Vaccine-associated reduction in symptom severity among patients with influenza A/H3N2 disease

Authors:

Robert G. Deiss^{1,2,3}, John C. Arnold¹, Wei-Ju Chen^{2,3}, Sara Echols^{1,2,3}, Mary P. Fairchok^{2,3,4}, Christina Schofield⁴, Patrick J. Danaher⁵, Erin McDonough^{3,6}, Michelande Ridoré^{2,3}, Deepika Mor^{2,3}, Timothy H. Burgess⁷, Eugene V. Millar^{2,3}

¹ Naval Medical Center of San Diego, San Diego, CA

² Infectious Disease Clinical Research Program, Department of Preventive Medicine and Biostatistics, Uniformed Services University, Bethesda, MD

³ The Henry M. Jackson Foundation for the Advancement of Military Medicine, Rockville, MD

⁴ Madigan Army Medical Center, Tacoma, WA

⁵ San Antonio Military Medical Center, San Antonio, TX

⁶ Naval Health Research Center, San Diego, CA

⁷ Walter Reed National Military Medical Center, Bethesda, MD

Keywords: influenza; vaccine; severity; military

Running title: Flu vaccine and A/H3N2 symptom severity

Corresponding Author:

Robert G. Deiss, MD
Assistant Professor
Uniformed Services University of the Health Sciences
Department of Preventive Medicine & Biostatistics
Infectious Disease Clinical Research Program
Naval Medical Center of San Diego
Division of Infectious Diseases
34800 Bob Wilson Drive
San Diego, CA 92134
(619) 532-6189 (tel)
(619) 532-7478 (fax)
robert.g.deiss.ctr@mail.mil

Alternate Corresponding Author:

Eugene V. Millar, PhD
Infectious Disease Clinical Research Program
Uniformed Services University of the Health Sciences
Department of Preventive Medicine & Biostatistics
11300 Rockville Pike Suite 1211
Rockville, MD 20852
Office: 301.816.8451
Fax: 301.816.8406
emillar@idcrp.org

Key points: Among patients with influenza A/H3N2 infection, prior receipt of seasonal influenza vaccine was associated with a reduction in the severity and duration of respiratory and systemic symptoms.

Clinical Trials Registration: This study is registered as a clinical trial with the U.S. National Institute of Health, NCT01021098.

Note: Drs. Arnold, Burgess and Danaher are service members (or employees of the U.S. Government). This work was prepared as part of their official duties. Title 17 U.S.C. §105 provides that 'Copyright protection under this title is not available for any work of the United States Government.' Title 17 U.S.C. §101 defines a U.S. Government work as a work prepared by a military service member or employee of the U.S. Government as part of that person's official duties.

Total Word Count: 2,792

ABSTRACT (word count 272)

Background: The moderate level of protection conferred by influenza vaccines is well-known, but the vaccine's ability to attenuate symptom severity among vaccinated individuals (i.e., vaccine failures) has not been established.

Methods: We enrolled otherwise healthy adults who presented with influenza-like illness (ILI) at five US military hospitals between 2009-2014. Influenza was diagnosed and subtyped by PCR. Individual and composite severity scores were compared between those who had vs. had not received the seasonal influenza vaccine >14d prior to enrollment.

Results: A total of 155 cases of influenza (A/H1N1, n=69; A/H3N2, n=66; A/untyped, n=3; B, n=17) were identified, of whom 111 (72%; A/H1N1, n=44; A/H3N2, n=52; A/untyped, n=3; B, n=12) had been vaccinated. Women were significantly less likely to be vaccinated than men (49% vs. 89%; $p<0.01$). In multivariate analysis, vaccinated individuals were significantly less likely to report a fever $>101^{\circ}\text{F}$ (OR 0.24; 95%CI [0.10, 0.62]) and more likely to report myalgias (OR 3.31; 95% CI [1.22, 8.97]) than vaccinated individuals. Among patients with A/H3N2 infection, upper respiratory and total symptom severity scores were significantly lower for vaccinated patients during the first two days of illness, and differences in total symptom severity persisted over seven days ($p<0.05$ for all comparisons). Differences across additional symptom categories (lower respiratory and systemic) were also observed throughout seven days of illness in bivariate analyses. Differences in symptom severity were not observed between vaccinated and unvaccinated participants with A/H1N1 infection.

Conclusions: Among patients with A/H3N2 infection, receipt of seasonal influenza vaccine was associated with reduced symptom severity. Patient-centered discussion about the benefits of influenza vaccination should be expanded to include the possibility that the vaccine could attenuate symptoms.

INTRODUCTION

Acute respiratory infections (ARI) are a significant cause of morbidity worldwide, with influenza alone accounting for an estimated 1 billion infections and 250,000-500,000 deaths each year [1]. As congregate military populations (e.g. trainees, shipboard personnel, and deployed personnel) are at increased risk for influenza outbreaks, annual influenza immunization is required of all active-duty military personnel. Nonetheless, influenza is still a major public health and military concern: outbreaks still occur among highly vaccinated military populations [2], as the vaccine confers only moderate (60-80%) protection [3-5].

Even with moderate effectiveness, annual vaccination remains the best available strategy for the prevention of influenza and its potential complications. Receipt of influenza vaccine has been associated with fewer hospitalizations [6], decreased risk of ICU admission or death among adults already hospitalized [7] and decreased risk of ICU admission in children [8]. However, the vaccine's relative effectiveness against each of the subtypes in the vaccine is poorly understood, and available data vary widely [3, 9-11]. Vaccination reduces the risk of influenza and its complications, but infections among recently vaccinated individuals (i.e. vaccine failures) occur frequently. Still, the extent to which vaccination reduces the severity of disease is largely unknown. A phase III trial in children demonstrated that the efficacy of quadrivalent inactivated influenza vaccine was higher against moderate-to-severe (73%) disease than disease of any severity (55%) [12]. Elsewhere, an observational study among elderly patients reported a reduction in symptom severity among vaccinated as compared to unvaccinated individuals [13].

