 1 local reactions (pain, tenderness, erythema, and/or pruritis) and 4 had grade 1 systemic events (headache, rhinorrhea, and/or chills) temporally related to vaccination. One fetal death of undetermined etiology at 26 weeks' gestation (32 days after dose 1; 7 days after dose 2) was judged unrelated to vaccine by site investigators. Six subjects had influenza-like illness visits; respiratory specimen influenza A virus PCR was negative in 5, and 1 did not have testing.

### Maternal pH1N1 Antibody Responses

Seroprotection rates and GMTs increased, respectively, from 21% and 14 at entry to 73% and 85 following vaccine dose 1



**Figure 1.** Flow diagram of subjects enrolled and analyzed. Numbers of vaccinated subjects, delivery timing, and numbers of maternal hemagglutination inhibition results at each time point are described. Abbreviation: HAI, hemagglutination inhibition.

**Table 1. Characteristics of the Analyzed Population (N = 127)**

Characteristic	Median (Range) or Percentage
Age at entry, y	29 (18–38)
Race	
Black	61
White	30
Other or unknown	9
Ethnicity	
Hispanic	34
Non-Hispanic	63
Unknown	3
CD4 count at entry, cells/mm <sup>3</sup>	472 (28–1383)
CD4 percentage at entry	31 (1–53)
CD8 count at entry, cells/mm <sup>3</sup>	680 (247–1951)
CD8 percentage at entry	46 (18–75)
HIV load at entry <sup>a</sup>	
HIV load at entry, copies/mL	75 (40–294 200)
Percentage with ≤50 copies/mL	35
Percentage with ≤400 copies/mL	79
Antiretroviral therapy <sup>b</sup>	
HAART <sup>c</sup>	94
Non-HAART	6
Gestational age at entry, wk	26 (14–34)
Seasonal influenza vaccination	31 <sup>d</sup>
Delivery characteristic (n = 125)	
Cesarean	54
Preterm (26–36 wk)	14
Vertical transmission of HIV infection	1 <sup>e</sup>

Abbreviations: HAART, highly active antiretroviral therapy; HIV, human immunodeficiency virus.

<sup>a</sup> HIV load values below the limit of detection were replaced with the lower limit of the assay used for each subject.

<sup>b</sup> Prior to or at the initial vaccine visit.

<sup>c</sup> HAART is defined as ≥3 medications from ≥2 antiretroviral classes.

<sup>d</sup> Thirty-five subjects received inactivated seasonal influenza vaccine 14–30 days before study vaccine dose 1, and 5 between 21 days after study vaccine dose 2 and study end.

<sup>e</sup> One infant was confirmed to be HIV-infected during the period of infant follow-up.

and 80% and 92 at 10 days after vaccine dose 2 (Table 2; Figure 2). Among subjects assessed after vaccine dose 1, all 25 with baseline titers ≥40 retained seroprotective titers, and 62 of 94 (66%) with baseline titers <40 attained seroprotective titers ( $P < .0001$ ). Among subjects who received both doses and were assessed after dose 1 and 10 days after dose 2, all with titers ≥40 after vaccine dose 1 had seroprotective titers after vaccine dose 2, and 8 of 29 (28%) with titers <40 after vaccine dose 1 achieved titers ≥40 after vaccine dose 2 ( $P = .008$ ). Overall, after vaccine dose 2, 63 of 84 (75%) subjects with baseline titers <40 had seroprotective titers.

Seroresponse was observed in 66% after vaccine dose 1 and 72% 10 days after vaccine dose 2. Complete responses

(seroprotection and seroresponse) were observed in 61% after vaccine dose 1, with no difference between complete responders and nonresponders in baseline median titer (10 in both groups;  $P = .7$ ). After vaccine dose 2, the complete response rate was 65%, including 7 of 41 (17%) nonresponders after vaccine dose 1 who achieved complete response and 3 of 66 (5%) complete responders after dose 1 who no longer met criteria after dose 2.

