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Antigen-specific H1N1 influenza antibody responses in acute respiratory tract infections and their relation to influenza infection and disease course^{☆, ☆☆}

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Abstract

Background—Early antibody responses to influenza infection are important in both clearance of virus and fighting the disease. Acute influenza antibody titers directed toward H1-antigens and their relation to infection type and patient outcomes have not been well investigated.

Objective—Using hemagglutination inhibition (HI) assays, we aimed to characterize the H1-specific antibody titers in patients with influenza infection or another respiratory infection before and after the H1N1-pandemic influenza outbreak. Among patients with acute influenza infection we related duration of illness, severity of symptoms, and need for hospitalization to antibody titers.

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Competing interests

The authors declare that they do not have anything to disclose regarding funding or conflict of interest with respect to this manuscript.

Ethical approval

Each participant was personally informed of the protocol and gave written informed consent. This protocol was approved by the institutional review board of Rhode Island Hospital (IRB number 4074-08).

Methods—There were 134 adult patients (average age 34.7) who presented to an urban academic emergency department (ED) from October through March during the 2008–2011 influenza seasons with symptoms of fever and a cough. Nasal aspirates were tested by viral culture, and peripheral blood serum was run in seven H1-subtype HI assays.

Results—Acutely infected influenza patients had markedly lower antibody titers for six of the seven pseudotype viruses. For the average over the seven titers (log units, base 2) their mean was 7.24 (95% CI 6.88, 7.61) compared with 8.60 (95% CI 8.27, 8.92) among patients who had a non-influenza respiratory illness, $p < 0.0001$. Among patients with seasonal influenza infection, titers of some antibodies correlated with severity of symptoms and with total duration of illness ($p < 0.02$).

Conclusion—In patients with acute respiratory infections, lower concentrations of H1-influenza-specific antibodies were associated with influenza infection. Among influenza-infected patients, higher antibody titers were present in patients with a longer duration of illness and with higher severity-of-symptom scores.

Keywords

Antibodies; Seasonal influenza; H1N1 influenza; Bacterial pneumonia

1. Background

Factors that increase the risk of severe influenza infection include host factors such as extremes of age, comorbid illnesses, pregnancy, and obesity [1–3]. Most patients infected with influenza virus either are asymptomatic or develop mild, nonlife-threatening respiratory tract infections. However, with the emergence of the pandemic 2009 H1N1 strain, the majority of serious illnesses and 90% of deaths occurred among young healthy adults who lacked any of these risk factors [4]. Unidentified factors must affect the host's innate immune response and thus lead to severe disease progression.

During the early stages of infection, neutralizing antibody titers are low, but crucial to fighting off infection [5]. Neutralizing antibody responses are induced strongly in healthy individuals and contribute to virus clearance [6]. Hospitalized influenza-infected individuals demonstrate lower total and neutralizing anti-influenza antibody titers than do respiratory syncytial virus-infected control subjects. This difference suggests a protective effect of higher influenza antibody titers [7]. An overall reduction in antibody titers produced during severe acute influenza infection occurs in both human and mouse models [8–10]. Clinically, little is known of how such variations in antibody responses may influence susceptibility to infection by new circulating H1 influenza viruses.

In 2009 a novel pandemic H1N1 influenza A virus (H1N1/09) spared the elderly but caused severe disease in middle-aged individuals, even though this younger population had more-robust preexisting immunity against seasonal H1 strains. One theory for this pattern was that cross-reactive antibodies to the pre-1957 circulating H1N1 virus protected the older population [11,12]. Elevated antibody titers against the H1N1/09 virus were associated with previous seasonal 2007 H1N1 infection, which may have contributed to the lower burden of the 2009 influenza pandemic [13].

Not all prior influenza vaccinations or infections leading to preexisting influenza antibodies may be beneficial. The presence of cross-reactive antibodies to one influenza virus strain can reduce production of B cell antibodies to a second encountered strain [14,15]. Low-affinity influenza antibodies have been detected in individuals with severe concurrent bacterial pneumonia [16]. Preexisting serum antibodies that cross-react with, but do not protect against, the H1N1/09 influenza virus in middle-aged adults may have led to higher disease severity and worse outcomes [16]. Such conflicting data and an overall lack of information on early antibody response to natural infection and patient outcomes indicate a need for further investigation of H1-specific antibody response to various H1 pseudotype viruses in patients with pre- and post- H1N1-pandemic influenza infection.

