Epidemiological and clinical profile of Influenza A(H1N1) pdm09 in Odisha, eastern India

B. Dwibedi a,*, J. Sabat b, S. Dixit b, S. Rathore b, S. Subhadra b, S. Panda b, S.S. Pati b, M. Mandal b, L.M. Ho b, B. Thakur b, S.K. Kar c

a Dept. of Pediatrics, AIIMS, Bhubaneswar, Odisha, India
b Virus Research and Diagnostic Laboratory, Regional Medical Research Centre (ICMR), Bhubaneswar, Odisha, India
c SOA University, Bhubaneswar, Odisha, India

ARTICLE INFO

Keywords:
Epidemiology
Infectious disease
Odisha
Real time RT-PCR
H1N1
India

ABSTRACT

Epidemic of flu is highly contagious and it spreads through air. In 2009 H1N1 influenza virus emerged after reassortment of North American TRIG and Eurasia Avian like virus of swine and started epidemic in Mexico. The first cases were reported from Hyderabad city on 16th May 2009 in India that spread rapidly within a short span of time. During this period large population of Odisha situated at the eastern side of India was also affected and incidences of H1N1 cases were recorded through state Government surveillance system. In this study real time RT-PCR based diagnosis was conducted for the throat swabs collected from suspected H1N1 cases in Odisha during 2009–2017. A total of 2872 throat swabs were received from 23 different Government and private hospitals and 21.1% positivity was confirmed. The disease affected mostly 46–60 years age group, males (50.6%) being more affected. The clinical features had shown that fever with cough (89.6%) was the most common symptom followed by shortness of breath (72.7%). Post monsoon was the peak season in which most of the cases were reported. Neurological signs, pregnancy, diabetes and hypertension were found to be risk factors for H1N1. The case fatality rate (CFR) was 15%.

1. Introduction

Flu epidemics occur every 6–10 years due to antigenic shift and that exposes human population with a new strain of influenza leading to higher or lower morbidity or mortality. The first human influenza epidemic of 21st century was an Influenza A(H1N1)pdm09 subtype virus that emerged through reassortment of North American Triple reassortment (TRIG) and Eurasian Avian (EA) like viruses of swine [1]. In 2009 world encountered a pandemic due to a novel Influenza A(H1N1)pdm09. First case was reported from Mexico in 2009 which was characterized by rapid spread and caused high morbidity [2] that subsequently spread to more than 214 countries with more than 18,366 deaths [3]. The first case of pandemic Influenza A(H1N1)pdm09 in India was reported from Hyderabad city on 16th May 2009 [2]. After that the virus soon spread to almost all major cities in India.

The pandemic A(H1N1)pdm09 started in the eastern part of Odisha in September, 2009 and spread rapidly to all parts of the state and lasted until the end of year 2010. A large number of Influenza A(H1N1)pdm09 cases (n = 118) and deaths (n = 32) were reported during this pandemic. In next two-three years only sporadic influenza activity was reported i.e. 2 confirmed cases of Influenza A(H1N1)pdm09 were reported in 2012, while only one case confirmed in 2013. Virulence of Influenza A(H1N1) pdm09 virus became once again active and caused another epidemic in 2015. The number of new cases, including fatal cases continued to increase since September, 2015. In this period, we reported 60 new Influenza A(H1N1)pdm09 cases and 5 deaths. We carried out a retrospective analysis of the available information, in order to study the clinico-epidemiological features and establish the magnitude and severity of recent Influenza A(H1N1)pdm09 epidemics in hospitalized patients from the state of Odisha.

2. Materials and methods

This retrospective record based study was conducted among patients...
of all age groups who were hospitalized and suspected of Influenza A(H1N1)pdm09 virus infection. The study was carried out at Virology Laboratory specially designated for influenza testing at Regional Medical Research Centre (ICMR), Bhubaneswar, Odisha, India from 2009 to 2017.

A suspected case of Influenza A(H1N1)pdm09 virus infection was defined as per WHO classification of category A and B patients or a patient with influenza-like illness (ILI) with a history of international travel from the country of confirmed Influenza A(H1N1)pdm09, or a close contact with a confirmed H1N1 infected person. ILI was defined as a case presenting with fever ≥38°C with at least one respiratory symptom such as cough, rhinorrhea, or sore throat. Confirmed case defined as a probable case that was tested positive for pandemic Influenza A(H1N1) pdm09 by real-time reverse transcription polymerase chain reaction (rRT-PCR) [4].