Seroprotection rates and GMTs declined to 67% and 54 at delivery, 60% and 39 three months postpartum, and 59% and 39 six months postpartum, respectively. Of subjects who had seroprotective titers 10 days after dose 2 and had subsequent assessments, 65 of 85 (76%) had titers ≥40 at delivery, 52 of 78 (67%) at 3 months postpartum, and 35 of 53 (66%) at 6 months postpartum. Subsequent to the 10-day postvaccine dose 2 visit, ≥4-fold declines in titers occurred in 26 of 106 (25%) subjects by delivery, 42 of 98 (43%) by 3 months postpartum, and 22 of 66 (33%) by 6 months postpartum. Eight subjects had ≥4-fold increases subsequent to the 10-day postvaccine dose 2 visit.

### Infant pH1N1 Antibody Concentrations

Cord and neonatal blood specimen titers were not statistically different (median, 40 and 80, respectively;  $P = .4$ ), allowing these to be combined. Infant seroprotection rates and GMTs were 65% and 55 at birth, 26% and 14 at 3 months, and 12% and 13 at 6 months, respectively (Table 3; Figure 2). There was no difference between maternal and infant delivery titers (median difference, 0 [interquartile range {IQR}, –30 to 5]; median ratio, 1 [IQR], .5–2]). Infant birth titers strongly correlated with maternal delivery titers (Spearman correlation coefficient = .86;  $P < .0001$ ), but not with the interval between maternal vaccine dose 2 and delivery (Spearman correlation coefficient = –.08;  $P = .4$ ). There was a weak negative association between gestational age at birth and infant birth titers (Spearman correlation coefficient = –.17;  $P = .08$ ).

### Predictors of Maternal Complete Response

Greater nadir and entry CD4% and CD4 count were associated with complete response to both vaccinations and lower entry CD8% with response to the first vaccination (Table 4). In multivariable logistic regression models, nadir CD4% and entry CD8% did not add significantly to entry CD4% for the first vaccine dose ( $\chi^2 = .1$  and  $.8$ , respectively) and nadir CD4% did not add significantly to entry CD4% for the second dose ( $\chi^2 = .08$ ). Viral load was not associated with response.

## DISCUSSION

No serious vaccine-related reactions were observed with two 30- $\mu$ g doses (twice the standard amount of hemagglutinin per

**Table 2. Maternal Hemagglutination Inhibition Antibody Responses**

Time Point	Subjects, No.	Seroprotection <sup>a</sup> (%) (95% CI)	Geometric Mean Titer (95% CI)	Seroresponse <sup>b</sup> (%) (95% CI)	Complete Response <sup>c</sup> (%) (95% CI)
Entry	127	21 (15–29)	14 (12–17) <sup>d</sup>	...	...
21 d post-dose 1	119	73 (64–81) <sup>e</sup>	85 (66–111) <sup>f</sup>	66 (57–75)	61 (51–69)
10 d post-dose 2	107	80 (72–87) <sup>e,g</sup>	92 (72–116) <sup>f</sup>	72 (62–80)	65 (56–74)
21 d post-dose 2	94	77 (67–85) <sup>e</sup>	81 (62–106) <sup>f,h</sup>	67 (57–76)	63 (52–73)
Delivery	115	67 (58–75) <sup>e,g</sup>	54 (43–69) <sup>f,h</sup>	57 (48–67) <sup>i</sup>	51 (42–61) <sup>j</sup>
3 mo postdelivery	105	60 (50–69) <sup>e</sup>	39 (32–49) <sup>f,h</sup>	51 (41–61)	46 (36–56)
6 mo postdelivery	66	59 (46–71) <sup>e</sup>	39 (30–51) <sup>f</sup>	58 (45–70)	48 (36–61)

Abbreviation: CI, confidence interval; HAI, hemagglutination inhibition.

Entry antibody titers were analyzed for subjects who received at least the first vaccination. Antibody responses after the first vaccination were analyzed for subjects with available data who received the first vaccine dose and had the day 21 postvaccine dose 1 visit prior to delivery. Responses 10 and 21 days after the second vaccination were analyzed for women with available data who received both doses of vaccine and had the respective visits prior to delivery. Maternal antibody levels at delivery and 3 and 6 months postdelivery were analyzed for those women with available data who received both doses of vaccine and delivered subsequent to the 10-day postvaccine dose 2 timepoint.

<sup>a</sup> Seroprotection defined as HAI titer  $\geq 40$ .

<sup>b</sup> Seroresponse defined as  $\geq 4$ -fold rise in HAI titer from entry.