Until now, the ability of influenza vaccine to attenuate symptoms among young, otherwise healthy adults has not been described. We therefore examined the effects of influenza vaccination on symptom severity among young healthy adults prospectively enrolled in our

natural history of influenza study. We hypothesized that among patients with influenza, those who had received seasonal influenza vaccine would experience a shorter and less severe course of symptoms than those who had not been vaccinated.

METHODS

Since 2009, the Acute Respiratory Infection Consortium (ARIC) of the Infectious Disease Clinical Research Program has enrolled otherwise healthy military personnel and beneficiaries into an observational, longitudinal study of influenza-like illness (ILI). Patients are enrolled at one of five military treatment facilities across the continental United States: Naval Medical Center Portsmouth, VA (NMCP); Naval Medical Center San Diego, CA (NMCSD); Madigan Army Medical Center, Tacoma (MAMC); WA; San Antonio Military Health System, TX (SAMMC); and Walter Reed National Military Medical Center, Bethesda, MD (WRNMCC). Patients are eligible if they present within 72 hours after the onset of illness, with symptoms including fever (temperature of 100.4°F or greater as documented at the time of medical evaluation, or by self-report), in addition to sore throat or one of the following respiratory symptoms: cough, sputum production, shortness of breath, or chest pain. Patients without fever who experience chills or night sweats are also eligible if they had a respiratory symptom or sore throat in addition to either headache, myalgia, or weakness/lethargy. Patients with underlying medical conditions (e.g. diabetes, high-risk pregnancy, immunodeficiency, obstructive lung disease, cystic fibrosis, etc.) are excluded.

After obtaining informed consent, patient demographics, clinical symptoms, and vital signs are recorded by clinical research personnel using a standard questionnaire, and a nasopharyngeal swab (Nylon-flocked, Copan Diagnostics, Corona, CA) for respiratory viral detection is collected. Following study enrollment, participants return for visits at three subsequent time points: days 3±1, 7±2 and 28±7; a daily symptom diary, which begins on the first day of illness, is collected

for a total of seven days following ILI onset. Clinical symptoms and vital signs are recorded at each visit. The presence of clinical symptoms and severity of each symptom are recorded both by self-report (diary) or interview based on the scoring system used by Hayden et al[14], with the exception that the word “chills” is substituted for “feverish,” and the term “nasal stuffiness” is not included. As such, symptom scores were as follows: 0 (none); 1 (*mild*: not changing activity or requiring treatment); 2 (*moderate*: requiring some modification in activity and/or medication); and 3 (*severe*: incapacitating, unable to perform normal activities, requiring bed rest and/or medication). Participants are trained by clinical research personnel on the definitions of each score.

All collected specimens for this study were analyzed by real-time reverse transcription polymerase chain reaction (rtRT-PCR) at the Naval Health Research Center (San Diego, CA). For specimens which tested positive for influenza virus, further analysis was conducted to identify viral subtype. Data collected from participants enrolled between November, 2009 and May, 2014 are included in this analysis.

Data analysis

Individuals 18 years or older with laboratory-confirmed influenza, were included in this analysis. Disease severity (via interview or diary) was measured by the severity of individual symptoms, including the presence of a symptom at any severity level (mild, moderate and severe); stratified comparisons were made based on moderate/severe versus none/mild, and severe versus mild/moderate/none. A second analysis involved the calculation of composite scores as the sum scores of individual symptoms in the following four categories: (1) lower respiratory symptoms: cough, breathing difficulty, hoarseness and chest pain, (2) upper respiratory symptoms: earache, runny nose, sore throat and sneezing; (3) systemic symptoms: chills, muscle ache, headache and fatigue; (4) total symptoms: sum of the above three categories.

We compared clinical individual symptom severity recorded at enrollment (interviewed) and daily composite scores for the first 7 days of ILI between those who had and had not received the seasonal influenza vaccine >14 days prior to enrollment. Continuous variables were compared using the Mann-Whitney or *t* test where appropriate, and dichotomous variables with the Pearson χ^2 or Fisher exact test. Two-sided *P* values <0.05 were considered statistically significant.

For all symptom comparisons with *P* values <0.05, we performed multivariate logistic regression to compare vaccinated and unvaccinated individuals, adjusting for sex, influenza season and use of antiviral medication. For composite score comparisons, we constructed a model using multivariate ordinal logistic regression to examine the effect of influenza vaccination on levels of severity scores, again adjusting for sex, influenza season and use of antiviral medication. We utilized ordinal logistic regression as this method would be less sensitive to individual extreme severity scores than other multivariate methods. Ordinal ranges were set as follows: Level 1 (0-3/and 0-9 for total symptom); Level 2 (4-6/10-18); Level 3 (7-9/19-27); and Level 4 (10-12/28-36). For each multivariate model, we performed sensitivity analysis by excluding data collected from pandemic influenza season 2009-10.

Statistical analyses were performed using SAS (Version 9.3; SAS Institute, Cary, NC) and SPSS (Version 19.0; IBM, Armonk, NY). The study was approved by the Infectious Disease Institutional Review Board of the Uniformed Services University of the Health Sciences (IDCRP-045).

RESULTS

Baseline characteristics of participants are shown in Table 1. Of 884 patients with ILI, we identified 157 patients (18%) with influenza. Vaccination status could not be determined for two patients, and they were excluded from analysis. Of the remaining 155 cases, 138 (89%) tested positive for influenza A (A/H1N1, n=69 (50%); A/H3N2, n=66 (48%); A/untyped, n=3 (2%). A total of 111 patients (72%) were immunized in the season of enrollment; 53.2% (n=59) received inactivated vaccine and 47% (n=52) received live attenuated vaccine. The distribution of influenza types (A vs. B) did not differ between vaccinated and unvaccinated groups.

The median (interquartile range, IQR) duration between last receipt of vaccine and ILI onset was 132 (IQR 102-158) days. The intervals differed significantly ($p=0.03$) when stratified by the viral type/sub-type associated with illness: A/H1N1 (median: 123; [IQR: 99-154.5]), A/H3N2 (median 139.5; IQR [113-165.5]), and B (median 101; [IQR 64.5-137.5]). Among individuals vaccinated within the current season of infection, the median number of doses of vaccine received in the past 5 years was 5 (IQR [4,5]; see table 1); there was no difference when stratified by viral type/sub-type associated with illness.

