

XB Experiment design

Reagents: XB final concentration 0.5 oz/gallon, or 2 oz/gallon solution; 1x Phosphate Buffered Saline (PBS)

Cell culture plates: 6-well plates; 4 wells for per virus

Equine influenza virus: 10^4 TCID₅₀ (PFU)/ml

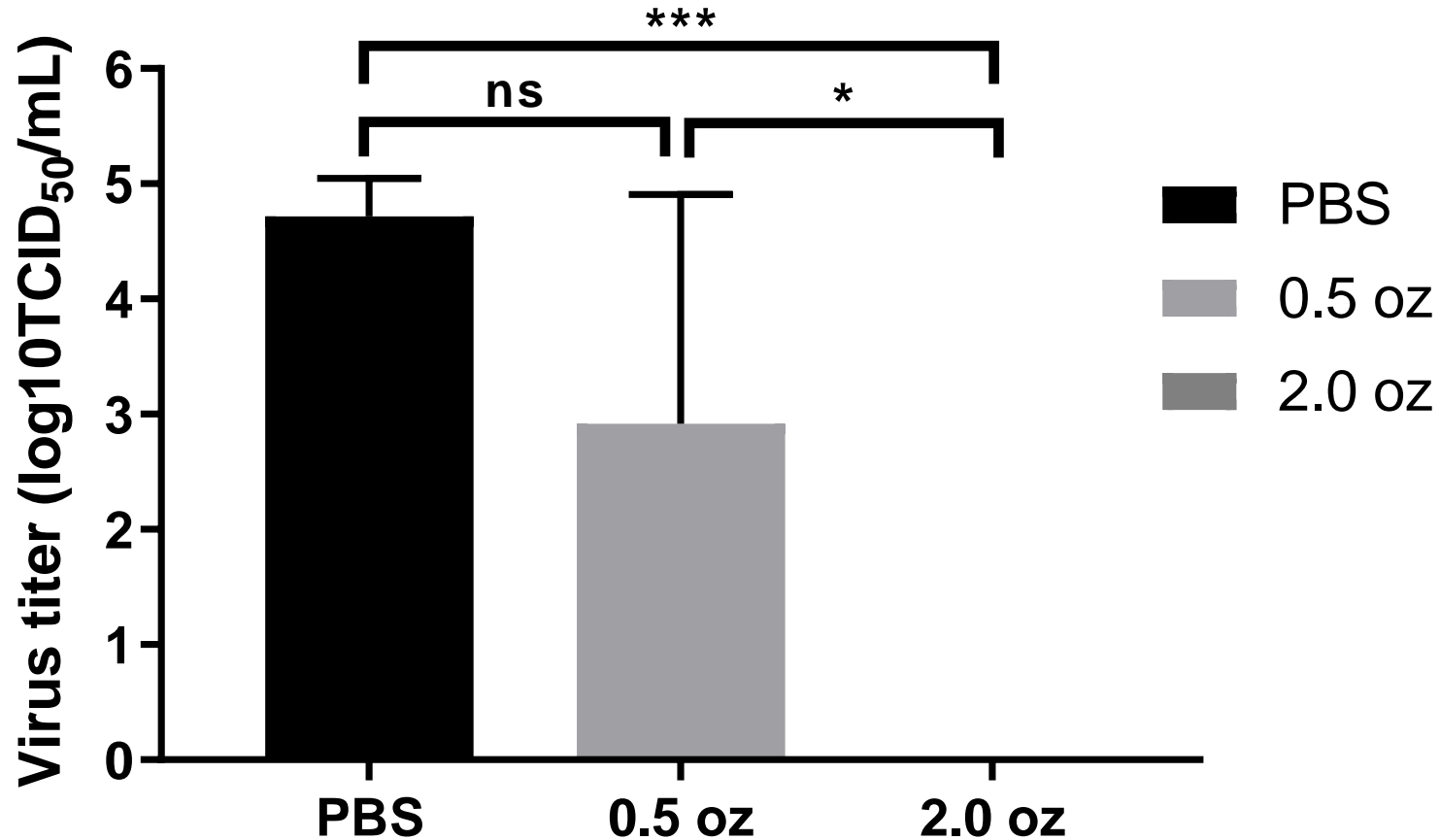
Avian influenza virus: 10^4 TCID₅₀ (PFU)/ml

