

## Estimated cumulative incidence of pandemic (H1N1) influenza among pregnant women during the first wave of the 2009 pandemic

Salaheddin M. Mahmud MD PhD, Marissa Becker MD, Yoav Keynan MD, Lawrence Elliott MD MSc, Laura H. Thompson MSc, Keith Fowke PhD, Lisa Avery MD MSc, Paul Van Caesele MD, Sande Harlos MD MSc, James Blanchard MD PhD, Magdy Dawood PhD

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### ABSTRACT

**Background:** Hospitalization and lab confirmed cases of H1N1 have been reported during the first wave of the 2009 pandemic but these are not accurate measures of influenza incidence in the population. We estimated the cumulative incidence of pandemic (H1N1) influenza among pregnant women in the province of Manitoba during the first wave of the 2009 pandemic.

**Methods:** Two panels of stored frozen serum specimens collected for routine prenatal screening were randomly selected for testing before (March 2009,  $n = 252$ ) and after (August 2009,  $n = 296$ ) the first wave of the pandemic. A standard hemagglutination inhibition assay was used to detect the presence of IgG antibodies against the pandemic (H1N1) 2009 virus. The cumulative incidence of pandemic (H1N1) influenza was calculated as the difference between the point prevalence rates in the first and second panels.

**Results:** Of the specimens collected in March, 7.1% were positive for the IgG antibodies (serum antibody titre  $\geq 1:40$ ). The corresponding prevalence was 15.7% among the specimens collected in August. The difference indicated a cumulative incidence of 8.6% (95% confidence interval [CI] 3.2%–13.7%). The rate differed geographically, the highest being in the northern regions (20.8%, 95% CI 7.9%–31.8%), as compared with 4.0% (95% CI 0.0%–11.9%) in Winnipeg and 8.9% (95% CI 0.0%–18.8%) in the rest of the province.

**Interpretation:** We estimated that the cumulative incidence of pandemic (H1N1) influenza among pregnant women in Manitoba during the first wave of the 2009 pandemic was 8.6%. It was 20.8% in the northern regions of the province.

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**D**uring the first wave of the pandemic (H1N1) 2009, the province of Manitoba was more severely affected than almost any other Canadian province.<sup>1</sup> Pregnant women in particular had higher rates of laboratory-confirmed infection and of severe illness.<sup>2</sup> However, the number of laboratory-confirmed cases is not an accurate measure of the incidence of influenza in the population. The number and geographic distribution of confirmed cases are influenced by differences in access to medical care, physicians' practices and other factors.<sup>3</sup>

We estimated the cumulative incidence of pandemic (H1N1) influenza among pregnant women in the province of Manitoba during the first wave of the 2009 pandemic. We did this by measuring the point seroprevalence in random samples of pregnant women presenting for routine prenatal screening before and after the first wave.

### Methods

The Manitoba Maternal Serum Screening Program is a province-wide prenatal screening program offered free of charge to all pregnant women between 15 and 22 weeks' gestation.<sup>4</sup> Participation rates in the program are high ( $> 70\%$ ).<sup>5,6</sup> All specimens from the program are tested, and subsequently stored for one year, at the Cadham Provincial Laboratory.

We tested a random sample of 296 serum specimens collected from women presenting for prenatal screening at the end of the first wave (August 2009) using a hemagglutination inhibition assay<sup>7</sup> to detect IgG antibodies against the pandemic strain of the virus (see Appendix 1, available at [www.cmaj.ca/cgi/content/full/cmaj.100488/DC1](http://www.cmaj.ca/cgi/content/full/cmaj.100488/DC1)). Because of concerns about potentially high levels of cross-reactivity with antibodies against pre-pandemic influenza strains,<sup>8</sup> we compared the seroprevalence estimates for the serum specimens collected after the first wave of the pandemic with estimates from a random sample of 252 specimens collected before the first wave (March 2009). Every woman's serum specimen had the same probability of being selected for testing, except for women residing in the less populous northern regions (North Eastman, NOR-MAN, Burntwood and Churchill regional health authorities); serum samples for all of these women were included to improve the precision of seroprevalence estimates in these regions. The study was conducted using anonymous, de-identified specimens. The study design was approved by the Health Research Ethics Board of the University of Manitoba.