All patients with ILI belonging to Category B and C were screened at different Government and private hospitals and medical colleges as per the state health programme and samples referred to viral laboratory of RMRC, Bhubaneswar for laboratory confirmation. The clinico-epidemiological information about all referred cases from these hospitals of the state was collected along with the respiratory sample using proforma prepared by the Ministry of Health and Family Welfare, Government of India. An additional proforma designed by the lab was also used to gather relevant information from all suspects and deaths from Influenza A(H1N1)pdm09. The four medical colleges namely SCB Medical College and Hospital at Cuttack, VIMSAR Medical College at Samalpur, MKCG Medical College, Berhampur, AIIMS, Bhubaneswar and state capital hospital at Bhubaneswar acted as five nodal centres for screening of Swine flu cases in the state. Besides private or public sector hospitals and medical colleges providing critical care to patients were also part of the Pandemic Flu management chain. After screening samples were referred to virology laboratory, Regional Medical Research Centre, ICMR, Bhubaneswar maintaining cold chain. Laboratory investigations were done with adequate quality control (QC) measures monitored by the National Institute of Virology (NIV), Pune. The QC measures included periodic training of the laboratory staff, standardizing reagents for RealTime PCR and EQAS proficiency testing.

Upper respiratory tract specimens including nasal and throat swabs were collected using plastic shafted dacron swabs placed into viral transport medium (VTM). RNA was extracted using the QIAamp viral RNA mini kit (QIAGEN, Germany) according to the manufacturer instruction. Amplification and detection for each RNA isolate was performed on Applied Biosystem’s RealTime PCR 7500 instrument by using primers and probe sets for Influenza A, Universal Swine (swA), Swine H1 (swH1) and RNaseP (Applied Biosystems, USA) as per the CDC real-time RT-PCR protocol (RealTime PCR, Applied Biosystems, USA) [5].

The epidemiological parameters were analysed which included demographic, clinical profile and disease outcome. Case Fatality Rate (CFR) was calculated by calculating numbers of deaths out of positive cases. All the relevant data collected and analyzed using Microsoft Excel.

3. Result

Odisha experienced the first pandemic during last quarter of 2009 when 16 districts (58 suspected cases) reported the outbreak. The outbreak was more widespread during 2017 in terms of more number of affected districts (n = 30) and suspected cases (n = 1703). A total of 2872 acute phase nasal/throat/nasopharyngeal swabs, tracheal aspirates from patients hospitalised in 23 different Government and private hospitals in the state of Odisha were referred to the virology laboratory during 2009–2017. Year wise distribution of cases is detailed in Table 1. Of these, 955 (32.2%) samples were positive for Influenza A and out of these cases, 606 (21.1%) samples were positive for Influenza A(H1N1)pdm09 through RealTime PCR. Remaining 349 cases were only Flu A positive. No sample was found positive for Influenza B. During this period 91 deaths occurred with CFR of 1.5% among the Influenza A(H1N1)pdm09 positive cases. The year wise distribution of Influenza A(H1N1)pdm09 cases (Table 1) reveals highest numbers of cases (n = 1703) received during 2017 with 67.33% H1N1 positivity followed by 63.94% during 2010. It was observed that CFR was high with 30.3% in 2010 followed by 15.79% in 2009. During 2015 though the number of confirmed Influenza A(H1N1)pdm09 cases was 12.5% but the CFR was low (6.58%).

Analysis of age and sex distribution of pandemic Influenza A(H1N1) pdm09 virus suspected and positive cases shown in Table 2. Out of the 606 positive cases; 307 (50.6%) were males representing 10.6% of suspected cases enrolled for laboratory testing. It was observed that the number of positive cases increased with age up to 60 years. Pandemic Influenza A(H1N1)pdm09 positivity rate was higher (29.70%) in the age group of 46–60 years followed by 31–45 years age group i.e. 22.77%.

The clinical features of the confirmed Influenza A(H1N1)pdm09 infected cases had shown that fever and cough were the most common (89.6%) symptom followed by shortness of breath (72.7%) where as sore throat and nasal catarrh were less common symptoms. As per chest radiograph, 47.5% positive cases had signs of pneumonia. It was observed that 18.15% of patients among positives were staying in the community where one or more infected people were Influenza A(H1N1)pdm09 positive (Table 3).

