Safety and Immunogenicity of a Baculovirus-Expressed Hemagglutinin Influenza Vaccine
A Randomized Controlled Trial

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A high priority in vaccine research is the development of influenza vaccines that do not use embryonated eggs as the substrate for vaccine production.

Objective To determine the dose-related safety, immunogenicity, and protective efficacy of an experimental trivalent influenza virus hemagglutinin (rHA0) vaccine produced in insect cells using recombinant baculoviruses.

Design, Setting, and Participants Randomized, double-blind, placebo-controlled clinical trial at 3 US academic medical centers during the 2004-2005 influenza season among 460 healthy adults without high-risk indications for influenza vaccine.

Interventions Participants were randomly assigned to receive a single injection of saline placebo (n=154); 75 µg of an rHA0 vaccine containing 15 µg of hemagglutinin from influenza A/New Caledonia/20/99(H1N1) and influenza B/Jiangsu/10/03 virus and 45 µg of hemagglutinin from influenza A/Wyoming/3/03(H3N2) virus (n=153); or 135 µg of rHA0 containing 45 µg of hemagglutinin from each of 3 components (n=153). Serum samples were taken before and 30 days following immunization.

Main Outcome Measures Primary safety end points were the rates and severity of solicited and unsolicited adverse events. Primary immunogenicity end points were the rates of 4-fold or greater increases in serum hemagglutinin inhibition antibody to each of the 3 vaccine strains before and 28 days after inoculation. The prespecified primary efficacy end point was culture-documented influenza illness, defined as development of influenza-like illness associated with influenza virus on a nasopharyngeal swab.

Results Rates of local and systemic adverse effects were low, and the rates of systemic adverse effects were not different in either vaccine group than in the placebo group. Hemagglutinin inhibition antibody responses to the H1 component were seen in 3% of placebo, 51% of 75-µg vaccine, and 67% of 135-µg vaccine recipients, while responses to B were seen in 4% of placebo, 65% of 75-µg vaccine, and 92% of 135-µg vaccine recipients. Responses to the H3 component occurred in 11% of placebo, 81% of 75-µg vaccine, and 77% of 135-µg vaccine recipients. Influenza infections in the study population were due to influenza B and A(H3N2), and influenza A infections were A/California/7/2004-like viruses, an antigenically drifted strain. Seven cases of culture-confirmed CDC-defined influenza-like illness occurred in 153 placebo recipients (4.6%) compared with 2 cases (1.3%) in 150 recipients of 75 µg of vaccine, and 0 cases in recipients of 135 µg of vaccine.

Conclusions In this study, a trivalent rHA0 vaccine was safe and immunogenic in a healthy adult population. Preliminary evidence of protection against a drifted influenza A(H3N2) virus was obtained, but the sample size was small. Inclusion of a neuraminidase component did not appear to be required for protection.

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recombinant baculovirus (rHA0). This alternative avoids dependence on eggs and is very efficient because of the high levels of protein expression under the control of the baculovirus polyhedrin promoter. Monovalent and bivalent baculovirus-derived influenza vaccines have been evaluated in young adults and in community-dwelling adults older than 65 years. These studies found that the vaccines are well tolerated and immunogenic. Recently, doses of a trivalent vaccine ranging from 15 µg to 135 µg per component were shown to be well tolerated in elderly persons and to induce antibody responses at rates comparable with or superior to the licensed trivalent vaccine. While a clear dose-response relationship has been shown for the H3 component in both healthy adults and in elderly persons, relatively little difference has been observed in the immune response to doses ranging from 15 µg to 135 µg of the H1 and B components. However, this analysis has been confounded by a poor correlation between the antigen content of the H1 and B components of rHA0 vaccines, as determined by measurement of total protein content, compared with the values determined by single radial immunodiffusion (SRID), the standard measurement used for trivalent inactivated vaccine (TIV).

Therefore, the primary objective of the current study was to evaluate the relative immunogenicity of the H1 and B components of the vaccine when formulated at either 15 µg or 45 µg per component, as determined by SRID. In addition, we used the opportunity to follow up participants throughout the influenza season to obtain preliminary evidence of protective efficacy in a healthy adult population.