Protocol: 4 wells for each virus (Avian or equine virus) that will be sprayed with XB (4 wells for XB at 0.5 oz/gallon or 2 oz/gallon solution) or PBS (4 wells for PBS)

- 1) Per well with 0.5 ml of each virus (10^4 TCID₅₀), then the 4 wells will be sprayed with XB or PBS for 5 times for incubation of 10 mins at room temperature, then the samples will be collected for virus titration.
- 2) Each collected sample will be titrated in MDCK cells for four repeats based on the fluorescence signals or cytopathic effects to detect the live virus.
- 3) The virus titers of each treatment groups will be analyzed.

Results: Both concentrations of XB (0.5 oz/gallon and 2.0 oz/gallon) can inactivate and reduce virus titers of both avian and equine influenza viruses. However, the high concentration of XB (2.0 oz/gallon) is significantly effective to inactivate both viruses as no live virus was detected in treated samples of both viruses. In contrast, the low concentration of XB (0.5 oz/gallon) can inactivate both avian and equine influenza viruses, but live viruses can still be detected in treated samples.

H9N2 virus treated with PBS or different concentrations of XB



The 10⁴ TCID₅₀ of H9N2 virus expressing GFP was treated with PBS or different concentrations of XB (0.5 oz/gallon and 2.0 oz/gallon) for 10 min at RT via unintentional spray 5 times, and the samples were collected and titrated on MDCK cells.

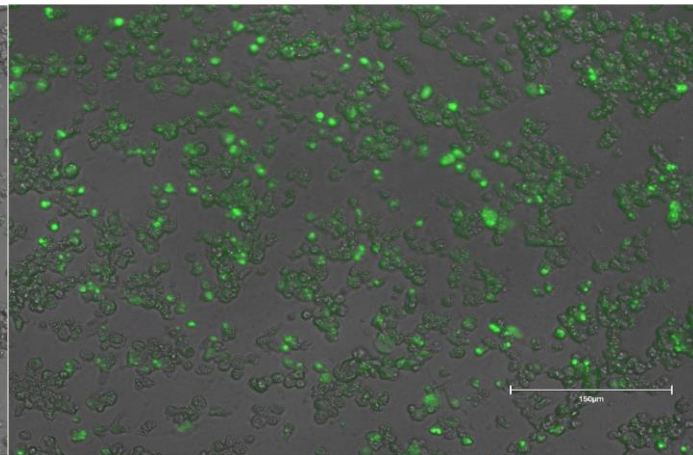
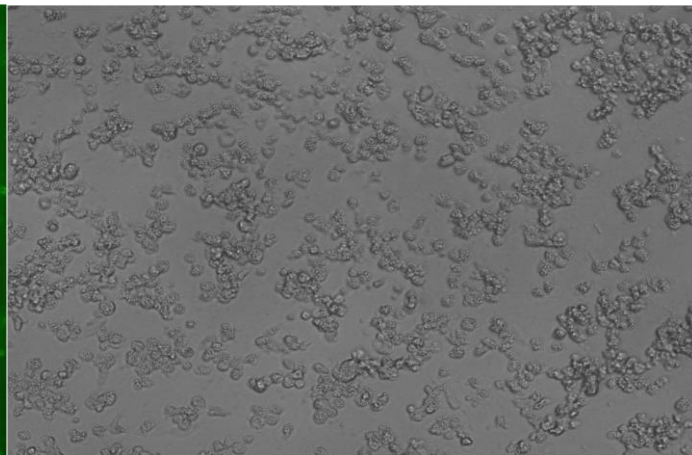
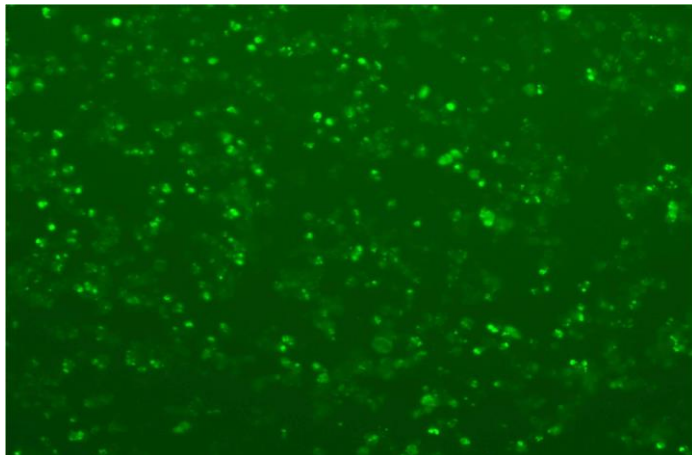
Result: 0.5 oz/gallon XB solution cannot significantly inactivate H9N2 virus, but more than one log decrease was observed. In contrast, 2.0 oz/gallon XB solution can completely inactivate H9N2 virus.