From the Departments of Community Health Sciences (Mahmud, Becker, Elliott, Thompson, Fowke, Avery, Harlos, Blanchard) and Medical Microbiology (Becker, Keynan, Elliott, Fowke, Van Caesele, Dawood), University of Manitoba, Winnipeg, Man.; and the Cadham Provincial Laboratory (Van Caesele, Dawood), Winnipeg, Man.

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Assuming a seroprevalence of 20% following the first wave of the pandemic (H1N1) 2009, with  $\alpha$  set at 0.05, we calculated a sample size of 246 for the panel of sera specimens obtained after the first wave, to ensure a 95% confidence interval (CI) no wider than 10%.

For each of the two panels of serum specimens, we calculated the point seroprevalence of pandemic (H1N1) influenza as the proportion of specimens with an antibody titre of 1:40 or greater in the hemagglutination inhibition assay.<sup>9–11</sup> The cumulative incidence was then calculated as the difference between the point prevalence rates for the March and August panels. Because of over-sampling from the northern regions of the province, each individual observation was weighted by the inverse of its sampling fraction. For each cumulative incidence estimate, we calculated a 95% CI using bias-corrected boot-strapped standard errors.<sup>12</sup>

## Results

The two panels of serum specimens were generally representative of the sampling frame and the population of all pregnant women in the province in terms of their age composition and geographic distribution (data not shown). The median age was 28 (range 16–40) years for the women whose samples were collected in March and 27 (range 16–43) years for those whose samples were collected in August ( $p = 0.52$ ).

Overall, 7.1% of the serum specimens collected in March and 15.7% of those collected in August were positive for antibodies against the pandemic (H1N1) virus (Table 1). From these rates, we calculated the cumulative incidence to be 8.6% (95% CI 3.2%–13.7%). The cumulative incidence was higher among younger women (age 14–27 years; 17.2%, 95% CI 8.6%–25.4%) than among older women (3.6%, 95% CI 0.0%–10.6%). The use of other cut-off points for positivity (titres of 1:20 and 1:80) did not change the estimates appreciably.

In the geographic analysis, which was limited by small numbers, the

highest cumulative incidence at the cut-off titre of 1:40 was observed among the women in the northern regions (20.8%, 95% CI 7.9%–31.8%), as compared with 4.0% (95% CI

**Table 1:** Estimated cumulative incidence of pandemic (H1N1) influenza among pregnant women during the first wave of the 2009 pandemic, by age group

Cut-off point for positivity; age group, yr	No.*	Point prevalence, %		Cumulative incidence,† % (95% CI)
		In March 2009 <i>n</i> = 252	In August 2009 <i>n</i> = 296	
<b>HIA titre <math>\geq</math> 1:20</b>				
14–27	150	23.9	36.0	12.1 ( 1.1–23.3)
28–44	146	19.3	23.3	4.0 ( 0.0–13.1)
All	296	21.4	27.3	5.9 ( 0.0–12.6)
<b>HIA titre <math>\geq</math> 1:40</b>				
14–27	150	6.8	24.0	17.2 ( 8.6–25.4)
28–44	146	7.4	11.0	3.6 ( 0.0–10.6)
All	296	7.1	15.7	8.6 ( 3.2–13.7)
<b>HIA titre <math>\geq</math> 1:80</b>				
14–27	150	2.6	14.0	11.4 ( 5.5–18.0)
28–44	146	1.5	6.8	5.4 ( 1.2–10.1)
All	296	2.0	10.0	8.0 ( 4.4–12.2)

Note: CI = confidence interval, HIA = hemagglutination inhibition assay.

\*The number of specimens tested in August.

†Calculated as the difference between the point prevalence estimates in March and August 2009.