Similarly, among the cases of pandemic Influenza A(H1N1)pdm09 negative but influenza A positives; fever (91.4%) and cough (87.67%) were most common symptoms followed by shortness of breath (77.6%). No follow up data was available for deaths among pandemic Influenza A(H1N1)pdm09 negative cases. It was also noted that there was no difference among pandemic Influenza A(H1N1)pdm09 and Influenza A positives as far as affection of age group is concerned.

During this period 91 deaths were reported among hospitalized Influenza A(H1N1)pdm09 lab confirmed patients (n = 606) (Table 4) belonged to the age group of 46–60 years (Fig 1). Number of deaths was more among male patients (n = 50, 54.9 %) than females (n = 41, 45.1%). Out of 91, 52.74% (48) had other underlying conditions like CNS involvement (15.4%) and ARDS (13.2%) etc. Other co-morbid conditions like hypertension, diabetes, coronary artery disease and pregnancy were also noted in low frequency (Table 4).

It was observed that among the deaths there was a mean gap of 5.7 days when the patients were admitted to the hospital after onset of the symptoms. Also a mean gap of 6.4 days between onset of symptom and intake of Tamiflu was noted among the hospitalized patients. Tamiflu was given on same day of admission as per requirement of C-category in 7 cases and 2nd day in 3 cases and beyond 2nd day in 3 cases when respiratory signs progressed. Only 4 cases reported to local doctor’s consultation before attending referral hospitals.

Year wise analysis of influenza activity revealed no case of Influenza A(H1N1)pdm09 in 2011 and 2014, whereas only one case was found positive during the years 2013 and 2016. Month wise break up showed highest number of positive cases admitted in the months of August during the respective years with larger case load i.e. in 2010, 2015 and 2017.
March in 2015. Apart from this a shorter peak was observed during the month of August. First peak started in February that in our region two peaks of epidemic of Influenza associated with Influenza A(H1N1)pdm09 infection was reported in some studies indicating different behaviour, hormonal response and susceptibility to infectious diseases may be the reason for this difference among males and females [8].

The characteristic feature of this pandemic influenza infection was that, case distribution was disproportionate among all age groups but the patients belonging to 46–60 years followed by 30–45 years of age group found to be more affected than the other age groups, which was also reflected in other studies [6]. This finding also corroborates with the study of Jin Lv et al., 2017 [7] reporting higher prevalence in elder age group which was not exposed previously. In our study it was observed that infection rate was higher among male individuals (50.6%) than female (49.3%) though the difference is not significant. Similar observation was reported in some studies indicating different behaviour, hormone response and susceptibility to infectious diseases may be the reason for this difference among males and females [8].

Among the confirmed positive cases for Influenza A(H1N1)pdm09 common symptoms observed were fever with cough (89.6%) followed by shortness of breath (72.7%). Similar symptoms of positive cases were also observed in various other studies [9, 10, 11]. This can be used for preliminary diagnosis of Influenza A infection during the influenza season prior to lab diagnosis report.

Month-wise analysis of influenza activity in the present study showed that in our region two peaks of epidemic of Influenza A(H1N1)pdm09 were observed. First peak started in winter season during February–March and second peak was observed in post monsoon season i.e. August–September where second was dominant. Increased influenza activity in monsoon and post monsoon seasons also observed in other studies [12, 13, 14]. First peak started in February–March i.e., in cold and

(Fig. 2). Apart from this a shorter peak was observed during the month of March in 2015.

### Table 2
Proportion of suspected and confirmed cases of influenza by age group and gender.

| Age group (yrs) | Male | | Female | | Total | |
|----------------|------| |--------| |--------| |
|                 | Suspected cases | Positive | % Positive | Suspected cases | Positive | % Positive | Suspected cases | Positive | % Positive |
| ≤5             | 88   | 20 | 6.51 | 58   | 11 | 3.67 | 146 | 31 | 5.11 |
| 5 to 15         | 70   | 21 | 6.84 | 77   | 22 | 7.35 | 147 | 43 | 7.09 |
| 16 to 30        | 304  | 49 | 15.96 | 277  | 62 | 20.73 | 581 | 111 | 18.31 |
| 31 to 45        | 323  | 64 | 20.84 | 269  | 74 | 24.74 | 592 | 138 | 22.77 |
| 46 to 60        | 426  | 89 | 28.9  | 334  | 91 | 30.43 | 760 | 180 | 29.70 |
| >60             | 437  | 64 | 20.84 | 209  | 39 | 13.04 | 646 | 103 | 16.99 |
| Total           | 1648 | 207 | 12.48 | 1224 | 299 | 19.32 | 2872 | 606 | 21.02 |

* % positive was calculated for each age group among total positive cases for all age groups.