Women, most of whom were dependents of active-duty spouses, were less likely to be vaccinated compared with men (49.3% vs. 88.6%, $p<0.01$). Vaccinated and unvaccinated participants were prescribed antiviral medications with similar frequency; however, when analyzed by influenza subtype, vaccinated subjects infected with A/H1N1 were more likely to have received antiviral prescriptions than those who had not been vaccinated (52.3% vs. 28.0%, respectively; $p=0.05$). No such differences were observed for those with infection due to influenza A/H3N2 or type B.

Symptom comparisons at baseline enrollment are displayed in Table 2. In multivariate analysis, vaccinated individuals were significantly less likely to report a fever $>101^{\circ}$ F (OR 0.24; 95%CI

[0.10, 0.62]) and more likely to report myalgias (OR 3.31; 95% CI [1.22, 8.97]). With regard to symptom duration, vaccinated individuals experienced significantly fewer days of fatigue, dyspnea and dizziness; in turn, they did not experience a longer duration of myalgias (Table 2).

Measures of disease severity, as determined by median composite scores, were derived from self-collected diary data and are presented in Figure 1 and Supplemental Table 1. In multivariate analysis, no severity differences between vaccinated and unvaccinated participants were noted when considering all cases of influenza (Panel A) or A/H1N1 cases (Panel B). However, after seven days of illness, upper/lower respiratory and total symptom severity scores were significantly lower among vaccinated participants in bivariate analysis (Panel A). With respect to individuals with A/H3N2 infection, our multivariate analysis showed significantly lower median composite scores for vaccinated patients than unvaccinated patients on the first day of illness with respect to upper/lower respiratory and total symptoms (Panel C). Our analysis further demonstrated that bivariate differences in lower respiratory and total symptom severity persisted through the first seven days of illness (Panel C). Among patients with influenza B infection, differences in symptom severity did not achieve statistical significance (data not shown).

Using interview-collected data as a source of validation, similar trends were observed. For influenza overall, unvaccinated individuals were more likely to report moderate/severe chest pain (43.2% vs. 21.6%, $p=0.007$), fatigue that was either moderate/severe (86.4% vs. 65.8%, $p=0.010$) or severe (47.7% vs. 30.6%, $p=0.045$), and severe rhinorrhea (27.3% vs. 13.5%, $p=0.042$) at the initial study visit (Table 3). These differences, however, did not retain statistical significance in multivariate analysis. No differences were observed for A/H1N1 infection (Table 4). Unvaccinated patients with A/H3N2 were significantly more likely to report moderate/severe sneezing (50.0% vs. 21.2%, $p=0.045$), chest pain (50.0% vs. 13.5%, $p=0.007$), appetite loss that

was either moderate/severe (85.7% vs. 48.1%, $p=0.015$) or severe (57.1% vs. 21.1%, $p=0.008$) and severe fatigue (64.3% vs. 30.8%, $p=0.022$) (Table 5). In multivariate analysis, vaccinated individuals with A/H3N2 infection were significantly less likely to report moderate/severe chest pain (OR 0.17; 95%CI [0.03, 0.89]).

Sensitivity analyses performed on all multivariate models (individual and composite scores) did not produce any statistically significant differences from the original models (Supplemental Table 2).

DISCUSSION

In this longitudinal, observational study of influenza, we provide the first evidence that annual recent receipt of influenza vaccine can attenuate the course of disease among young, otherwise healthy individuals. In bivariate and multivariate analysis, vaccinated individuals who were nonetheless infected with influenza A/H3N2 reported decreased severity and duration of numerous symptoms when compared to unvaccinated individuals, and these differences persisted for up to seven days. Thus, our findings suggest that influenza vaccine, while moderately effective in preventing disease outright, mitigates the perceived severity and duration of symptoms, while potentially providing benefit in lessening the likelihood of severe disease and/or complications due to influenza.

Demonstrations of the impact of vaccination on the clinical course of infection are not limited to influenza alone. Nearly fifty years ago, receipt of an experimental respiratory syncytial virus vaccine was associated with a more severe illness among subsequently infected children [15], and typical manifestations of measles infection were re-classified as “modified measles” among recipients of inactivated measles vaccine [16]. More recently, receipt of varicella-zoster vaccine has been shown to attenuate disease severity in breakthrough cases [17]. Our study suggests

that similar disease attenuation may occur with influenza vaccination, at least in A/H3N2 infection. We did not observe such differences with A/H1N1, though the treatment patterns we observed with respect to A/H1N1 cases, and the variability of vaccine to circulating strain match, may have confounded this association. Moreover, it is interesting to note that individuals who had received influenza vaccine were significantly more likely to report myalgias, though neither increased severity nor duration of this symptom were noted. Whether the presence of myalgias is reported more frequently by vaccine recipients in light of decreased severity of other symptoms (such as inactivity), or whether it is the result of an exuberant inflammatory response, warrants further study.

Influenza vaccine-associated disease attenuation has also been observed in several pediatric studies, though there has been considerable variability in study design and the definition of severity. Jain and colleagues reported decreased “moderate-to-severe” disease in vaccinated children, where severity was described as a combination of body temperature $>39^{\circ}\text{C}$, otitis media, lower respiratory disease or serious extra-pulmonary manifestations [12]. More recently, post-hoc analyses of live attenuated influenza vaccine (LAIV) efficacy trials demonstrated that in $>1,600$ children aged 2-17 years, relative efficacy was higher in the prevention of severe as opposed to mild disease. Variability of these effects by season and influenza types and subtypes was also observed [18].

Our data extend findings from several additional studies conducted among diverse adult populations. Using a 0-3 scale of severity, Henkle and colleagues reviewed twelve symptoms among 58 hospital personnel, of whom 76% had received influenza vaccine. Although there was a trend toward lower symptom scores in vaccinated subjects, no statistically significant differences were observed, potentially due to the small sample size [19]. More recently, Van Wormer conducted a secondary analysis of vaccine efficacy trial data and found that mean

symptom severity scores were 31% lower among adults ≥ 65 years old who had been vaccinated, though differences were not seen in individuals < 65 years old [13].