**METHODS**

**Vaccine**

The vaccine used in this study consisted of purified HA proteins produced in insect cells using a baculovirus expression system, as previously described. Because the HA produced in this system is not cleaved, the resulting product is referred to as rHA0. The vaccine was formulated as a trivalent preparation containing the purified rHA0 of the A/New Caledonia/20/99 (H1N1), A/Wyoming/30/03 (H3N2), and B/Jiangsu/10/03 influenza viruses expressed from genes cloned by reverse transcriptase polymerase chain reaction from the same Centers for Disease Control and Prevention (CDC)–derived vaccine seed viruses used for the licensed TIV and formulated in phosphate-buffer saline containing 0.005% detergent without preservative. Sodium dodecyl sulfate–polyacrylamide gel electrophoresis of the purified monovalent materials indicated that hemagglutinin constitutes approximately 95% of the total protein. The experimental vaccine was formulated at 2 different concentrations, as determined by SRID. The high-dose formulation contained 45 µg of each component, for a total dose of 135 µg of rHA0 per 0.5-mL dose based on the SRID values on the preblend bulk. After formulation, it was determined that this dose contained 35 µg rather than 45 µg of the H1 component. The low-dose formulation contained 45 µg of the H3 rHA0 and 15 µg each of the H1 and B rHA0, for a total dose of 75 µg of rHA0. Placebo consisted of normal saline for injection. Vaccine and placebo were supplied in coded, identical-appearing single-dose vials.

**Clinical Study Design**

The study was conducted as a randomized, double-blind, placebo-controlled study at 3 medical centers (University of Rochester, Rochester, NY; Cincinnati Children's Hospital, Cincinnati, Ohio; and University of Virginia, Charlottesville) during the 2004-2005 influenza season. Participants were healthy adults aged 18 to 49 years who did not belong to high-priority target groups for influenza vaccination as defined by the Advisory Committee on Immunization Practices and who had not received previous influenza vaccination for the 2004-2005 season. Women of childbearing potential had to have a negative urine pregnancy test result at the time of randomization and be willing to use an adequate form of contraception during the course of the study. Participants were recruited by newspaper and radio advertisements, posters, and word of mouth. Race/ethnicity data for all participants were collected from self-reports. Participants were compensated for each visit ($25).

After screening medical history and physical examination to determine eligibility, 10 mL of blood was collected from an arm vein for serologic studies, and participants were randomly assigned to receive a single dose of either rHA0 at 135 µg, rHA0 at 75 µg, or saline placebo using a permuted-block randomization scheme stratified by study site. Vaccine was administered as a single intramuscular injection in the upper deltoid.

Participants measured their oral temperature daily and maintained a diary card for 7 days after vaccination on which they recorded local and systemic reactions graded as mild (noticeable but not interfering with normal activities), moderate (some interference with normal activities), and severe (symptom prevented normal daily activities). Participants also returned on day 2 and day 7 after vaccination for review of the diary card, concomitant medications and medical history, and examination of the vaccination site. Participants returned approximately 28 days after vaccination for review of interim medical history. In addition, 10 mL of venous blood was obtained from an arm vein for assessment of antibody to influenza virus. A final study visit occurred at day 180, during which participants underwent a physical examination and interval medical history.

The study protocol was reviewed and approved by the investigational review boards at all 3 clinical sites, and written informed consent was obtained from all participants prior to study entry.

**Surveillance for Influenza**

Following the day 28 visit, participants started to complete a weekly diary to record influenza symptoms, and after influenza was recognized in the community, participants received weekly telephone calls to review the diary and ascertain presence or absence of respiratory illness symptoms. Participants were instructed to return to the clinic for illness evaluations if they observed any acute respira-
ory tract symptoms or fever. During these illness visits, symptoms were reviewed, a brief physical examination was conducted, and nasopharyngeal swabs for virus culture were obtained.

