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Background

To control influenza successfully, the different ways that could promote influenza virus transmission in humans need to be known. Droplets and aerosols are considered as the main route of transmission. Nevertheless, human influenza A viruses can survive for a prolonged period of time in the environment as fomites. In addition, avian influenza viruses are excreted at high concentrations in stools, thus increasing the dissemination potential of this virus.

The aim of our study was to assess the survival of human influenza viruses on banknotes knowing that billions of them are exchanged daily.

Methodology

Viral stocks

Influenza A/Moscow/10/99 (H3N2), influenza A/Wisconsin/67/2005 (H3N2), influenza A/New Caledonia/20/99 (H1N1) and influenza B/Hong Kong/335/2001 were used at different concentrations.

Detection of virus survival

50 µl of a viral suspension were deposited on a small pieces of banknotes that were then conserved at room temperature (Figure 1a, 1b). During the experiments, temperature remained at 22°C and relative humidity was maintained between 30% and 50%. At predefined period of time, standardized pieces of banknotes were eluted in culture medium (Figure 1c, 1d) for 10min. 0.4 ml of the eluate was then used for cell inoculation. Cells were incubated at 33°C for 10 days and harvested for screening by immunofluorescence. 50 Francs Swiss Banknotes were provided by the Swiss National bank.

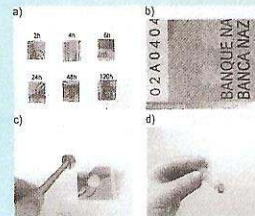


Figure 1 : Infectiousness determination

Results

1) Survival of influenza viruses on banknotes

With a low concentration, the duration of infectiousness for influenza A (H1N1) and influenza B was limited to 1h to 2h respectively (Figure 2). Survival of influenza A (H3N2) viruses, like influenza A/Wisconsin/67/05 and A/Moscow/10/99, tested at higher concentration, was significantly longer with a duration of up to 24 and 72h respectively (Figure 2).

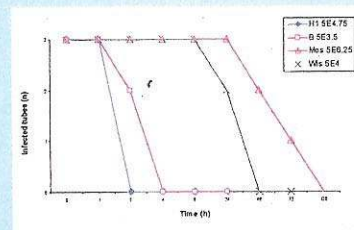


Figure 2 : Survival of different influenza viruses on banknotes

2) Impact of the initial concentration and the presence of mucus on survival of influenza viruses

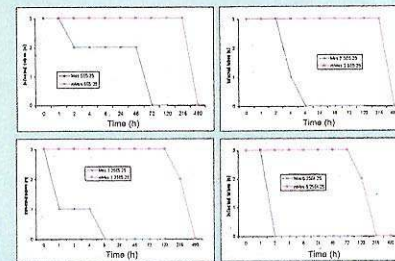


Figure 3 : Impact of concentration and mucus on survival of influenza viruses on banknotes

Different concentrations of influenza A/Moscow/10/99 (H3N2) and influenza B/Hong Kong/335/2001 were tested up to 120 hours in the presence and absence of respiratory mucus. The recovery rate was directly related to the size of the inoculum and we observed a rapid loss of the infectivity when the inoculum was low (Figure 3). At any concentration tested we observed a significant increase of influenza survival in the presence of mucus. This was the case for any type tested. For an influenza A/Moscow/10/99 (H3N2) suspension at $6,25 \cdot 10^{4,25} \text{TCID}_{50}/\text{ml}$, the virus could be recovered after 120 hours (5 days) after inoculation, whereas the virus survived less than two hours in the absence of mucus. Survival of A/Moscow/10/99 (H3N2) was prolonged up to 408h (17 days) in the presence of mucus. B/Hong Kong/335/2001 virus was still infectious after 24h on banknotes in the presence of mucus whereas in absence the virus did not survive more than 2h (data not shown). Detection of the RNA by real-time RT-PCR showed also a stability for more than 10 days (data not shown).

3) Survival of influenza viruses on banknotes

To assess whether these in vitro experiments could be reproduced under "natural conditions", we used nasopharyngeal secretions from infected individuals. Nasopharyngeal secretions of fourteen influenza-positive cases (detected by near patient test and cell culture) collected during the 2007 season were inoculated on banknotes. In 7/14 cases, influenza virus survived for at least 24h and in 5/14 cases influenza survival was $\geq 48\text{h}$ (Figure 4).

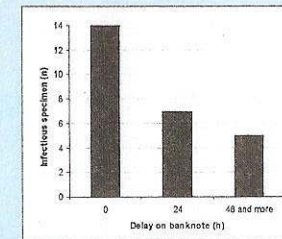


Figure 4: Survival of influenza virus in human nasopharyngeal secretions

Conclusions

- Influenza A viruses in respiratory secretions can survive up to 17 days on banknotes.
- Time of survival of infectious virus is related to the inoculum size and the presence of mucus.
- Contamination of environmental surfaces such as banknotes need to be considered as a potential vector for influenza in case of a pandemic.