**Table 2:** Estimated cumulative incidence of pandemic (H1N1) influenza among pregnant women during the first wave of the 2009 pandemic, by region of residence

Cut-off point for positivity; region*	No.†	Point prevalence, %		Cumulative incidence,‡ % (95% CI)
		In March 2009 <i>n</i> = 252	In August 2009 <i>n</i> = 296	
<b>HIA titre <math>\geq</math> 1:20</b>				
Winnipeg	126	17.3	22.2	4.9 ( 0.0–14.7)
Northern Manitoba	90	25.6	43.3	17.8 ( 2.0–33.1)
Elsewhere	80	25.6	26.3	0.6 ( 0.0–14.9)
All	296	21.4	27.3	5.9 ( 0.0–12.6)
<b>HIA titre <math>\geq</math> 1:40</b>				
Winnipeg	126	7.1	11.1	4.0 ( 0.0–11.9)
Northern Manitoba	90	7.0	27.8	20.8 ( 7.9–31.8)
Elsewhere	80	7.3	16.3	8.9 ( 0.0–18.8)
All	296	7.1	15.7	8.6 ( 3.2–13.7)
<b>HIA titre <math>\geq</math> 1:80</b>				
Winnipeg	126	1.6	7.1	5.6 ( 1.0–11.5)
Northern Manitoba	90	0.0	13.3	13.3 ( 7.0–21.0)
Elsewhere	80	3.7	12.5	8.8 ( 1.0–17.0)
All	296	2.0	10.0	8.0 ( 4.4–12.2)

Note: CI = confidence interval, HIA = hemagglutination inhibition assay.

\*Northern Manitoba refers to four northern regional health authorities (North Eastman, NOR-MAN, Burntwood and Churchill).

†The number of specimens tested in August.

‡Calculated as the difference between the point prevalence estimates in March and August 2009.

0.0%–11.9%) in the Winnipeg region and 8.9% (95% CI 0.0%–18.8%) in the rest of the province (Table 2). At this cut-off titre, positivity levels for the specimens collected in March were similar across all regions (range 7.0%–7.3%). Despite the small numbers, similar patterns emerged when the data were stratified by both region and age group (data not shown).

## Interpretation

Our results are consistent with the rates of laboratory-confirmed pandemic (H1N1) influenza and of hospital admissions and severe illness observed in the first wave of the 2009 pandemic, all of which were much higher in northern Manitoba.<sup>13</sup> This geographic pattern was reversed during the second wave (October–December 2009), possibly because of higher levels of herd immunity after the first wave in the northern regions of the province.

The reasons for the higher cumulative incidence in the northern regions during the first wave are not clear. Aboriginal communities, with younger populations,<sup>14</sup> represent the majority of the population of northern Manitoba, which may explain some of the differences. Unfortunately, we could not reliably adjust our estimates for age-related differences because of the small numbers. It has been proposed that the uniformity of the patterns of pandemic (H1N1) influenza among Aboriginal communities across the globe, despite their genetic diversity, suggests a role for socio-economic and environmental factors, such as overcrowding, poor housing and limited access to health care services.<sup>15</sup>

## Limitations

Our estimates may not be generalizable to the rest of the young adult population in Manitoba, because pregnant women may be at increased risk of pandemic (H1N1) influenza, if one assumes that pregnancy is a marker of increased contact with children.<sup>16</sup> However, our results are comparable to those reported in a study from England that used a similar design to ours but was not restricted to pregnant women.<sup>17</sup>

The relatively small sample limited our ability to measure the cumulative incidence precisely, particularly in the geographic analysis. Also, pregnant women who did not undergo routine prenatal screening were not represented; however, the included women appeared to be representative of the population of all pregnant women. Women whose serum specimens were collected in August, after the first wave of the pandemic, were slightly younger than women whose specimens were in the March panel, which may have inflated our estimates.

The hemagglutination inhibition assay is not very specific.<sup>7</sup> However, we compared seroprevalence estimates before and after the first wave of the pandemic to minimize the resulting upward bias. Finally, the antibody titre corresponding to adequate protection against pandemic (H1N1) influenza is not known; we therefore used a range of cut-off points, all of which yielded consistent results.

## Conclusion

Our study showed that the cumulative incidence of pandemic (H1N1) influenza during the first wave of the 2009 pandemic was 8.6% among pregnant women in Manitoba. The rate was 20.8% in the northern regions of the province.

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**Competing interests:** None declared.

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**Correspondence to:** Dr. Salaheddin Mahmud, Department of Community Health Sciences, University of Manitoba, Rm. S113, Medical Services Building, 750 Bannatyne Ave., Winnipeg MB R3E 0W3; [salah.mahmud@gmail.com](mailto:salah.mahmud@gmail.com)