**Laboratory Assays**

Serum samples were assessed for antibody to each of the 3 components of the vaccine by hemagglutination inhibition (HAI) using standard methods. Egg-grown influenza A/New Caledonia/20/99 and influenza A/Wyoming/03/03 were obtained from the CDC, Atlanta, Ga, while assays against the influenza B/Jiangsu/10/03 used antigen prepared in Madin-Darby canine kidney cells from a seed virus supplied by the CDC. Serum samples were treated with neuraminidase (receptordestroying enzyme, Denka Seiken, Tokyo, Japan) to remove nonspecific inhibitors of hemagglutination prior to testing and were tested in serial 2-fold dilutions at an initial dilution of 1:4. Serum samples with no reactivity at 1:4 were assigned a value of 1.2. Assays were performed using chick red blood cells (Colorado Serum Company, Denver) for influenza A/New Caledonia/20/99 and B/Jiangsu/10/03 viruses and turkey red blood cells (Viromed Laboratories, Minnetonka, Minn) for the influenza A/Wyoming/03/03 virus.

Nasopharyngeal swabs for virus culture were stored at −70°C and shipped on dry ice to a central laboratory (Cincinnati Children’s Hospital Medical Center), where virus isolation was performed in primary rhesus monkey kidney cells (Diagnostic Hybrids Inc, Athens, Ohio). The presence of influenza A or B viruses in the culture was determined by immunofluorescence using type-specific monoclonal antibodies (Diagnostic Hybrids Inc). Influenza A isolates were further subtyped at Protein Sciences Corp by sequence analysis of the entire HA1 region after reverse transcriptase-polymerase chain reaction amplification of Madin-Darby canine kidney cell–grown virus.

**Definition of End Points**

The primary safety end points for this study were the rates and severity of solicited and unsolicited adverse events. The primary immunogenicity end points were the rates of 4-fold or greater increases in serum HAI antibody to each of the 3 vaccine strains comparing prevaccination and 28-day postvaccination samples. The pre-specified primary efficacy end point was culture-documented influenza illness, defined as development of a CDC-defined influenza-like illness associated with recovery of influenza virus from a nasopharyngeal swab. A CDC-defined influenza-like illness was defined as the presence of documented fever with body temperature higher than 37.7°C (99.8°F) plus either sore throat or cough.

**Statistical Analyses**

Rates of safety end points were based on the most severe response and were evaluated by χ² test. Differences between the proportions of participants with at least a 4-fold increase in HAI antibody for each pairwise treatment group comparison were tested using a χ² test.

The sample size for this study was chosen primarily based on the immunogenicity end point. Assuming that from 60% to 80% of the participants in an active treatment group would have a 4-fold or greater serum HAI antibody response to any specific strain, inclusion of 150 participants per group would have 80% power to detect an approximately 13% to 14% difference in response rates between study groups. Although not designed primarily as an efficacy trial, with a 5% attack rate in the placebo group, the study had power ranging from 14% to 53% to detect vaccine efficacy ranging from 40% to 80%. A P < .05 level was considered to be statistically significant. The analyses were conducted using SAS software, version 9.1 (SAS Institute Inc, Cary, NC).

**RESULTS**

A total of 460 participants were randomized, 458 were vaccinated, and 451 (98.5%) completed all study procedures. The disposition of the participants is shown in the figure. Of the 460 enrolled participants, 153 were randomized to 75 µg of rHA0 vaccine, 153 were randomized to 135 µg of rHA0 vaccine, and 154

