IDENTIFICATION OF VIRAL PATHOGENS RESPONSIBLE FOR SEVERE AND COMPLICATED RESPIRATORY INFECTION DURING THE PANDEMIC AND POST-PANDEMIC PERIOD (2009-2011)

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BACKGROUND

Most cases of suspected influenza A(H1N1)pdm09 infection presented with a mild outcome, but some presented as severe acute respiratory infection (SARI) and required admission to intensive care unit (ICU), mainly due to acute respiratory distress syndrome (ARDS), a pulmonary inflammatory syndrome characterized by diffuse alveolar damage that causes edema. Although influenza A(H1N1)pdm09 strain was the prevalent circulating virus during the pandemic and post-pandemic period (2009-2011), many other viruses are able to cause SARI. Up to now, more than 200 antigenically diverse viral serotypes, belonging to the main viral families have been associated with occurrence of acute respiratory infections.

OBJECTIVE

To determine viral pathogens responsible for SARI/ARDS during the pandemic and post-pandemic (2009-2011) in Lombardy, Italy.

MATERIALS AND METHODS

In the capacity of reference laboratory operating within the InfluNet network (http://www.iss.it/iflu/) in charge of carrying out the virological surveillance of severe forms of influenza infection in Lombardy (northern Italy, nearly 10 million inhabitants), 206 SARI/ARDS cases were identified during the pandemic and post-pandemic period (from October 1, 2009, to April 30, 2011). Of these, 61.2% were males with a median age of 44.3 years (IQR: 49.7 years; range: 1 month - 89 years). A respiratory sample was collected and then analyzed by real-time RT-PCR (CDC-2009 protocol) to identify influenza viruses. A panel for respiratory pathogens (Respiratory MWS r-gene™ Real-time PCR, bioMérieux) was used to detect RSV A/B, hMPV A/B, hRV/hEV, hAdV, hBoV 1-4, hCoV 229E, NL63, OC43, HKU1, hPIV 1-4. Besides investigating the common respiratory viruses, the VIDISCA-454 (VIrus DIScovery using CDNA Amplified fragment-length polymorphism combined with Roche 454 high-throughput sequencing) methodology was applied (Fig .1, Poster REF 407).