<sup>c</sup> Complete response defined as HAI titer  $\geq 40$  and  $\geq 4$ -fold rise in HAI titer from entry.

<sup>d</sup> Baseline titers did not differ according to whether seasonal influenza vaccine had been received prior to entry ( $P = .7$ ).

<sup>e</sup>  $P < .0001$  (McNemar test) for the comparison to seroprotection at entry.

<sup>f</sup>  $P < .0001$  (sign test) for the comparison to geometric mean titer at entry.

<sup>g</sup>  $P \leq .008$  (McNemar test) for the comparison to seroprotection at previous timepoint.

<sup>h</sup>  $P \leq .04$  (sign test) for the comparison to geometric mean titer at previous timepoint.

<sup>i</sup>  $P = .007$  (McNemar test) for the comparison to seroresponse at previous timepoint.

<sup>j</sup>  $P = .007$  (McNemar test) for the comparison to complete response at previous timepoint.

dose administered for seasonal and pH1N1 vaccination) of unadjuvanted pH1N1 vaccine in HIV-infected women in the second or third trimester of pregnancy. Previous reports provide evidence of safety of unadjuvanted and adjuvanted monovalent pH1N1 vaccines during pregnancy [5, 14–19] and in HIV-infected, nonpregnant adults [20] and children [13, 21]. Although our study was not powered to detect uncommon events, it adds to the safety record supporting influenza vaccination during pregnancy.

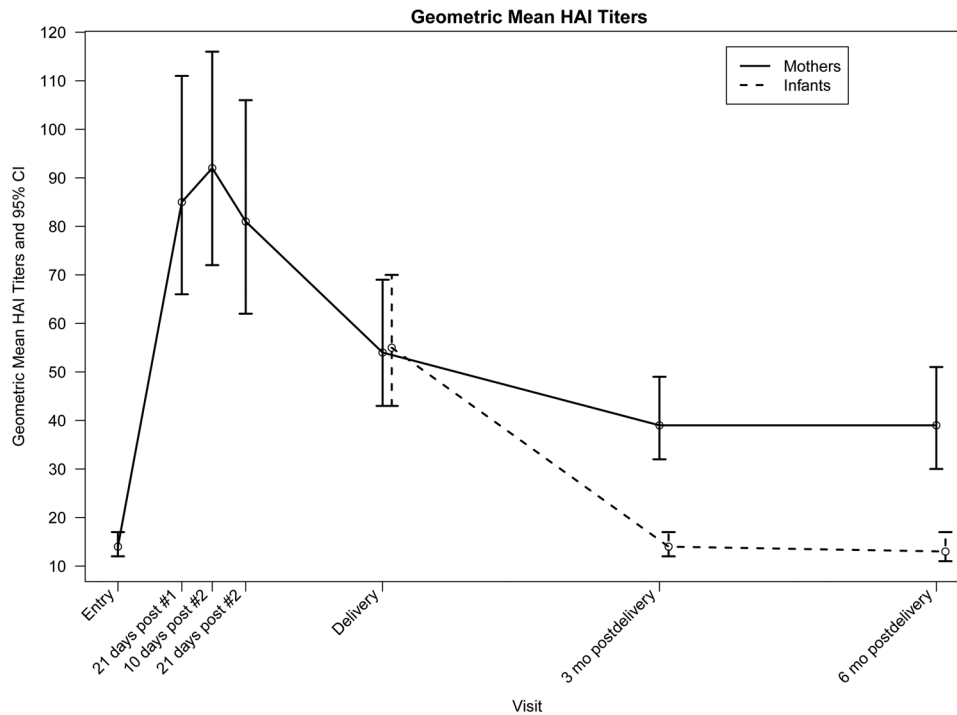
The 2 double-dose regimen was moderately immunogenic, yielding seroprotective titers in 73% after 1 dose, including 66% of those lacking seroprotective titers at baseline, and seroresponse in 66%. After the second dose, 80% achieved seroprotective titers, including 75% of those lacking seroprotective titers at baseline and 28% of those lacking seroprotective titers after the first dose, and 72% demonstrated seroresponse. GMTs attained were relatively modest. Seroprotective titers were present in 67% of mothers and 65% of infants at delivery; however, rates of seroprotection in infants dropped rapidly over the first 3–6 months of life.