2. Objectives

We aimed to characterize H1-specific antibody titers before and after the H1N1-pandemic influenza outbreak among patients with influenza infection and patients with other respiratory tract infections. The resulting data allowed us to compare those two groups' antibody responses on individual H1-pseudotype viruses, to compare responses among those viruses, and to compare H1N1/09 patients versus seasonal influenza patients. Among influenza-infected patients, we also explored the relation between H1-specific antibody titers and severity of patient-reported symptoms, total duration of symptoms, and need for hospitalization.

3. Methods

3.1. Study design and setting

This prospective observational study involved patients presenting to the ED with symptoms of influenza-like illness (ILI), which included a cough and a fever [17]. The study setting was an urban academic Level 1 trauma center with an annual census greater than 100,000 patients. The study involved three influenza seasons. The first season spanned the months of January through March of 2009 and included subjects with seasonal influenza infection. The second and third influenza seasons spanned the months of October through March 2009–2010 and 2010–2011. The pandemic H1N1 influenza virus emerged during this time period, and subjects with H1N1 influenza infection were enrolled.

3.2. Selection of participants

Patients presenting to the adult ED, 18 years of age or older, whose chief complaints were fever and cough were approached for enrollment. Patients with symptoms lasting more than 3 days, a history of immunosuppressant medications, a recent diagnosis of pneumonia, or current antibiotics were excluded. The exclusion of patients with symptoms lasting longer than 3 days draws on our previous work on cytokine markers in this group [18] and captures subjects with an acute infection [19]. The primary analysis excluded subjects with a diagnosis of concurrent influenza and bacterial pneumonia infection. Secondary analysis compared those patients with patients who had influenza infection alone.

3.3. Clinical data

Patients completed a survey about their past medical history, current illness, and symptom severity. During this initial ED visit, both nasal washing and blood samples were collected from each patient and vital signs recorded. After enrollment patients were followed by chart review (if admitted to the hospital) or with a telephone survey at four weeks. The follow-up aimed to determine duration of symptoms and the presence of any potential complications. A score for severity of symptoms (SOS) was calculated for each patient as they presented to the ED. The previously validated instrument asked patients to rate, on a scale from 0 (none) to 3 (severe), the severity of seven symptoms: cough, nasal obstruction, sore throat, fatigue, headache, myalgia, and feverishness [20,21].

3.4. Laboratory assays

During January through March of 2009, we used the rapid point-of-care Enzyme-Linked Immunosorbent Assay (ELISA) to screen patients presenting to the ED who were suspected of having an acute infection with influenza. We enrolled only subjects who had a positive test confirming influenza A antigen which included both H3N2 and H1N1 subtypes.

With the emergence of the pandemic 2009 H1N1 virus, the rapid ELISA assay was discontinued because it lacked sensitivity in detecting the novel H1N1 influenza strain [22]. Without a reliable rapid test to identify influenza-infected individuals, we altered the study design to enroll all patients (ages 18–65) who presented with symptoms of ILI. To test for the presence of influenza virus, we used a viral culture technique. In brief, after centrifugation specimens were processed for culture in rhesus monkey kidney cells for two days. Then, using monoclonal antibodies, they were assessed for the presence of viral particles. The Rhode Island State Laboratory confirmed pandemic-H1N1/09-positive cultures by virus particle detection through polymerase chain reaction, used nationally at that time [23].

3.4.1. Pseudotype lentiviral vectors—Recombinant lentiviral vectors expressing a luciferase reporter gene were produced as previously described [24,25]. We used 7 pseudotype viruses expressing H1 antigen, representing over 30 years of H1 influenza virus in circulation in the United States. Pseudotype viruses (psv) were constructed from the following H1N1 influenza strains: A/Brazil/78(BZ78), A/Chile/1/83(CH83), A/Taiwan/1/86(TW86), A/Texas/36/91(TX91), A/Bayern/7/95(BY95), A/Beijing/262/95(BJ95), and A/Solomon Islands/3/06(SI06) (Table 1). The pseudovirus titers were determined by single-round infection of 293A cells [26].