The comparability of findings from multiple studies continues to be hampered by the lack of a uniform measure for symptom and disease severity due to influenza. While clinically validated severity scores are used for a number of conditions including sepsis [20] and pneumonia [21], no such validated score currently exists for influenza. A number of different scales have been used in the literature, including Hayden I [22], Hayden II [14], Keech [23], ICCSQ [24], and FluiiQ [25]. The Hayden I scale is a multi-symptom, 4-point severity scale used during the clinical trials of neuraminidase inhibitors, particularly zanamavir [14, 22]. Our scale mirrored the Hayden II scale, as described above, and therefore is somewhat consistent with these prior studies of symptom severity.

The strengths of our study include its prospective design, frequent visits during the first week of illness, and the comprehensive collection of laboratory, demographic and clinical data, including vaccination history. While focused on young healthy adults in the military health system, our sample was relatively heterogeneous by gender and ethnicity, and sufficiently large to permit conclusions from even a moderate number of cases. For data collection, and in order to mitigate uncertainties which relate to self-reporting of data, we utilized two methods to assess symptoms, including a self-completed daily diary and an interview-administered questionnaire. Data from these two methods were consistent. All subjects had laboratory-confirmed influenza diagnoses, and subtype information was available on 98% of cases with influenza A infection. Last, the use of composite scores provided an aggregate measure for a diverse group of individual symptoms which may vary considerably between individuals. Moreover, findings were confirmed with multivariate analysis.

Limitations of our study include the potential for selection bias, as all subjects were seeking care for their illness, most commonly in ambulatory settings. Much of our data were self-reported, though this potential bias was minimized by validation of patient reports via in-person interview. We noted important differences between our vaccinated and unvaccinated populations, in that the former tended to be active-duty military members, among whom annual influenza vaccination is mandatory. Lastly, a significantly higher portion of unvaccinated individuals were women, and thus, we adjusted for this variable in multivariate analyses to provide further confidence in our findings. Potential differences between women and men with respect to reporting of symptom severity remain undetermined.

In summary, this is the first study demonstrating the association between recent vaccination and a significant reduction in severity of symptoms among individuals with breakthrough influenza A/H3N2 infection. The mechanism by which this occurs is not known, and future studies examining potential correlates of these clinical outcomes (e.g. viral load, viral shedding and/or pro-inflammatory cytokines) are necessary. In addition, whether this finding can be extended to populations at increased risk for complications due to influenza (e.g. pregnant women, elderly) and whether there are sex-based differences with respect to symptom severity should be evaluated in prospective fashion. In the meantime, the ability of influenza vaccine to attenuate symptoms should be highlighted as a benefit of routine immunization, and widespread dissemination of this message would likely increase overall vaccination coverage.

FUNDING

The work was supported by the Infectious Disease Clinical Research Program (IDCRP), a Department of Defense (DoD) program executed through the Uniformed Services University of the Health Sciences. This project has been funded in whole, or in part, with federal funds from the National Institute of Allergy and Infectious Diseases, National Institutes of Health (NIH), under Inter-Agency Agreement [Y1-AI-5072].

CONFLICT OF INTEREST

All authors. No reported conflicts.

ACKNOWLEDGEMENTS

We are indebted to the study team of clinical research coordinators, laboratory personnel, and data management staff for their dedication to the project. The views expressed in this paper are those of the authors and do not necessarily represent the views of the Uniformed Services University of the Health Sciences, the Department of Defense (DoD) or other federal agencies.

REFERENCES

1. World Health Organization. Influenza (Seasonal) Fact Sheet. 2014. Available at <http://www.who.int/mediacentre/factsheets/fs211/en/>. Accessed August 14, 2015.
2. Aquino TL, Brice GT, Hayes S, Myers CA, McDowell J, White B et al. Influenza Outbreak in a Vaccinated Population—USS Ardent, February 2014. *MMWR*. 2014;63(42):947.
3. Herrera GA, Iwane MK, Cortese M, Brown C, Gershman K, Shupe A et al. Influenza vaccine effectiveness among 50–64-year-old persons during a season of poor antigenic match between vaccine and circulating influenza virus strains: Colorado, United States, 2003–2004. *Vaccine*. 2007;25(1):154-60.
4. Jackson LA, Gaglani MJ, Keyserling HL, Balser J, Bouveret N, Fries L et al. Safety, efficacy, and immunogenicity of an inactivated influenza vaccine in healthy adults: a randomized, placebo-controlled trial over two influenza seasons. *BMC Inf Dis*. 2010;10(1):71.
5. Monto AS, Ohmit SE, Petrie JG, Johnson E, Truscon R, Teich E et al. Comparative efficacy of inactivated and live attenuated influenza vaccines. *N Engl J Med*. 2009;361(13):1260-7.
6. Kostova D, Reed C, Finelli L, Cheng P-Y, Gargiullo PM, Shay DK et al. Influenza illness and hospitalizations averted by influenza vaccination in the United States, 2005–2011. *PloS one*. 2013;8(6):e66312.