### Table 3
Comparison of clinical features and travel history of suspected and confirmed cases of H1N1.

| Clinical condition | Suspected ILI cases (n = 2872) | Positive for H1N1 (n = 606) | Negative for H1N1 (n = 2266) |
|-------------------|--------------------------------|-----------------------------|-----------------------------|
|                   | No. of cases | % | No. of cases | % | No. of cases | % |
| Fever             | 2481         | 86.39 | 543 | 89.60 | 1938 | 85.53 |
| Cough             | 2417         | 84.16 | 543 | 89.60 | 1874 | 82.70 |
| Sore throat       | 1512         | 52.65 | 343 | 56.60 | 1169 | 51.59 |
| Nasal catarrh     | 895          | 31.16 | 197 | 32.51 | 698  | 30.80 |
| Shortness of breath | 2284      | 79.53 | 441 | 72.77 | 1843 | 81.33 |
| Pneumonia signs   | 1555         | 54.14 | 288 | 47.52 | 1267 | 55.91 |
| Travel details    |               | | | | | |
| Visit to other locality/state/country | 114 | 3.97 | 27 | 4.46 | 87 | 3.84 |
| History of close contact with confirmed case of H1N1 positive (within 7 days) | 172 | 5.99 | 37 | 6.11 | 135 | 5.96 |
| Travel to a community reported of H1N1 cases | 237 | 8.25 | 53 | 8.75 | 184 | 8.12 |
| Resides in the community where there are one or more confirmed influenza cases | 444 | 15.4 | 110 | 18.15 | 334 | 14.74 |

### Table 4
Conditions associated with Influenza A(H1N1)pdm09 confirmed deaths (n = 91).

| Parameters                  | Number (%) |
|-----------------------------|------------|
| Age group (Years)           |            |
| <15                         | 8 (8.8)    |
| 16-30                       | 15 (16.5)  |
| 31-45                       | 19 (20.9)  |
| 46-60                       | 35 (38.6)  |
| >60                         | 14 (15.4)  |
| Hypertension                | 4 (4.4%)   |
| Diabetes                    | 6 (6.6%)   |
| Coronary artery disease     | 4 (4.4%)   |
| Chronic renal failure       | 4 (4.4%)   |
| Pregnancy                   | 4 (4.4%)   |
| Neurological complication   | 14 (15.4)  |
| Acute Respiratory Distress Syndrome | 12 (13.2) |
| Mean gap between onset of illness & hospitalization | 5.7 days (Median 5.5) |
| Mean gap between onset of illness & Tami flu intake | 6.4 days (Median 6) |
| Mean gap between hospital admission & Tami flu intake | 1.2 days |
| Survival after hospitalization (Mean period) | 3.4 days (Median 2.5) |

(Fig. 1). Numbers of Influenza A 2009 associated deaths by year and age distribution.

4. Discussion

India reported its first case of pandemic Influenza A(H1N1)pdm09 in May 2009 from Hyderabad city, Telangana [2] and Odisha reported first confirmed case of Influenza A(H1N1)pdm09 in October 2009. Through 2009 to 2017, a total of 2872 suspected patients with flu like symptoms were hospitalized and tested for pandemic influenza Influenza A(H1N1) pdm09 infection, of which 606 (21.1%) were found to be positive.
low solar radiation season with a decreasing trend in number of cases during April–June; dry and sunny season which indicated increased influenza activity in cold temperature and in low solar radiation [15, 16, 17]. Exact reason for increased influenza activity in winter is not known but when temperature drops people spend more time indoors, making it easier for the virus to spread in-house (8). No/low number of cases were reported in 2011, 2013–14 and 2016 which may indicate that most of the people were vaccinated during 2009–10 out of Influenza A(H1N1)pdm09 positives for which decline in transmission occurred over 2013–14. On lowering of the host immunity over few years, the population was again susceptible to acquire infection and thus, epidemic resurgence again in 2015. Presumably the situation was repeated in 2016 and again due to lack of immunity majority of the population affected in 2017. This could be due to absence of a programme for flu vaccination in the country, where pandemic panic was the driving force for the community to take a preventive vaccine. This force became weak over time possibly because of the increase in the health system preparedness in handling emergen-
cles that adults as well as young aged were mostly affected by the wave of this pandemic Influenza A(H1N1)pdm09 where as in other studies age group of 20–49 years was suggested to be more affected [30]. In our investigation age group 20–50 year was also found to have significantly high CFR.