3.4.2. Hemagglutination inhibition (HI) assay—HI assay was performed as described previously [27]. Human sera were treated with receptor-destroying enzyme (RDE) by diluting one part serum with three parts enzyme and incubated overnight at 37°C. This serum-RDE mix was 2-fold serially diluted and run in duplicate wells. Subject serum was incubated with pseudotype virus for 1 h before the addition of red blood cells (RBCs). HI assays were performed in V-bottom 96-well plates using four hemagglutinating units (HAU) of virus and 0.5% turkey or guinea pig RBC. Results were expressed as the reciprocal of the highest dilution of human serum that still produced complete hemagglutination.

3.5. Statistical analysis

To assess the significance of differences between groups, the analysis used the Mann–Whitney test for measured variables and Fisher’s exact test for dichotomous variables. We calculated Pearson correlations (r) between antibody titers (in a logarithmic scale, base 2) and Spearman rank correlations (ρ) for other pairwise relations. The “averaged antibody titer” was the arithmetic average of the titers for the 7 (or 6) pseudotype viruses; analyses used the logarithm (base 2) of that average. Confidence intervals for means and tests of significance for correlations used t -distributions. Odds ratios were estimated by logistic regression. Individual p -values <0.05 were considered statistically significant. The software used for the analyses was Stata, Release 12 (StataCorp LP, College Station, TX).

4. Results

4.1. Study population

A total of 250 patients were screened over the three influenza seasons, and a total of 172 patients were enrolled. Of those enrolled, 38 (22.1%) were excluded from final analysis, leaving a total of 134 patients. Reasons for exclusion were loss to follow-up (68.4%), taking immunosuppressant medication (10.5%), dual diagnosis of influenza and bacterial pneumonia (10.5%), inability to comply with nasal wash protocol (7.9%), and inability to draw blood (2.6%). The 4 dual-diagnosis patients all had infiltrate demonstrated on chest X-ray and were treated with antibiotics for community-acquired pneumonia (CAP).

We enrolled 33 of the patients during the 2008–2009 season and the remaining 101 over the 2009–2010 and 2010–2011 seasons. The average age was 34.7 years, and 61.2% were female. Concerning vaccination history, 34 patients (25.4%) had received that year’s flu vaccine, and 69 (51.5%) reported a history of flu vaccination in the past. Of the 134 patients 53 were confirmed to have influenza A infection: 33 pre-H1N1 outbreak enrolled during the 2008–2009 season (and thus termed seasonal influenza) and 20 laboratory-confirmed pandemic H1N1/09 enrolled over the 2009–2011 seasons. A total of 81 patients enrolled over the 2009–2011 seasons were influenza-culture negative and thus were presumed to be infected with another viral process or to have a bacterial infection, via an infiltrate demonstrated on chest X-ray. No patients upon follow-up experienced any additional complications or infection diagnoses. Table 2 lists the characteristics of the patient population for each influenza year.

The three patient groups designated seasonal influenza, H1N1/09, and non-influenza-infection group were similar in average age (31.1, 29 and 37.9) percentage female (57.6, 50.0 and 64.6) and percentage with a previous history of vaccination (48.5, 50.0 and 53.7). The H1N1/09 group had a lower percentage of patients receiving that year’s immunization (5.0) compared to the other two groups (24.2 and 29.6). All three groups had similar duration of illness in days (10.6, 9.0 and 9.9) and varied slightly in the percentage admitted (12.1, 25.0 and 31.7). Of the non-influenza-infection group 24.7% had the diagnosis of bacterial pneumonia while the remaining 75.3% were diagnosed with a viral syndrome other than influenza.

4.2. Antibody titers and infection type

Fig. 1 contains a scatterplot of all of the patients' antibody levels for each pair of H1-pseudotype viruses. Because of the wide range of serial dilutions, the antibody titers are expressed in a logarithmic scale (base 2). Because of reagent availability, only 101 subjects had their BJ95 H1-antibody titers measured. All 21 pairs of pseudotype viruses had statistically significant correlation; the strongest correlation was between TW86 and TX91 ($r = 0.89$, $p < 0.001$) (Table 3). All but 3 of the 21 p -values were < 0.001 .

Patients with an acute influenza infection (H1N1 or seasonal) had markedly lower mean averaged antibody titer, 7.24 (95% CI 6.88, 7.61), than patients with another respiratory infection, 8.60 (95% CI 8.27, 8.92) $p < 0.0001$ (Fig. 2a). The influenza infection group and the non-influenza group had statistically significant differences on all seven antibody types (Fig. 2b). The pseudotype viruses that had lower antibody concentrations among influenza-infected patients were BZ78 ($p = 0.017$), CH83 ($p < 0.0001$), TW86 ($p = 0.0001$), TX91 ($p < 0.0001$), BY95 ($p < 0.0001$), and SI06 ($p = 0.0003$). Only BJ95 had higher antibody titers among the influenza-infected patients ($p = 0.041$). The influenza-positive and -negative groups were similar in age, sex, and vaccination history, with no statistically significant differences. Table 4 shows odds ratios (ORs) for each antibody type for the diagnosis of an acute influenza infection.