7. Castilla J, Godoy P, Domínguez Á, Martínez-Baz I, Astray J, Martín V et al. Influenza vaccine effectiveness in preventing outpatient, inpatient, and severe cases of laboratory-confirmed influenza. *Clin Inf Dis*. 2013;57(2):167-75.
8. Ferdinands JM, Olsho LE, Agan AA, Bhat N, Sullivan RM, Hall M et al. Effectiveness of influenza vaccine against life-threatening RT-PCR-confirmed influenza illness in US children, 2010–2012. *J Inf Dis*. 2014;210(5):674-83.
9. Ohmit SE, Thompson MG, Petrie JG, Thaker SN, Jackson ML, Belongia EA et al. Influenza vaccine effectiveness in the 2011–2012 season: protection against each circulating virus and the effect of prior vaccination on estimates. *Clin Inf Dis*. 2014;58(3):319-27.
10. Chen Q, Griffin MR, Nian H, Zhu Y, Williams JV, Edwards KM et al. Influenza Vaccine Prevents Medically-Attended Influenza-Associated Acute Respiratory Illness in Adults 50 Years or Older. *J Inf Dis*. 2015;211(7):1045-1050.
11. CDC. CDC Health Advisory Regarding the Potential for Circulation of Drifted Influenza A (H3N2) Viruses. Health Action Network. 2014.
12. Jain VK, Rivera L, Zaman K, Espos RA, Jr., Sirivichayakul C, Quiambao BP et al. Vaccine for prevention of mild and moderate-to-severe influenza in children. *N Engl J Med*. 2013;369(26):2481-91.
13. VanWormer JJ, Sundaram ME, Meece JK, Belongia EA. A cross-sectional analysis of symptom severity in adults with influenza and other acute respiratory illness in the outpatient setting. *BMC Inf Dis*. 2014;14(1):231.
14. Hayden FG, Fritz R, Lobo MC, Alvord W, Strober W, Straus SE. Local and systemic cytokine responses during experimental human influenza A virus infection. Relation to symptom formation and host defense. *J Clin Invest*. 1998;101(3):643-9.
15. Kapikian AZ, Mitchell RH, Chanock RM, Shvedoff RA, Stewart CE. An epidemiologic study of altered clinical reactivity to respiratory syncytial (RS) virus infection in children previously vaccinated with an inactivated RS virus vaccine. *Am J Epidemiol*. 1969;89(4):405-21.
16. Fulginiti VA, Eller JJ, Downie AW, Kempe CH. Altered reactivity to measles virus. Atypical measles in children previously immunized with inactivated measles virus vaccines. *JAMA*. 1967;202(12):1075-80.
17. Vazquez M, LaRussa PS, Gershon AA, Steinberg SP, Freudigman K, Shapiro ED. The effectiveness of the varicella vaccine in clinical practice. *N Engl J Med*. 2001;344(13):955-60.
18. Ambrose CS, Wu X, Caspard H, Belshe RB. Efficacy of live attenuated influenza vaccine against influenza illness in children as a function of illness severity. *Vaccine*. 2014;32(43):5546-5548.
19. Henkle E, Irving SA, Naleway AL, Gaglani MJ, Ball S, Spencer S et al. Comparison of laboratory-confirmed influenza and noninfluenza acute respiratory illness in healthcare personnel during the 2010-2011 influenza season. *Inf Cont Hosp Epidemiol*. 2014;35(5):538-46.
20. Knaus WA, Zimmerman JE, Wagner DP, Draper EA, Lawrence DE. APACHE-acute physiology and chronic health evaluation: a physiologically based classification system. *Crit Care Med*. 1981;9(8):591-7.
21. Flanders WD, Tucker G, Krishnadasan A, Martin D, Honig E, McClellan WM. Validation of the pneumonia severity index. Importance of study-specific recalibration. *J Gen Intern Med*. 1999;14(6):333-40.
22. Hayden FG, Gubareva LV, Monto AS, Klein TC, Elliot MJ, Hammond JM et al. Inhaled zanamivir for the prevention of influenza in families. Zanamivir Family Study Group. *N Engl J Med*. 2000;343(18):1282-9.
23. Keech M, Scott AJ, Ryan PJ. The impact of influenza and influenza-like illness on productivity and healthcare resource utilization in a working population. *Occup Med (Lond)*. 1998;48(2):85-90.
24. Devoulyte K, Sullivan MJ. Pain catastrophizing and symptom severity during upper respiratory tract illness. *Clin J Pain*. 2003;19(2):125-33.

25. Osborne RH, Norquist JM, Elsworth GR, Busija L, Mehta V, Herring T et al. Development and validation of the Influenza Intensity and Impact Questionnaire (FluiiQ). *Value Health*. 2011;14(5):687-99.

TABLES AND FIGURE

Table 1. Baseline Characteristics of Individuals with Laboratory-Confirmed Influenza Diagnosis by Recent Vaccination Status

	All (n=155)	Unvaccinated (n=44)		Vaccinated (n=111)		P-value
Median Age (IQR)	30.6 (24.9-37.3)	29.3	(24.8-38.3)	31.1	(24.9-37.3)	0.61
Mean BMI (SD)	27.6 (5.0)	28.8	(5.5)	27.1	(4.9)	0.07
Median doses of flu vaccine received in the past 5 years (IQR)	5 (2-5)	1	(0-3)	5	(4-5)	<0.01
	N	N	%	N	%	
Sex						
Female	67	34	(50.8)	33	(49.3)	<0.01
Male	88	10	(11.4)	78	(88.6)	
Military activity						
Active Duty	106	6	(5.7)	100	(94.3)	<0.01 ¹
Retired	8	4	(50.0)	4	(50.0)	
Dependent	41	34	(82.9)	7	(17.1)	
Influenza season						
2009-10	9	7	(77.8)	2	(22.2)	0.02
2010-11	56	14	(25.0)	42	(75.0)	
2011-12	16	4	(25.0)	12	(75.0)	
2012-13	38	8	(21.1)	30	(79.0)	
2013-14	36	11	(30.6)	25	(69.4)	
Site of enrollment						
WRNMMC	6	0	(0.0)	6	(100.0)	0.16
NMCSD	39	11	(28.2)	28	(71.8)	
NMCP	62	18	(29.0)	44	(71.0)	
MAMC	29	6	(20.7)	23	(79.3)	
SAMMC	19	9	(47.4)	10	(52.6)	
Ethnicity						

White	98	27	(27.6)	71	(72.5)	0.82 ¹
Black	32	8	(25.0)	24	(75.0)	
Asian	11	4	(36.4)	7	(63.6)	
Other	12	4	(33.3)	8	(66.7)	
Missing	2	1		1		
Obesity						
Yes	104	24	(23.1)	80	(76.9)	0.10
No	38	14	(36.8)	24	(63.2)	
Missing	13	6		7		
Smoking status						
Current	23	5	(21.7)	18	(78.3)	0.68
Former	29	8	(27.6)	21	(72.4)	
Never	101	31	(30.7)	70	(69.3)	
Missing	2	0		2		
Influenza Type						
A	138	39	(28.3)	99	(71.7)	0.92 ²
H1N1	69	25	(36.2)	44	(63.8)	
H3N2	66	14	(21.2)	52	(78.8)	
Untyped	3	0	(0.0)	3	(100.0)	
B	17	5	(29.4)	12	(70.6)	
Hospitalized						
Yes	11	4	(36.4)	7	(63.6)	0.48 ¹
No	136	35	(25.7)	101	(74.3)	
Missing	11	5		3		
Prescribed anti-viral						
Yes	52	13	(25.0)	39	(75.0)	0.51
No	103	31	(30.1)	72	(69.9)	

IQR: interquartile range; BMI: body mass index; SD: standard deviation

¹ P-value of exact test.

