



# Comparative analysis of human and chicken influenza virus A(H7N9)-induced inflammatory response and virus replication kinetics

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## Introduction

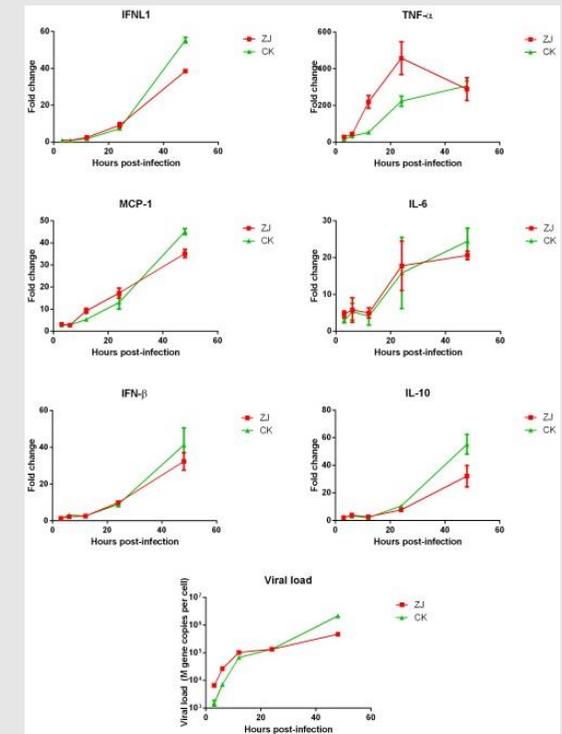
Avian influenza virus subtype A(H7N9) has caused over 430 human infections with over 30% mortality. Comparison of inflammatory response and viral replication between human and avian isolates of A(H7N9) virus is lacking. Here, we inoculated influenza viruses onto a human lung adenocarcinoma cell line Calu-3 and measured the cytokine/chemokine response and the kinetics of the viral replication.

## Materials and Methods

A/Zhejiang/DTID-ZJU01/2013 (ZJ) was isolated from a patient with A(H7N9) virus infection, and A/chicken/Zhejiang/1/2013 (CK) was isolated from a chicken in an epidemiologically-linked poultry market. The viruses were inoculated onto Calu-3 cells at three multiplicity of infection. After 1 h of viral adsorption, the medium was removed and cells were washed twice with medium before further incubation for 3, 6, 12, 24 and 48 h. The viral loads in the supernatant were determined by quantitative reverse-transcriptase polymerase chain reaction. Cell lysates were collected for cytokine mRNA expression assays.

## Results

At 12 hours post-infection, the cytokine/chemokine levels were similar between the H7N9 viruses, except that ZJ induced a greater increase in the mRNA level of TNF- $\alpha$  than CK ( $P = 0.010$ ) (Figure 1). There was <10 fold increase in the mRNA levels of the antiviral cytokines IFNL1 and IFN- $\beta$  for both viruses. At 48 hours post-infection, CK induced significantly higher levels of IFNL1 ( $P < 0.001$ ) and MCP-1 ( $P < 0.014$ ) than the human H7N9 virus ZJ, but there was no significant differences in the levels of IFN- $\beta$ , TNF- $\alpha$ , IL-6, IFN- $\beta$  and IL-10. The viral load of CK were about 1 log lower than that of ZJ at 3 and 6 hours post-infection. The viral load at 48 hours post-infection for CK were about 1 log higher than that for ZJ ( $P < 0.001$ ).



## Conclusions

The human A(H7N9) virus induced a higher level of pro-inflammatory cytokine TNF- $\alpha$  and was able to reach a higher titer in viral culture than the chicken A(H7N9) virus. These attributes may enhance the ability of the virus to infect humans.

## Contact

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