**Figure. Flow of Participants Through the Trial**

154 Randomized to Receive Placebo 154 Received Placebo as Assigned
153 Randomized to Receive rHA0, 75 µg 151 Received Vaccine as Assigned 2 Did Not Receive Vaccine as Assigned
153 Randomized to Receive rHA0, 135 µg 153 Received Vaccine as Assigned
1 Did Not Meet Entry Criteria (Received Trivalent Influenza Vaccine) 2 No Day 28 Titer
1 Did Not Meet Entry Criteria (Pregnant) 2 No Day 28 Titer
154 Included in Safety Analysis 151 Included in Safety Analysis 2 Excluded (Did Not Receive Vaccine)
151 Included in Safety Analysis 3 Excluded 1 Received Trivalent Influenza Vaccine 2 No Day 28 Titer
150 Included in Immunogenicity Analysis 3 Excluded 1 Received Trivalent Influenza Vaccine 2 No Day 28 Titer
153 Included in Efficacy Analysis 1 Excluded (Received Trivalent Influenza Vaccine) 150 Included in Efficacy Analysis 3 Excluded 2 Did Not Receive Vaccine 1 Pregnant
151 Included in Efficacy Analysis 2 Excluded (Received Trivalent Influenza Vaccine)
were randomized to placebo. There were 9 participants who did not complete the study, including 1 who withdrew consent, 5 who were lost to follow-up, 2 participants in the 75-µg rHA0 treatment group who were randomized but not vaccinated, and 1 who was incarcerated. The majority of participants were white (86%) and female (63%) (Table 1). The mean age was 31.7 years (range, 18-49 years). There were no differences with respect to age, sex, or race/ethnicity between the groups.

**Assessment of Vaccine Safety**

The rates and severities of local and systemic symptoms reported on the diary card are shown in Table 2. Injection of rHA0 vaccine was associated with local injection site pain that was significantly more frequent than after saline placebo injection (P=.001). How-ever, 97% of all reports of pain after rHA0 vaccine were rated as mild. Systemic symptoms following vaccination also did not occur at significantly different rates in vaccine and placebo recipients (P>.07 for all comparisons). The most frequently reported systemic symptom following vaccination was headache. The majority (86%) of reports of headache were also rated as mild, and there was no difference in the frequency of headache between vaccine and placebo recipients (P>.07). There were no reported fevers (oral temperature >37.7°C [>99.8°F]) following vaccination.

Two participants (1%) in the 135-µg vaccine group experienced serious adverse events that were considered to be unrelated to vaccine (1 seizure related to hypoglycemia and 1 newly diagnosed lobular carcinoma in situ). Two additional participants (1 in the 75-µg vaccine group and 1 in the 135-µg vaccine group) experienced severe adverse events (1 infected nevus and 1 knee injury) that were also considered to be unrelated to vaccine. No participants discontinued the study because of adverse events and no participants died.

**Immunogenicity**

Serum antibody responses to vaccination are summarized in Table 3. Overall, among participants vaccinated with rHA0 vaccine, the frequencies of HAI antibody responses (≥4-fold increase comparing day 0 with day 28) to influenza A/New Caledonia, B/Jiangsu, and A/Wyoming were significantly greater (range, 51%-92%) than was observed for participants vaccinated with placebo (range, 3%-11%; P<.001). Antibody responses were seen in most participants receiving either the 75-µg or the 135-µg dose of rHA0 vaccines. However, the frequency of responses to both the A/New Caledonia/99 and B/Jiangsu/03 influenza viruses were significantly higher in the group receiving the 135-µg dose (P=.003), consistent with the higher doses of H1 and B components in the 135-µg vaccine. The frequency of HAI antibody response to the A/Wyoming/03 (H3N2) influenza virus was not different between the 75-µg and 135-µg doses, which contained identical amounts of the H3 component. Similarly, there were significant differences in the day 28 geometric mean titer of HAI antibody between the 75-µg and 135-µg doses for both the H1 and B components but not the H3 component.

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**Table 1. Baseline Participant Characteristics**