Among the influenza-positive patients, the seasonal and H1N1/09 groups did not differ significantly on age (31.1 vs. 29.0), current influenza vaccination status (33.3% vs. 5.0%), or previous vaccination history (48.5% vs. 50.0%). Patients infected with the pandemic H1N1/09 influenza strain had significantly higher averaged antibody titers than those with seasonal influenza infection ($p = 0.0005$). Antibody types TW86 ($p = 0.0004$), TX91 ($p = 0.0005$), and BY95 ($p < 0.0001$) had significantly higher titers in the H1N1 group, but BJ95 ($p = 0.015$) had lower antibody concentrations (Fig. 3).

4.3. Antibody titers and influenza outcomes

In subgroup analysis of the seasonal-influenza-infected patients, higher severity-of-symptoms scores were associated with higher averaged antibody titers ($\rho = 0.393$, $p = 0.024$) and also with three of the seven antibody types: BZ78 ($\rho = 0.477$, $p = 0.0050$), CH83 ($\rho = 0.509$, $p = 0.0026$), and SI06 ($\rho = 0.594$, $p = 0.0003$). However, longer duration of illness (measured as days with flu-like symptoms) was associated with higher antibody titers only for BZ78 ($\rho = 0.530$, $p = 0.0045$) and CH83 ($\rho = 0.503$, $p = 0.0075$). None of the antibody titers differed significantly between admitted and discharged patients.

In contrast, in the pandemic-H1N1/09-influenza-infected patients, neither SOS scores nor duration of symptoms was significantly correlated with any of the antibody titers. The averaged antibody titers were significantly higher in admitted patients ($p = 0.032$) (Fig. 4).

Among the influenza-positive patients, 4 (not included in the primary analysis) were diagnosed with concurrent bacterial pneumonia. In comparison with the other influenza-positive individuals, these patients had higher mean H1-specific antibody titers for BZ78, 7.25 (95% CI 4.86, 9.64) compared with 5.38 (95% CI 4.90, 5.86) [$p = 0.031$] and for CH83, 9.00 (95% CI 5.56, 12.44) compared with 6.58 (95% CI 6.25, 6.92) [$p = 0.005$].

5. Discussion

To our knowledge, this report is the first to relate H1-specific influenza antibody titers to clinical infection among patients prior to and then during the 2009 pandemic influenza outbreak. In patients with acute influenza infection, six of the seven H1-specific antibody titers were lower than in patients with another cause of acute respiratory illness. This finding suggests that higher antibody titers provide a level of protection against influenza infection or at least protection against clinically relevant symptoms that would lead a patient to seek care in the ED.

During acute H1N1/09 influenza infection, H1 antibody titers were higher among subjects admitted to the hospital than among those sent home from the ED. This was true for the averaged H1 titer and for four of the seven H1-specific antibodies. This result leads to the conclusion that higher titers among influenza-infected patients are related to more-severe influenza infection. Interestingly, among the seasonal-influenza-infected patients the H1 antibody titers correlated with both the severity of patient-reported symptoms and the total duration of symptoms. This further result suggests that antibody titers may relate to both severity of symptoms and duration of illness. The absence of a similar correlation in the H1N1/09-influenza-infected may be due to either the smaller number of subjects in this group ($n = 20$) or the fact that these subjects overall reported less-severe symptoms than their seasonal-influenza counterparts, average SOS score of 8.95 compared with 15.0.

Our patient population was predominantly young with an average age of 35 years, and our initial analysis excluded four patients (one seasonal and 3 H1N1/09) who had concurrent bacterial and influenza infections and were treated for CAP. In a secondary analysis these four subjects had higher H1-specific antibody titers for BZ78 and CH83 than those in the seasonal and H1N1/09 influenza-infection groups. This finding is in line with the other clinical outcomes observed, in which patients with higher H1-specific antibody titers had worse outcomes.