Several studies demonstrated that pH1N1 vaccines are immunogenic in HIV-uninfected pregnant women, producing high levels of seroprotection (89%–100%) and seroresponse (89%–93%) after a single, standard dose of unadjuvanted or adjuvanted vaccine [5, 8, 19, 22, 23], and maternal titers at

delivery similar to those of women infected with pH1N1 virus during pregnancy [7]. In contrast, one small, unpublished study suggested that a standard 15- $\mu$ g dose of unadjuvanted vaccine induced lower rates of seroprotection (85%) and seroresponse (69%) compared with a 30- $\mu$ g dose (96% seroprotection and 93% seroresponse) in pregnant women or compared with a standard dose in nonpregnant women (93% seroprotection and 86% seroresponse) [24]. Nonetheless, findings of adequate immunogenicity of pH1N1 vaccines during pregnancy in most studies are consistent with studies of seasonal influenza vaccination in pregnancy [8, 19].

Nonpregnant, HIV-infected populations generally had inferior responses to standard doses of unadjuvanted pH1N1 vaccines compared with immunocompetent cohorts, with seroprotection and/or seroresponse rates after 1 dose between 31%–76% vs rates of 56%–97% in HIV-uninfected adult vaccinees and lower GMTs among HIV-infected vaccinees [20, 21, 25–29]. A higher (30  $\mu$ g) dose increased seroprotection, seroresponse, and GMTs, but responses were still poorer than in HIV-uninfected populations [30]. A second dose improved responses in some studies, but not others [25, 26, 30]. Among HIV-infected children, a single, standard dose induced seroprotection in 54%–94% and seroresponse in 49%–63%, increasing to 68%–100% and 65%–68%, respectively, after a second dose, compared with seroprotection in 62%–100% and





**Figure 2.** Geometric mean hemagglutination inhibition titers of pregnant women and their infants. Titers and 95% confidence intervals at study entry; 21 days after the first dose of pH1N1 vaccine; 10 and 21 days after the second dose of vaccine; delivery; and 3 and 6 months postpartum are shown. Abbreviations: CI, confidence interval; HAI, hemagglutination inhibition.

98%–100% of HIV-uninfected children after 1 and 2 standard doses, respectively. Two 30- $\mu$ g vaccinations produced a 80%–85% seroprotection and 80%–84% seroresponse in HIV-infected children, with incremental gains after the second vaccine, but seroprotection was lower than rates of 88%–100% in HIV-uninfected children [13, 21, 31] (R. Pass, S. Nachman, P. Flynn, et al, unpublished data). Poorer responses of HIV-infected persons to influenza vaccines, even with preserved CD4 counts, have been attributed to immune dysregulation affecting T- and B-cell quality and function, immune activation,

immunosenescence, and high regulatory T-cell numbers [20, 32]. The responses we observed are lower than those in HIV-uninfected pregnant women and more similar to those described in other HIV-infected populations [32].

Response rates and GMTs in nonpregnant, HIV-infected cohorts were improved with adjuvanted pH1N1 vaccines [10]. Seroprotection and seroresponse rates after 1 ASO3-adjuvanted vaccine dose were 70%–93% and 68%–89%, increasing to 94%–98% and 86%–97%, respectively, after 2 doses and approaching >95% response rates in HIV-uninfected populations [26, 33–35]. With MF59-adjuvanted vaccine, seroprotection rates among HIV-infected adults were 88% after 1 dose and 91% after 2 doses, similar to HIV-uninfected controls, although GMTs of the HIV-infected group were lower. In HIV-infected children, seroprotection rates were 94%–100% after 1 MF59-adjuvanted vaccine dose and 100% after 2 doses [13, 36]. These observations are consistent with greater responses to adjuvanted seasonal influenza vaccines in HIV-infected populations and adjuvanted pH1N1 vaccines in immunocompetent adults and children [26, 35]. The safety and immunogenicity of adjuvanted pH1N1 vaccines in HIV-infected pregnant women have not been described.

Goals of influenza immunization during pregnancy include inducing a sufficiently durable antibody response to protect maternal health through the postpartum period, reduce

**Table 3. Infant Hemagglutination Inhibition Antibody Levels**

Timepoint	Subjects No.	Seroprotection <sup>a</sup> (%) (95% CI)	Geometric Mean Titer (95% CI)
Birth	106	65 (55–74)	55 (43–70)
3 mo	97	26 (17–36) <sup>b</sup>	14 (12–17) <sup>c</sup>
6 mo	59	12 (5–23) <sup>b</sup>	13 (11–17) <sup>c</sup>

Abbreviation: CI, confidence interval.