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Placebo (n = 154)</th>
<th>75-µg Vaccine (n = 151)</th>
<th>135-µg Vaccine (n = 153)</th>
<th>Overall (N = 458)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Race/ethnicity, No. (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>139 (90)</td>
<td>126 (83)</td>
<td>130 (85)</td>
<td>395 (86)</td>
</tr>
<tr>
<td>Black/African American</td>
<td>9 (6)</td>
<td>12 (8)</td>
<td>9 (6)</td>
<td>30 (7)</td>
</tr>
<tr>
<td>Latino/Hispanic</td>
<td>1 (1)</td>
<td>2 (1)</td>
<td>5 (3)</td>
<td>8 (2)</td>
</tr>
<tr>
<td>Asian</td>
<td>4 (3)</td>
<td>10 (7)</td>
<td>4 (3)</td>
<td>18 (4)</td>
</tr>
<tr>
<td>American Indian/Alaskan Native</td>
<td>0</td>
<td>0</td>
<td>1 (1)</td>
<td>1 (&lt;1)</td>
</tr>
<tr>
<td>Native Hawaiian/Pacific Islander</td>
<td>0</td>
<td>1 (1)</td>
<td>1 (1)</td>
<td>2 (&lt;1)</td>
</tr>
<tr>
<td>Other/not stated</td>
<td>1 (1)</td>
<td>0</td>
<td>3 (2)</td>
<td>4 (1)</td>
</tr>
<tr>
<td>Male, No. (%)</td>
<td>65 (42)</td>
<td>48 (32)</td>
<td>57 (37)</td>
<td>170 (37)</td>
</tr>
<tr>
<td>Age, median (range), y</td>
<td>32 (18-49)</td>
<td>32 (18-49)</td>
<td>30 (18-49)</td>
<td>31 (18-49)</td>
</tr>
</tbody>
</table>

**Table 2. Local and Systemic Symptoms Experienced in the 7 Days Following Vaccination**

<table>
<thead>
<tr>
<th>Symptoms</th>
<th>Placebo (n = 154)</th>
<th>75-µg Vaccine (n = 151)</th>
<th>135-µg Vaccine (n = 153)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mild</td>
<td>Moderate</td>
<td>Severe</td>
</tr>
<tr>
<td>Anthraxias</td>
<td>7 (5)</td>
<td>1 (1)</td>
<td>0</td>
</tr>
<tr>
<td>Chills</td>
<td>2 (2)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Fatigue</td>
<td>21 (14)</td>
<td>7 (5)</td>
<td>0</td>
</tr>
<tr>
<td>Headache</td>
<td>48 (31)</td>
<td>15 (10)</td>
<td>0</td>
</tr>
<tr>
<td>Myalgias</td>
<td>16 (10)</td>
<td>3 (2)</td>
<td>0</td>
</tr>
<tr>
<td>Nausea</td>
<td>9 (6)</td>
<td>1 (1)</td>
<td>0</td>
</tr>
<tr>
<td>Pain</td>
<td>24 (16)</td>
<td>1 (1)</td>
<td>0</td>
</tr>
<tr>
<td>Sweats</td>
<td>6 (4)</td>
<td>1 (1)</td>
<td>0</td>
</tr>
<tr>
<td>Tenderness</td>
<td>3 (2)</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

*Data are expressed as No. (%) of patients experiencing local and systemic symptoms in the 7 days following vaccination based on the most severe responses as reported on the diary cards. Severity was graded as mild (no interference with daily activities), moderate (some limitation of activity due to the symptom), or severe (the symptom prevents normal daily activity).
Protection Against Influenza Illness
Participants in this study were followed throughout the subsequent influenza season with weekly telephone calls and instructed to return to the study clinics for any acute respiratory illness, at which time a nasopharyngeal swab for viral culture was obtained. A total of 116 such cultures were obtained, 43 in the placebo group, 39 in the 75-µg vaccine group, and 34 in the 135-µg vaccine group. The primary efficacy end point for this study was the development of culture-confirmed influenza illness meeting the influenza-like illness case definition of the CDC; ie, presence of fever higher than 37.7°C (99.8°F) and sore throat, cough, or both.

There were a total of 13 positive cultures for influenza in the study population, of which 10 were influenza A (all of which were confirmed as influenza H3) and 3 were influenza B. Sequence analysis of these viruses further revealed that all of the 10 H3N2 isolates were A/California/7/2004- like. Of these 13 cases, 9 (69%) occurred in individuals meeting the CDC influenza illness case definition. The rates of culture-positive influenza illness, the prespecified primary efficacy end point, were 7 (4.6%) of 153 placebo recipients, 2 (1.4%) of 150 recipients of the 75-µg vaccine dose, and 0 of 151 recipients of the 135-µg vaccine dose. Among the 4 positive cultures in individuals who did not meet the CDC case definition, 1 occurred in a placebo recipient, 2 occurred in recipients of the 75-µg vaccine, and 1 occurred in a recipient of the 135-µg vaccine, so the rates of participants with a positive culture associated with any acute respiratory illness were 8/153 (5.2%), 4/150 (2.7%), and 1/151 (0.7%) in placebo, 75-µg vaccine, and 135-µg vaccine recipients, respectively.