Because samples were available for only 101 patients, testing of the BJ95 antibody was incomplete. Unfortunately, only BJ95 demonstrated antibody titer trends opposite to those of the other 6 types. This unexpected and unexplained pattern warrants further investigation. Additionally we were unable to produce a H1N1/09 pseudotype virus which could have shown correlations to SOS and duration of illness.

Our results are similar to previous observations on hospitalized patients, in whom higher influenza antibody titers were protective against serious illness requiring hospitalization [7]. Our investigation demonstrates this phenomenon among patients treated in the emergency department, the vast majority of whom are not admitted to the hospital. This report goes further by relating antibody responses to both severity of infection and patient outcomes; we observed differences in the seasonal-influenza and the H1N1/09-influenza infection groups. We conclude that influenza-specific antibody titers may serve as a marker of both infection type and outcomes during the influenza respiratory season.

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Fig. 1.

Pairwise scatterplots depict the associations between the titers for the seven H1-specific influenza antibody types. Because of the wide range of serial dilutions, the antibody titers are expressed in a logarithmic scale (base 2). In each of the small rectangles, a point represents one or more patients (because of the granularity of the data), the vertical variable represents the antibody type named in the box on the diagonal in the same row, and the horizontal variable represents the antibody type named in the box on the diagonal in the same column.

Fig. 2.

H1-specific influenza titers among patients who had an acute influenza infection ($n = 53$) or another viral or bacterial respiratory illness ($n = 81$). (A) Antibody titers averaged over the seven antibody types, and (B) antibody titers for each antibody type. Titers are expressed in the logarithmic scale (base 2). (A) Boxes extend from the 25th to the 75th percentile with a line at the median; vertical lines extend from the box to the data values whose distance to the box is closest to, but not greater than, 1.5 times the IQR ($=75\text{th percentile} - 25\text{th percentile}$); data values farther from the box are plotted individually. (B) Symbols represent the mean for

influenza-negative (◆) and influenza-positive (■) patients; vertical lines represent 95 percent confidence intervals. Abbreviations: –, negative and +, positive influenza viral culture.

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Antibody Titers of Influenza Positive Patients

Fig. 3.

H1-specific influenza titers among influenza-positive patients comparing seasonal and H1N1/09 infections. Four antibody types reached statistical significance (TW86, $p = 0.0004$; TX91, $p = 0.0005$; BY95, $p < 0.0001$; and BJ95, $p = 0.015$) with the average of all antibody types also being significant ($p = 0.0005$). The difference on the averaged antibody titer and on four of the seven H1-specific types reached statistical significance. Titers are expressed in the logarithmic scale (base 2). Symbols represent the mean of the seasonal (◆) and H1N1/09 (■) patient groups; vertical lines represent 95 percent confidence intervals. Abbreviations: avg., averaged H1-antibody titer, S, seasonal and H, H1N1/09-infected patients.

Fig. 4.

H1-specific influenza titers among H1N1/09-positive patients by whether discharged from the ED or admitted to the hospital for further treatment. (A) Antibody titers averaged over the seven antibody types, and (B) for each of the four antibody types that showed statistical significance (BZ78, $p = 0.046$; CH83, $p = 0.003$; BJ95, $p = 0.050$; and SI06, $p = 0.036$). Titers are expressed in the logarithmic scale (base 2). (A) Boxes extend from the 25th to the 75th percentile with a line at the median; vertical lines extend from the box to the data values whose distance to the box is closest to, but not greater than, 1.5 times the IQR (=75th

percentile – 25th percentile); data values farther from the box are plotted individually. (B) Symbols represent the mean for discharged (◆) and admitted (■) patients; vertical lines represent 95 percent confidence intervals. Abbreviations: DC, discharged patients and A, admitted patients.

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Table 1

H1N1 vaccine strains used in this study emerged from 1978 to 2006. Depicted are the grouping by antigenic drift, short name, H1N1 influenza strain designation, and years in which the influenza strain was used as part of the yearly vaccine.

	Name	H1N1 influenza strain	Vaccine years
Group 1	BZ78	A/Brazil/78(H1N1)	1982–1984
	CH83	A/Chile/1/83(H1N1)	1985–1986
Group 2	TW86	A/Taiwan/1/86(H1N1)	1986–1992
	TX91	A/Texas/36/91(H1N1)	1992–1996
	BY95	A/Bayern/7/95(H1N1)	1996–1997
Group 3	BJ95	A/Beijing/262/95(H1N1)	1997–2000
	SI06	A/Solomon Islands/3/06(H1N1)	2007–2008

Table 2

Characteristics of patient population.