² Comparison between influenza A and B

Table 2. The presence of ILI symptoms and duration of each symptom between influenza vaccinated and unvaccinated cases (all influenza cases)

Symptom	Presence of Symptoms					Median duration with Symptoms ¹ (IQR)			
	Bivariate analysis			Multivariate analysis		Bivariate analysis			
	Unvaccinated n=44	Vaccinated n=111	P-value	OR ²	95% CI	Unvaccinated n=44	Vaccinated n=111	P-value ³	
Chills	34 (77.3)	92 (82.9)	0.419			3 (2, 5)	4 (2.5, 5)	0.550	
Cough	44 (100)	110 (99.1)	1.000 ⁴			7 (6, 7)	7 (6, 7)	0.179	
Rhinorrhea	35 (79.6)	88 (79.3)	0.971			5.5 (4, 7)	5 (3, 7)	0.451	
Earache	23 (52.3)	47 (42.3)	0.263			2 (0, 4)	1 (0, 4)	0.281	
Sneezing	25 (56.8)	57 (51.4)	0.539			3 (2, 5)	3 (0, 5)	0.842	
Sore throat	31 (70.5)	88 (79.3)	0.241			4 (2, 6)	5 (2, 6)	0.862	
Eye issues	19 (43.2)	47 (42.3)	0.924			1.5 (0, 4)	2 (0, 4)	0.760	
Nausea	19 (43.2)	40 (36.0)	0.409			1 (0, 3)	1 (0, 3)	0.646	
Vomiting	5 (11.4)	17 (15.3)	0.525			0 (0, 0)	0 (0, 0)	0.446	
Diarrhea	2 (4.6)	12 (10.8)	0.352 ⁴			0 (0, 1)	0 (0, 1)	0.245	
Hoarseness	30 (68.2)	82 (73.9)	0.475			5 (2, 6)	4 (2, 6)	0.875	
Joint pain	28 (75.7)	89 (80.9)	0.494			4 (2, 6)	4 (2, 6)	0.706	
Dyspnea	28 (63.6)	56 (50.5)	0.137			4 (1, 6)	2 (0, 5)	0.013**	
Myalgia	30 (68.2)	93 (83.8)	0.031**	3.31**	(1.22, 8.97)	4 (2, 6)	4 (2, 6)	0.650	
Headache	37 (84.1)	99 (89.2)	0.383			4 (3, 6)	4 (3, 5)	0.371	
Fatigue	44 (100)	102 (91.9)	0.061 ⁴	N/A ⁵		6.5 (5, 7)	5 (4, 7)	0.016**	
Dizziness	22 (50.0)	42 (37.8)	0.166			2.5 (0, 5)	1 (0, 4)	0.037**	
Appetite loss	37 (84.1)	91 (82.0)	0.755			5 (3, 7)	5 (3, 6.5)	0.764	
Abdominal pain	16 (36.4)	36 (32.4)	0.640			1 (0, 4)	0 (0, 2)	0.069	
Chest pain	25 (56.8)	51 (46.0)	0.222			3.5 (0, 5)	1 (0, 4)	0.057	
Fever >101°F	31 (70.5)	52 (46.9)	0.008**	0.24**	(0.10, 0.62)	N/A N/A	N/A N/A	N/A	

¹Any severity.

²Odds ratios from multivariate logistic regression, adjusting for sex, influenza season and use of antiviral drugs. MVA is only performed on symptoms with p-value <0.05 in bivariate analysis.

³P-value of exact test

⁴By Mann-Whitney U test; equal variances assumed.

IQR: interquartile range, N/A: Not Available, 95% CI: 95% confidence interval, **p<0.05.

Table 3. Comparison of symptom severity at study enrollment among individuals with influenza infection of any type.

	Moderate/Severe (vs. None/Mild)					Severe only (vs. None/Mild/Moderate)				
	Bivariate analysis			Multivariate analysis		Bivariate analysis			Multivariate analysis	
	Unvaccinated	Vaccinated	P-value	OR ²	95% CI	Unvaccinated	Vaccinated	P-value	OR ²	95% CI
All influenza cases	n=44	n=111				n=44	n=111			
Chills	25 (56.8)	60 (54.1)	0.755			11 (25.0)	20 (18.0)	0.327		
Cough	36 (81.8)	86 (77.5)	0.552			20 (45.5)	42 (37.8)	0.383		
Rhinorrhea	27 (61.4)	58 (52.3)	0.304			12 (27.3)	15 (13.5)	0.042**	0.55	(0.20, 1.48)
Earache	12 (27.3)	30 (27.0)	0.975			4 (9.1)	11 (9.9)	1.000 ¹		
Sneezing	11 (25.0)	24 (21.6)	0.650			3 (6.8)	10 (9.0)	0.760 ¹		
Sore throat	21 (47.7)	54 (48.7)	0.918			8 (18.2)	20 (18.0)	0.981		
Eye issues	9 (20.5)	18 (16.2)	0.531			3 (6.8)	6 (5.4)	0.714 ¹		
Nausea	10 (22.7)	19 (17.1)	0.419			2 (4.6)	6 (5.4)	1.000 ¹		
Vomiting	1 (2.3)	8 (7.2)	0.447 ¹			0 (0)	2 (1.8)	1.000 ¹		
Diarrhea	1 (2.3)	3 (2.7)	1.000 ¹			0 (0)	1 (0.9)	1.000 ¹		
Hoarseness	16 (36.4)	43 (38.7)	0.784			7 (15.9)	15 (13.5)	0.700		
Joint pain	21 (56.8)	62 (56.4)	0.967			12 (32.4)	32 (29.1)	0.701		
Dyspnea	15 (34.1)	28 (25.2)	0.266			5 (11.4)	11 (9.9)	0.789		
Myalgia	27 (61.4)	72 (64.9)	0.682			14 (31.8)	34 (30.6)	0.885		
Headache	31 (70.5)	71 (64.0)	0.442			14 (31.8)	28 (25.2)	0.405		
Fatigue	38 (86.4)	73 (65.8)	0.010**	0.423*	(0.15, 1.16)	21 (47.7)	34 (30.6)	0.045**	0.73	(0.32, 1.70)
Dizziness	14 (31.8)	22 (19.8)	0.111			6 (13.6)	8 (7.2)	0.208		
Appetite loss	32 (72.7)	62 (55.9)	0.053*			17 (38.6)	28 (25.2)	0.097		
Abdominal pain	8 (18.2)	13 (11.7)	0.289			1 (2.3)	3 (2.7)	1.000 ¹		
Chest pain	19 (43.2)	24 (21.6)	0.007**	0.48*	(0.20, 1.14)	7 (15.9)	11 (9.9)	0.293		