Infant antibody levels at birth, 3 months, and 6 months were analyzed for those infants with available data whose mothers received both doses of vaccine and delivered subsequent to the 10-day postvaccine dose 2 timepoint.

<sup>a</sup> Seroprotection defined as hemagglutination inhibition titer  $\geq$ 40.

<sup>b</sup>  $P < .0001$  (McNemar test) for the comparison to seroprotection at birth.

<sup>c</sup>  $P < .0001$  (sign test) for the comparison to geometric mean titer at birth.

**Table 4. Predictors of Maternal Complete Response to Study Vaccinations (Univariate Analyses)**

Characteristic	Dose 1			Dose 2		
	No. <sup>a</sup>	OR (95% CI)	P Value	No. <sup>a</sup>	OR (95% CI)	P Value
Age	119	1.062 (.992–1.136) <sup>b</sup>	.09	107	1.056 (.982–1.136) <sup>b</sup>	.1
Black race	112	.989 (.449–2.175)	1.0	100	1.364 (.589–3.158)	.5
Hispanic ethnicity	115	1.546 (.691–3.455)	.3	103	1.268 (.533–3.020)	.6
Nadir CD4 percentage	113	1.071 (1.031–1.113) <sup>c</sup>	<.001	102	1.054 (1.014–1.096) <sup>c</sup>	.01
Nadir CD4 count	113	1.002 (1.000–1.004) <sup>d</sup>	.02	102	1.003 (1.001–1.005) <sup>d</sup>	.01
P1061s CD4 percentage	119	1.068 (1.028–1.109) <sup>c</sup>	.001	107	1.043 (1.004–1.083) <sup>c</sup>	.03
P1061s CD4 count	116	1.002 (1.000–1.004) <sup>d</sup>	.02	105	1.002 (1.000–1.004) <sup>d</sup>	.03
P1061s CD8 percentage	119	.963 (.931–.996) <sup>c</sup>	.03	107	.976 (.941–1.011) <sup>c</sup>	.2
P1061s CD8 count	109	.999 (.998–1.000) <sup>d</sup>	.2	98	.999 (.998–1.001) <sup>d</sup>	.4
P1061s CD19 percentage	108	.983 (.912–1.059) <sup>c</sup>	.7	98	.983 (.909–1.063) <sup>c</sup>	.7
P1061s CD19 count	97	.999 (.996–1.002) <sup>d</sup>	.6	88	.999 (.996–1.002) <sup>d</sup>	.6
P1061s HIV viral load <400 copies/mL	119	1.253 (.570–2.759)	.6	107	1.047 (.446–2.459)	.9
HAART	119	.596 (.111–3.205)	.5	107	.743 (.137–4.028)	.7
Gestational age at entry	119	1.017 (.954–1.083) <sup>e</sup>	.6	107	1.010 (.943–1.080) <sup>e</sup>	.8
Seasonal influenza vaccination prior to entry	119	.907 (.403–2.039)	.8	107	.945 (.394–2.268)	.9
pH1N1 HAI titer ≥40 at entry	119	.974 (.396–2.397)	1.0	107	.616 (.240–1.581)	.3
pH1N1 log <sub>10</sub> HAI titer at entry	119	1.054 (.396–2.803) <sup>f</sup>	.9	107	.695 (.249–1.944) <sup>f</sup>	.5

Complete responses after dose 2 were based on antibody titers measured at the 10 days post-dose 2 timepoint.

Abbreviations: CI, confidence interval; HAART, highly active antiretroviral therapy; HAI, hemagglutination inhibition; OR, odds ratio.

<sup>a</sup> No. of participants with available data.

<sup>b</sup> Odds ratio for every year increase.

<sup>c</sup> Odds ratio for every 1% increase.

<sup>d</sup> Odds ratio for every cell/mm<sup>3</sup> increase.

<sup>e</sup> Odds ratio for every week increase.