COMMENT
We evaluated the safety, immunogenicity, and efficacy of a trivalent recombinant hemagglutinin (rHA0) vaccine. We have shown that the rHA0 vaccine is well tolerated in healthy adults and immunogenic at both doses evaluated, and we obtained preliminary evidence of protection against influenza infection and disease. The safety data generated in this study are consistent with the safety profile observed in previous studies of rHA0 vaccine.3 8 These vaccines have been well tolerated at all doses administered and are associated with low rates of local reactions. Because of widespread shortages of licensed TIV in the United States during the 2004-2005 season, we were unable to perform a direct comparison of trivalent rHA0 vaccines and TIV. In other studies of TIV in healthy adult populations, pain at the injection site has been reported in 54% to 67% of recipients,8, 11 and systemic symptoms have been similar to placebo, suggesting that the trivalent rHA0 vaccine and TIV have a similar safety profile. This is consistent with previous direct comparisons of rHA0 and TIV suggesting similar rates of local reactions between the 2 vaccines.3 7 11

Both doses of the recombinant hemagglutinin vaccine evaluated induced serum HAI antibody responses to all 3 components (H1, H3, and B) in the majority of recipients. As expected, there were no differences in either the frequency of responses or the postvaccination geometric mean titer to influenza A/Wyoming (H3N2) between the 75-µg and 135-µg doses, since both doses contain the same 45 µg of the H3 component. Significant differences in both the frequency and the magnitude of the HAI response were demonstrated for both the H1 and B components. Although the responses to the 75-µg dose exceeded the European Union criteria for influenza vaccine licensure,12 the 135-µg dose (45 µg of each component) was more immunogenic and might be predicted to provide greater or longer-lasting protection. Therefore, further development of the rHA0 vaccine should use a formulation of 45 µg per component, and future studies should directly compare the safety and immunogenicity of this dose with that of TIV. A major advantage of the rHA0 approach is that these doses are well within the production capacity of the system at an economically and logistically feasible scale.

We also found that recipients of the rHA0 vaccine had reduced rates of culture-positive CDC-defined influenza-like illness compared with placebo recipients, although the study was small. When considering both vaccine groups combined, the cumulative incidence of culture positive CDC-defined influenza-like illness was reduced by 86%. For comparison, in a recently reported study conducted in the same influenza season as was our study, the efficacy of TIV in healthy adults against culture-confirmed influenza meeting a similar case definition was 77% (95% confidence interval, 37%-92%).11

The majority of cases in this study were due to influenza A, and all of the influenza A viruses isolated in this study that were further subtyped were of the H3N2 subtype, consistent with the report that 98.5% of all influenza A viruses typed in the United States during the 2004-2005 season were H3N2.11 In addition, all of the influenza A (H3N2) viruses isolated from