¹ P-value of exact test.

² Odds ratios from multivariate logistic regression, adjusting for sex, influenza season and use of antiviral drugs. Multivariate analysis is only performed on symptoms with p-value <0.05 in bivariate analysis.

95% CI: 95% confidence interval, **p<0.05

Table 4. Comparison of symptom severity at study enrollment among individuals with pH1N1 infection

	Moderate/Severe (vs. None/Mild)					Severe only (vs. None/Mild/Moderate)				
	Bivariate analysis			Multivariate analysis		Bivariate analysis			Multivariate analysis	
	Unvaccinated	Vaccinated	P-value	OR ²	95% CI	Unvaccinated	Vaccinated	P-value	OR ²	95% CI
	n=25	n=44				n=25	n=44			
Chills	14 (56.0)	23 (52.3)	0.765			6 (24.0)	11 (25.0)	0.926		
Cough	19 (76.0)	36 (81.8)	0.564			12 (48.0)	22 (50.0)	0.873		
Rhinorrhea	11 (44.0)	20 (45.5)	0.907			5 (20.0)	5 (11.4)	0.327		
Earache	5 (20.0)	11 (25.0)	0.636			2 (8.0)	3 (6.8)	1.000 ¹		
Sneezing	3 (12.0)	9 (20.5)	0.515 ¹			1 (4.0)	5 (11.4)	0.406 ¹		
Sore throat	9 (36.0)	22 (50.0)	0.261			4 (16.0)	10 (22.7)	0.756 ¹		
Eye issues	4 (16.0)	3 (6.8)	0.245 ¹			1 (4.0)	1 (2.3)	1.000 ¹		
Nausea	6 (24.0)	10 (22.7)	0.904			1 (4.0)	2 (4.6)	1.000 ¹		
Vomiting	1 (4.0)	5 (11.4)	0.406 ¹			0 (0)	1 (2.3)	1.000 ¹		
Diarrhea	0 (0)	2 (4.6)	0.531 ¹			0 (0)	0 (0)	NA		
Hoarseness	10 (40.0)	19 (43.2)	0.797			5 (20.0)	7 (15.9)	0.667		
Joint pain	9 (50.0)	25 (58.1)	0.559			5 (27.8)	14 (32.6)	0.713		
Dyspnea	8 (32.0)	15 (34.1)	0.859			3 (12.0)	5 (11.4)	1.000 ¹		
Myalgia	13 (52.0)	27 (61.4)	0.449			8 (32.0)	14 (31.8)	0.988		
Headache	17 (68.0)	32 (72.7)	0.677			9 (36.0)	10 (22.7)	0.236		
Fatigue	20 (80.0)	30 (68.2)	0.291			10 (40.0)	14 (31.8)	0.493		
Dizziness	7 (28.0)	9 (20.5)	0.475			4 (16.0)	1 (2.3)	0.054 ¹		
Appetite loss	17 (68.0)	29 (65.9)	0.859			7 (28.0)	14 (31.8)	0.740		
Abdominal pain	5 (20.0)	5 (11.4)	0.327			1 (4.0)	1 (2.3)	1.000 ¹		
Chest pain	11 (44.0)	14 (31.8)	0.312			3 (12.0)	7 (15.9)	0.737 ¹		

¹ P-value of exact test.

² Odds ratios from multivariate logistic regression, adjusting for sex, influenza season and use of antiviral drugs. Multivariate analysis is only performed on symptoms with p-value <0.05 in the bivariate analysis.

95% CI: 95% confidence interval, ** $p < 0.05$

Table 5. Comparison of symptom severity at study enrollment among individuals with H3N2 infection