	2008–2009 season	2009–2010 season	2010–2011 season
Number of patients	33	61	40
Age	31.1 (10.9)	28.5 (11.6)	47.2 (23.5)
Female (%)	57.6	63.9	60.0
Current vaccine (%)	24.2	9.8	50.0
History vaccine (%)	48.5	49.2	57.5
Influenza-positive (%)	100 ^a	29.5 ^b	5.0 ^b
Symptoms PTA	2.6 (0.8)	1.8 (1.2)	3.1 (1.7)
SOS score	15.0 (3.8)	13.8 (3.8)	12.6 (5.0)

Characteristics of the 134 patients by influenza season in which they were enrolled in the study. Means are represented followed by standard deviation in parentheses. Current vaccine represents influenza vaccine received prior to presentation for that year. History vaccine refers to patients that receive yearly influenza vaccination in the past five years.

Symptoms PTA = number symptom days prior to arrival, SOS = severity of symptoms score.

^a All patients were influenza-positive.

^b All patients who were influenza-positive had PCR testing for H1N1 confirmation.

Table 3

Pairwise correlations of titers for the seven H1-specific influenza antibody types. In each cell below the diagonal, the upper number is the Pearson correlation coefficient, and the lower number is the (individual) *p*-value.

	BZ78	CH83	TW86	TX91	BY95	BJ95	SI06
BZ78	1.000						
CH83	0.7403 <i>p</i> < 0.001	1.000					
TW86	0.3693 <i>p</i> < 0.001	0.4866 <i>p</i> < 0.001	1.000				
TX91	0.3976 <i>p</i> < 0.001	0.5256 <i>p</i> < 0.001	0.8944 <i>p</i> < 0.001	1.000			
BY95	0.3193 <i>p</i> < 0.001	0.4529 <i>p</i> < 0.001	0.6851 <i>p</i> < 0.001	0.7022 <i>p</i> < 0.001	1.000		
BJ95	0.6211 <i>p</i> < 0.001	0.5525 <i>p</i> < 0.001	0.2426 <i>p</i> = 0.015	0.2507 <i>p</i> = 0.012	0.2075 <i>p</i> = 0.037	1.000	
SI06	0.3550 <i>p</i> < 0.001	0.5026 <i>p</i> < 0.001	0.7668 <i>p</i> < 0.001	0.7414 <i>p</i> < 0.001	0.6154 <i>p</i> < 0.001	0.3258 <i>p</i> < 0.001	1.000

Table 4

Odds ratios (with 95% confidence intervals) for a diagnosis of influenza. Each H1-specific antibody type has an adjusted odds ratio in the multiple-predictor model (adjusting for the contributions of the other antibody types) and a simple odds ratio in its single-predictor model. The analysis used data from the subjects for whom the results of all seven pseudotype-virus hemagglutination inhibition assays were available ($n = 101$). Titers are serial dilutions, expressed in a logarithmic scale (base 2). For patients who have a middling value of the $\log_2$ titer for each of the pseudotype viruses (BZ78, 6; CH83, 7; TW86, 8; TX91, 8; BY95, 7; BJ 95, 6; SI06, 7), the multiple-predictor model gives 0.576 as the predicted probability of an influenza infection. In the multiple-predictor model, combining all seven antibody types to predict influenza infection, the adjusted odds ratios of CH83, BY95, and BJ95 reached statistical significance. In the single-predictor models, all antibody types except BZ78 and BJ95 had statistically significant odds ratios.

Antibody titers in predicting influenza infection		
Pseudotype virus	Multiple-predictor model ($n = 101$) Adjusted odds ratio (95% CI)	Single-predictor models ($n = 101$) Simple odds ratio (95% CI)
BZ78	0.88 (0.54, 1.43)	0.83 (0.66, 1.04)
CH83	0.34 (0.17, 0.67)	0.57 (0.42, 0.79)
TW86	1.91 (0.76, 4.82)	0.62 (0.47, 0.81)
TX91	0.67 (0.30, 1.48)	0.60 (0.46, 0.79)
BY95	0.35 (0.20, 0.62)	0.43 (0.30, 0.62)
BJ95	3.04 (1.65, 5.62)	1.21 (0.99, 1.49)
SI06	0.97 (0.58, 1.61)	0.68 (0.53, 0.89)