Table 3. Serum Hemagglutination-Inhibiting Antibody Response to Vaccination

<table>
<thead>
<tr>
<th>Influenza Subtype</th>
<th>Placebo (n = 151)</th>
<th>75-µg Vaccine (n = 150)</th>
<th>135-µg Vaccine (n = 150)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A/New Caledonia/20/99(H1N1)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Prevaccination*</td>
<td>26.4 (19.9-35.0)</td>
<td>23.9 (18.0-31.7)</td>
<td>22.0 (16.6-29.2)</td>
</tr>
<tr>
<td>Postvaccination*</td>
<td>28.8 (22.8-36.4)</td>
<td>115.6 (91.5-146.2)</td>
<td>206.0 (163.0-260.5)</td>
</tr>
<tr>
<td>Response, %</td>
<td>3</td>
<td>51</td>
<td>67</td>
</tr>
<tr>
<td>A/Wyoming/3/03(H3N2)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Prevaccination*</td>
<td>72.8 (56.4-93.9)</td>
<td>65.5 (50.7-84.6)</td>
<td>74.2 (57.5-95.8)</td>
</tr>
<tr>
<td>Postvaccination*</td>
<td>68.9 (57.9-81.9)</td>
<td>933.6 (784.4-1111.2)</td>
<td>1028.7 (864.3-1224.5)</td>
</tr>
<tr>
<td>Response, %</td>
<td>11</td>
<td>81</td>
<td>77</td>
</tr>
<tr>
<td>B/Jiangsu/10/03</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Prevaccination*</td>
<td>6.1 (5.1-7.3)</td>
<td>6.4 (5.4-7.6)</td>
<td>5.5 (4.6-6.5)</td>
</tr>
<tr>
<td>Postvaccination*</td>
<td>5.7 (4.7-6.9)</td>
<td>33.4 (27.6-40.4)</td>
<td>74.9 (61.9-90.6)</td>
</tr>
<tr>
<td>Response, %</td>
<td>4</td>
<td>65</td>
<td>92</td>
</tr>
</tbody>
</table>

*Four-fold or greater increase.
†Day 0 (prevaccination) and day 28 (postvaccination) geometric mean titers (95% confidence intervals).

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BACULOVIRUS-EXPRESSED HEMAGGLUTININ INFLUENZA VACCINE

participants in this trial were genetically similar to influenza A/California/7/2004, a significant antigenic variant which re-acts poorly with antisera from persons who received the 2004-2005 for-mulation of TIV.14 as were 75% of influ-enza A(H3N2) isolates throughout the United States.15 It has been suggested that the neuraminidase component of vaccine may be important for protection in situ-ations in which there is not a close anti-genic match in the hemagglutinin.15 The current study suggests that it is possible to generate a substantial amount of pro-tection in an immunologically primed population against influenza with a pure hemagglutinin vaccine, even in the presence of significant antigenic drift.

The development of functional anti-body responses and the preliminary evi-dence of protective efficacy of the vaccine suggest that in adults, the minor differ-ences in HA glycosylation seen in insect cells compared with mammalian cells, the presence of HA as an uncleaved precur-sor, and the lack of neuraminidase in the rHAO vaccine do not have a major effect on the actual protection provided by the vaccine. Although this study and other studies in adults have shown excellent HAI and neutralizing antibody responses to rHAO vaccines,1,2 further studies in im-munologically naive young children are clearly needed.

The use of recombinant DNA tech-niques to express proteins in cell cul-ture has been a successful approach for generation of highly effective vaccines for the prevention of hepatitis B virus and human papillomavirus. Among the avail-able expression technologies, recombi-nant baculovirus is especially well suited for production of influenza vaccine be-cause the rapidity with which genes can be cloned and inserted into this vector facilitates updating the vaccine at regu-lar intervals. In addition, the extraordi-narily high yields of protein possible in this system provide the opportunity to use much higher and potentially more effective doses of vaccine. Expression of the HA protein in insect cells using recom-binant baculovirus also avoids the need to work with potentially patho-genic live influenza viruses and the at-tendant bioccontainment issues that would be a particular concern for gen-eration of pandemic vaccines.8 The pre-liminary demonstration of protective ef-ficacy in adults provides further support for the development of this promising ap-proach for prevention of seasonal and pandemic influenza.

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Author Contributions: Dr Treanor had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Study concept and design: Treanor, Schiff, Hayden, Cox.

Acquisition of data: Treanor, Schiff, Hayden, Brady, Hay, Meyer.

Analysis and interpretation of data: Treanor, Hayden, Hay, Holden-Wiltse, Liang, Gilbert, Cox.

Drafting of the manuscript: Treanor, Meyer, Gilbert, Cox.

Critical revision of the manuscript for important in-tellectual content: Treanor, Schiff, Hayden, Brady, Hay, Meyer, Holden-Wiltse, Liang, Cox.

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