Among the Influenza A(H1N1)pdm09 deaths, average time lag between onset of illness and hospital admission was 5.7 days (Range 0–15 days) and the mean duration of hospital stay of cases leading to death was 3.4 days (Range 0–13 days). There was an average delay of 6.4 days for Tamiflu use. Hence it can be presumed that delay in antiviral therapy can be a main determinant for mortality.

Though our study gives an impression on associated risk factors for mortality, it is limited by certain amount of missing clinical data like pre-existing respiratory disease, small numbers, incomplete records on epidemic dynamics within the population and health structure/referral chain.

According to the data from WHO till March 2010, this new Influenza A(H1N1)pdm09 was estimated to have a case-fatality rate (CFR) of 1.28%. The CFR in our study was much higher (15%) in comparison to other studies [30] which may be due to under reporting of cases of H1N1 especially those of mild illness. The CFR during the wave was high in the fall season.

5. Conclusion

The study results are worth noting on its strengths and limitations. We used detailed hospitalization data from an enhanced surveillance system that covered all hospitals in the state, and individual-level clinical data also allowed us to assess the effect of age, gender and underlying co-morbidities on the risk of death among Influenza A(H1N1)pdm09 patients. It reflects that adults as well as young aged were mostly affected by the wave of this pandemic Influenza A(H1N1)pdm09 suggesting risk of acquisition of infection by most active and productive age group which should be considered in any future preventive plan. Although children are the principal targets in any immunization programme, the adults also need emphasis in such a situation where they are equally susceptible and risk is high due to movement and exposure. The co-morbid conditions associated with mortality should be taken into account and health providers can recommend influenza vaccination on priority to patients with discussed medical conditions. These high-risk patients also need careful attention when become hospitalized with flu like illness. Besides, early antiviral therapy can be targeted during such pandemics in countries like India, where laboratory investigation for all suspected patients may not be feasible.

The ethical clearance for the study was not required since samples were referred to us for diagnosis as a public health concern to mitigate the pandemic. However the investigations were performed following GCP guideline of ICMR as well as Institutional Ethics Committee recommendations for the ICMR Virology grade-1 laboratory project.
Declarations

Author contribution statement

B. Dwivedi et al. Heliyon 5 (2019) e02639

Funding statement

Acknowledgements

Additional information

References

Author wishes to thank DSIP, Govt of Odisha for providing assistance during outbreak investigation. Authors also thank DHR, ICMR, New Delhi for financial support.

J. Lv, Z.Y. Ren, Y.Y. Zhang, et al., Study on age-dependent pre-existing 2009 pandemic influenza virus T and B cell responses from Chinese population, BMC Infect. Dis. 17 (1) (2017) 136. Published 2017 Feb 10.

W. Choi, B.H. Rho, M.Y. Lee, Male predominance of pneumonia and hospitalization in pandemic influenza A (H1N1) 2009 infection, BMC Res. Notes 4 (2011) 351. http://www.biomedcentral.com/1756-0500/4/351.

S. Revudwala, S. Mulia, T. Panwala, et al., Clinical characterisation of H1N1 influenza Taqman real time PCR positive cases, Natl. J. Med. Res. 2 (1) (2012) 12-14.

M. Khalid, J. Tahir, T. Muhammad, et al., Swine flu experience in local population of Lahore, Biomedia 26 (19) (2010) 50-53.

A. Choudhry, S. Singh, S. Khare, et al., Emergence of pandemic 2009 influenza A H1N1, India, Indian J. Med. Res. 135 (4) (2012) 534-537.