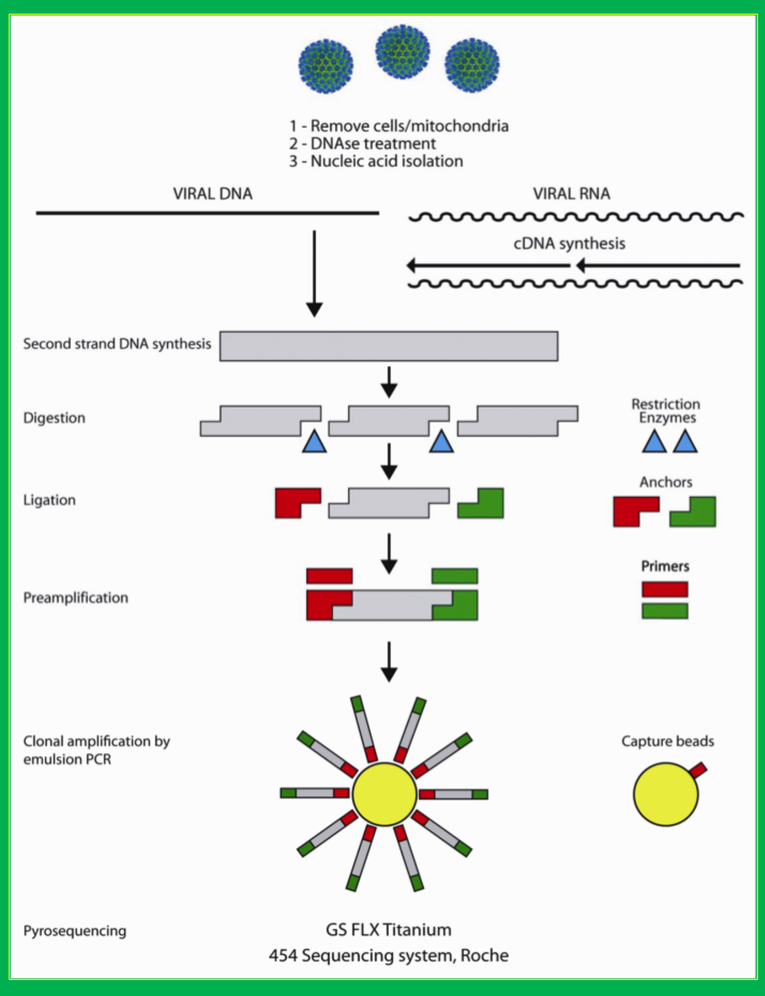

Fig.1: VIDISCA-454 methodology

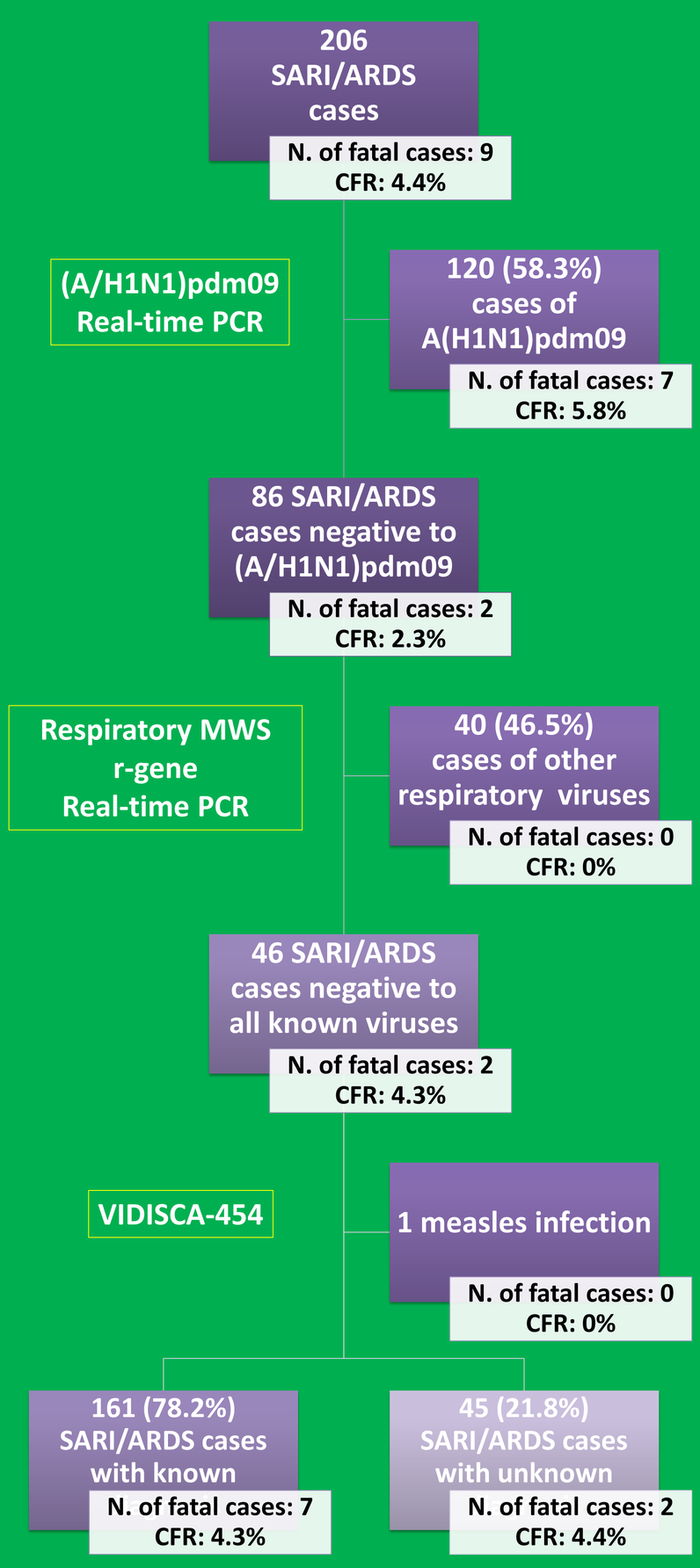

Fig. 2: Study design and results overview (CFR: case fatality risk)

RESULTS

58.3% (120/206) samples resulted positive for A(H1N1)pdm09 virus infection (61.7% males; median age: 42.9 years, IQR: 40.9 years).

40/86 (46.5%) SARI/ARDS samples that resulted negative to A(H1N1)pdm09 virus received a diagnosis of viral infection. These were 64.5% males with a median age of 44.2 years (IQR: 57.1 years). These infections were mainly sustained by hRV/hEV (11/40: 27.5%) and influenza A(H3N2) virus (8/40: 20%) (Table 1).

The remaining 46 negative samples (58.2% males; median age: 49.6 years, IQR: 61.3 years) were further investigated by VIDISCA-454 methodology which allowed the identification of 1 measles positive sample.

The other samples (21.8%) remained negative to any virus.

PATHOGEN	N. of positive samples	% of positive samples
hRV/hEV	11	27.5
Influenza A(H3N2) virus	8	20
RSV	4	10
hCoV	4	10
hPIV	4	10
AdV	2	5
Influenza B virus	1	2.5
hBoV	1	2.5
hMPV	0	0
C. pneumoniae	0	0
M. pneumoniae	0	0
Co-infections	5	12.5

Table 1: Impact of respiratory pathogens other than influenza A(H1N1)pdm09 virus

CONCLUSIONS

During the pandemic a post-pandemic period influenza A(H1N1)pdm09 virus had the greatest impact (58.3%) on severe respiratory infections. Nearly 22% of SARI/ARDS cases did not receive a certain diagnosis and two of these (4.4%) were fatal: thus, in view of the severity of the SARI/ARDS disease, it is important to investigate the specimens that are negative to all standard assays, and to apply innovative techniques able to identify unexpected pathogens, novel variants or emerging agents.

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