	Moderate/Severe (vs. None/Mild)					Severe only (vs. None/Mild/Moderate)				
	Bivariate analysis			Multivariate analysis		Bivariate analysis			Multivariate analysis	
	Unvaccinated	Vaccinated	P-value	OR ²	95% CI	Unvaccinated	Vaccinated	P-value	OR ²	95% CI
	n=14	n=52				n=14	n=52			
Chills	9 (64.3)	29 (55.8)	0.567			5 (35.7)	7 (13.5)	0.055*		
Cough	13 (92.9)	39 (75.0)	0.269 ¹			6 (42.9)	16 (30.8)	0.394		
Rhinorrhea	12 (85.7)	30 (57.7)	0.066 ¹			5 (35.7)	9 (17.3)	0.135		
Earache	5 (35.7)	18 (34.6)	0.939			1 (7.1)	7 (13.5)	1.000 ¹		
Sneezing	7 (50.0)	11 (21.2)	0.032**	0.39 ³	(0.08, 1.83)	1 (7.1)	4 (7.7)	1.000 ¹		
Sore throat	10 (71.4)	24 (46.2)	0.093			4 (28.6)	7 (13.5)	0.227 ¹		
Eye issues	4 (28.6)	12 (23.1)	0.730 ¹			1 (7.1)	4 (7.7)	1.000 ¹		
Nausea	3 (21.4)	6 (11.5)	0.387 ¹			1 (7.1)	2 (3.9)	0.517 ¹		
Vomiting	0 (0)	1 (1.9)	1.000 ¹			0 (0)	1 (1.9)	1.000 ¹		
Diarrhea	0 (0)	1 (1.9)	1.000 ¹			0 (0)	1 (1.9)	1.000 ¹		
Hoarseness	4 (28.6)	16 (30.8)	1.000 ¹			2 (14.3)	8 (15.4)	1.000 ¹		
Joint pain	10 (71.4)	29 (55.8)	0.290			6 (42.9)	13 (25.0)	0.190		
Dyspnea	6 (42.9)	10 (19.2)	0.067			1 (7.1)	4 (7.7)	1.000 ¹		
Myalgia	10 (71.4)	37 (71.2)	0.984			5 (35.7)	15 (28.9)	0.620		
Headache	11 (78.6)	31 (59.6)	0.227 ¹			4 (28.6)	16 (30.8)	1.000 ¹		
Fatigue	13 (92.9)	34 (65.4)	0.052			9 (64.3)	16 (30.8)	0.022**	0.44	(0.10, 1.84)
Dizziness	4 (28.6)	10 (19.2)	0.473 ¹			1 (7.1)	4 (7.7)	1.000 ¹		
Appetite loss	12 (85.7)	25 (48.1)	0.0151**	0.27	(0.04, 1.72)	8 (57.1)	11 (21.2)	0.008**	0.27*	(0.06, 1.21)
Abdominal pain	3 (21.4)	7 (13.5)	0.431 ¹			0 (0)	1 (1.9)	1.000 ¹		
Chest pain	7 (50.0)	7 (13.5)	0.003**	0.173**	(0.03, 0.89)	3 (21.4)	3 (5.8)	0.104 ¹		

¹ P-value of exact test.

² Odds ratios from multivariate logistic regression, adjusting for sex, influenza season and use of antiviral drugs. Multivariate analysis is only performed on symptoms with p-value <0.05 in the bivariate analysis.

95% CI: 95% confidence interval, **p<0.05.

Supplemental table 1. Multivariate ordinal logistic regression characterizing the effect of influenza vaccine on clinical symptom severity measured by composite scores. Multivariate analysis was performed only among those variables with significant (p<0.05) differences between influenza vaccinated and unvaccinated cases in bivariate analysis (Figure 1).

(A) Among all influenza cases

	Day 6 of illness		Day 7 of illness	
	OR	(95% CI)	OR	(95% CI)
Upper respiratory			1.10	(0.39, 3.08)
Lower respiratory	0.52	(0.22, 1.20)	0.80	(0.32, 2.01)
Total symptom			0.67	(0.27, 1.69)

Note All the models were adjusted for sex, antiviral use and influenza season.

(B) Among cases with H3N2 infection

	Day 1 of illness		Day 2 of illness		Day 4 of illness		Day 5 of illness	
	OR	(95% CI)	OR	(95% CI)	OR	(95% CI)	OR	(95% CI)
Upper respiratory	0.22*	(0.05, 0.92)	0.18*	(0.04, 0.70)				
Lower respiratory	0.17*	(0.04, 0.73)	0.37	(0.09, 1.47)	0.60	(0.16, 2.22)	0.44	(0.11, 1.74)
Systemic symptom	0.26	(0.06, 1.02)						
Total symptom	0.10*	(0.02, 0.49)	0.21*	(0.05, 0.86)			0.38	(0.10, 1.46)

(H3N2 table continued)

	Day 6 of illness		Day 7 of illness	
	OR	(95% CI)	OR	95% CI
Upper respiratory	0.49	(0.11, 2.19)	0.41	(0.08, 2.12)
Lower respiratory	0.39	(0.09, 1.60)	0.51	(0.09, 2.97)
Systemic symptom	0.30	(0.07, 1.29)	0.33	(0.05, 2.22)
Total symptom	0.23	(0.05, 1.01)	0.13*	(0.02, 0.81)

Note All the models were adjusted for sex, antiviral use and influenza season. * p-value<0.05

Supplemental table 2. Sensitivity analysis of multivariate ordinal logistical regression. Cases from the 2009-2010 influenza season were excluded to determine if the effect of vaccination on symptom severity persisted without the pandemic year. Multivariate analysis was performed only among those variables with significant (p<0.05) differences between influenza vaccinated and unvaccinated cases in bivariate analysis (Figure 1).

(A) Among all influenza cases

Composite Scores:

	Day 6 of illness		Day 7 of illness	
	OR	(95% CI)	OR	(95% CI)
Upper respiratory			0.97	(0.34, 2.78)
Lower respiratory	0.54	(0.23, 1.28)	0.83	(0.32, 2.11)
Total symptom			0.57	(0.22, 1.48)

Note All the models were adjusted for sex, antiviral use and influenza season.

(B) Sensitivity analysis not performed for A/H3N2 cases as none were identified in the 2009-2010 season.

Supplemental table 3. Sensitivity analysis of multivariate logistical regression. Cases from the 2009-2010 influenza season were excluded to determine if the effect of vaccination on symptom severity persisted without the pandemic year. Multivariate analysis was performed only among those variables with significant (p<0.05) differences between influenza vaccinated and unvaccinated cases in bivariate analysis (Tables 2 and 3). Models were adjusted for sex, antiviral use and influenza season.

(A) Among all influenza cases

<u>Symptom</u>	<u>Odds Ratio (95%CI)</u>
Myalgias (presence)	3.00 (1.10, 8.15)
Fever (presence)	0.30 (0.12, 0.78)
Fatigue (moderate/severe)	0.50 (0.18, 1.43)
Chest pain (moderate/severe)	0.41 (0.17, 1.00)

(B) Sensitivity analysis not performed for A/H3N2 cases as none were identified in the 2009-2010 season.

Figure 1. Median composite severity scores of vaccinated (blue) and unvaccinated (red) patients on the first seven days of illness.

Figure 1