S.A. Rasmussen, D.J. Jamieson, K. Macfarlane, et al., Pandemic influenza and pregnant women: summary of a meeting of experts, Am. J. Public Health (2009 Jun 18) [Epub ahead of print].

S.R. Balaganasekumkar, M.V. Murbekar, K.K. Swamy, et al., Risk factors associated with death among influenza A (H1N1) patients, Tamil Nadu, India, 2010, J. Postgrad. Med. 59 (1) (2013) Jan-Mar 9-14.

A. Campbell, R. Rodin, R. Kropp, et al., Risk of severe outcomes among patients admitted to hospital with pandemic (H1N1) influenza, CMAJ 162 (4) (2010) 249–255.

S. Jain, L. Kamimoto, A.M. Bramley, et al., Hospitalized patients with 2009 H1N1 influenza in the United States, april–june 2009, N. Engl. J. Med. 361 (2009) 1935-1944.

R.T. Yokota, L.M. Skalinski, C.N. Igani, et al., Risk factors for death from pandemic (H1N1) 2009, southern Brazil, Emerg. Infect. Dis. 17 (8) (2011) 1467-1471.

R. Chudasama, U. Patel, P. Verma, et al., A two wave analysis of hospitalizations and mortality from seasonal and pandemic 2009 (H1N1) influenza in Saurashtra, India: 2009-2011, Ann. Med. Health Sci. Res. 3 (3) (2013) 334-340.

B.A. Cunha, U. Syed, N. Mickail, S. Strollo, Rapid clinical diagnosis in fatal swine influenza (H1N1) pneumonia in an adult with negative rapid influenza diagnostic tests (RDTs): diagnostic swine influenza trial, Heart Lung 39 (1) (2010 Jan-Feb) 78-86.

J.K. Louie, M. Acosta, K. Wintner, et al., Factors associated with death or hospitalization due to pandemic 2009 influenza A(H1N1) infection in California, J. Am. Med. Assoc. 302 (17) (2009) 1896-1902.

S. Echevarria-Zuno, J.M. Mejia-Arangure, A.J. Mar-Obeso, et al., Infection and death from influenza A H1N1 virus in Mexico: a retrospective analysis, Lancet 374 (9707) (2009) 2072–2079, 19; Epub 2009 Nov 11.

J.K. Louie, M. Acosta, D.J. Jamieson, et al., Severe, 2009 H1N1 influenza in pregnant and postpartum women in California, N. Engl. J. Med. 362 (1) (2010 Jan 7) 27–35.

The ANZIC Influenza Investigators and Australasian Maternity Outcomes Surveillance System: critical illness due to 2009 A/H1N1 influenza in pregnant and postpartum women: population based cohort study, BMJ (2010) 340.423.

R. Zarychanski, T.L. Stuart, A. Kumar, et al., Correlates of severe disease in patients with 2009 pandemic influenza (H1N1) virus infection, CMAJ 182 (3) (2010) 257-264.

G. Palacios, M. Hornig, D. Cisterna, et al., Streptococcus pneumoniae coinfection is correlated with the severity of H1N1 pandemic influenza, PLoS One 4 (12) (2009), e8540. Published 2009 Dec 31.

World Health Organization, New influenza A (H1N1) virus: global epidemiological situation, June 2009, Wkly. Epidemiol. Rec. 84 (25) (2009) 249–257.

L. Vaillant, G. La Ruche, A. Tarantola, et al., Epidemic Intelligence Team at In VS, World Health Organization, New influenza A/H1N1 virus in the United States following the 2009 pandemic, PLoS One 7 (10) (2012), e48187.

K. Tan, A. Prema, Y. Leo, Surveillance of Influenza A(H1N1)pdm09-related neurological complications, Lancet Neurol. 9 (2) (2010) 142-143.

C. Reed, J.M. Katz, K. Hancock, A. Balish, A.M. Fry, H1N1 Serosurvey Working Group, Prevalence of seropositivity to pandemic influenza A/H1N1 virus in the United States following the 2009 pandemic, PLoS One 7 (10) (2012), e48187.

H. Rana, P. Parikh, A.N. Shah, S. Gandhi, Epidemiology and clinical outcome of H1N1 in Gujarat from July 2009 to March 2010, J. Assoc. Phys. India, 60 (2012) 95-97.