<sup>f</sup> Odds ratio for every log<sub>10</sub> increase in titer.

exposure of the neonate to maternal influenza, and provide passive immunization to protect the newborn before his/her eligibility for vaccination at 6 months of life. In HIV-uninfected pregnant women vaccinated with various pH1N1 regimens in the second or third trimesters, protective titers were maintained in 62%–100% of women at delivery [5, 8, 19, 22, 24], with protective titers persisting in 90%–100% at 2–3 months [8, 19] and 100% at 5 months after delivery [8]. We found at delivery that, despite a drop in GMT, protective titers were present in 67% of women, including 76% who had achieved seroprotective titers after the second vaccine dose, and protective titers persisted in 60% and 59% at 3 and 6 months postpartum, respectively. Thus, the majority of HIV-infected pregnant women maintained protective titers at delivery and postpartum.

Efficient transplacental antibody delivery occurs following pH1N1 vaccination of HIV-uninfected pregnant women, yielding neonatal seropositivity rates of 77%–96%. Neonatal titers exceeded maternal titers in some studies, consistent with active transplacental immunoglobulin G transport, and were lower than maternal titers in others [5, 8, 19, 22, 24, 37]. In

one study of infants born to HIV-uninfected women who received MF59-adjuvanted vaccine during the third trimester, seroprotective antibody levels were present in 96% of neonates at delivery and were still present in 96% at 2 months and 81% at 5 months [8]. Transplacental pH1N1 antibody likely confers clinical protection similar to that conferred by transplacental antibody following maternal seasonal influenza vaccination during gestation [5, 8, 19, 38, 39]. In the current study, maternal pH1N1 vaccination was associated with efficient transplacental antibody transfer; the 65% level of seroprotection in neonates and the neonatal GMT were virtually identical to those of mothers at delivery, and there was a strong correlation between maternal delivery titers and neonatal titers. However, because GMTs were modest, with the expected decline of passively acquired antibody, infant seroprotection rates dropped rapidly to 26% at 3 months and 12% at 6 months; infant protection against severe influenza infection was therefore likely short-lived.

Twenty-one percent of subjects had seroprotective titers at entry, consistent with baseline seroprotection rates of 25%–70% in other studies of pH1N1 vaccination in HIV-infected

populations [13, 21, 25, 27, 28, 36]. Because our study was performed in the fall of 2009, well into the pandemic which began in the preceding spring, baseline seroprotection may reflect prior pH1N1 infection and/or cross-reactivity with prior seasonal influenza immunization or infection. Similarly,  $\geq 4$ -fold antibody rises observed in small numbers of subjects subsequent to the postvaccination dose 2 visit may have been due to subclinical or mild pH1N1 infection that did not trigger clinical evaluation.

Nadir and entry CD4% and CD4 count were the primary predictors of maternal vaccine response, whereas HIV RNA load was not predictive. In studies of pH1N1 vaccination of HIV-infected adults and children, predictors included younger age, shorter duration of HIV infection, absence of AIDS-associated conditions, highly active antiretroviral therapy, higher nadir CD4 count, higher current CD4 count, CD8 count (lower in some studies, higher in others) and lower HIV RNA load [20, 21, 27–29, 30, 33, 36]. In some studies, a higher baseline antibody titer was associated with greater responses [13, 21, 27, 30], whereas the opposite was found in other studies [25, 33]. Our study population may have lacked sufficient variation in parameters such as receipt of highly active antiretroviral therapy and HIV RNA load to demonstrate an impact of these factors.

In this first study examining pH1N1 vaccination in HIV-infected pregnant women, a 2 double-dose vaccination regimen in the second and third trimesters had moderate immunogenicity and no concerning vaccine-related safety signals. Seroprotection persisted in a majority through delivery and postpartum, and efficient transplacental transfer of antibody was demonstrated, although seroprotection in infants waned rapidly. These results demonstrate feasibility of vaccination to protect HIV-infected pregnant women and their newborns from new influenza viruses. Nevertheless, pH1N1 vaccination was less immunogenic than in HIV-uninfected pregnant women. Although lack of a comparator group precludes definite conclusions about the optimal vaccination regimen in HIV-infected pregnant women, gains after a second vaccine dose were marginal. Alternative vaccine platforms, including adjuvanted vaccines, should be evaluated in HIV-infected pregnant women to improve upon protection that vaccination can provide to this highly vulnerable group against novel influenza strains.

## Notes

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