GFP

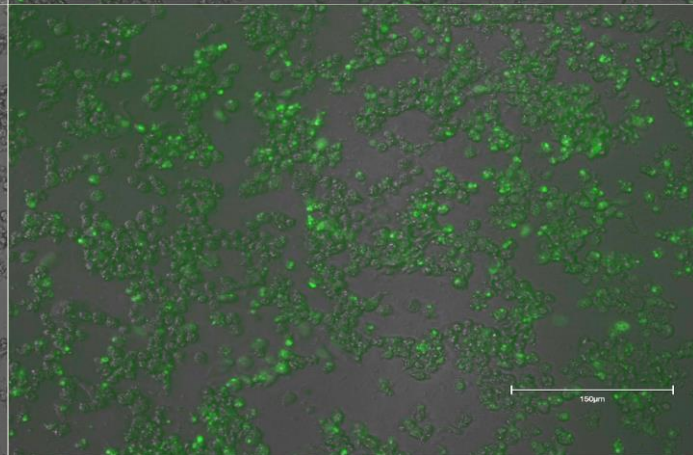
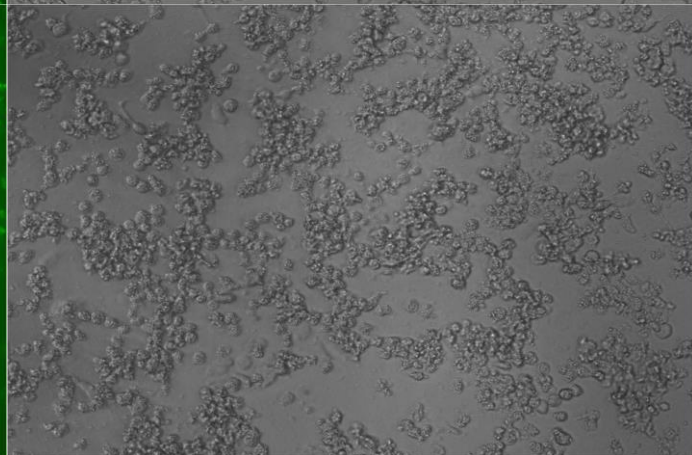
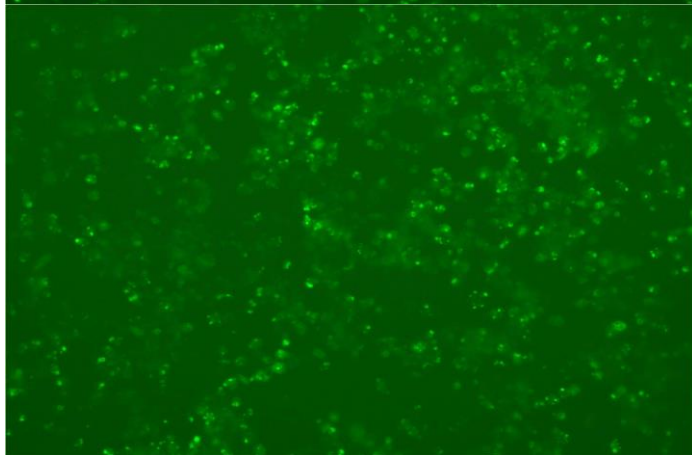
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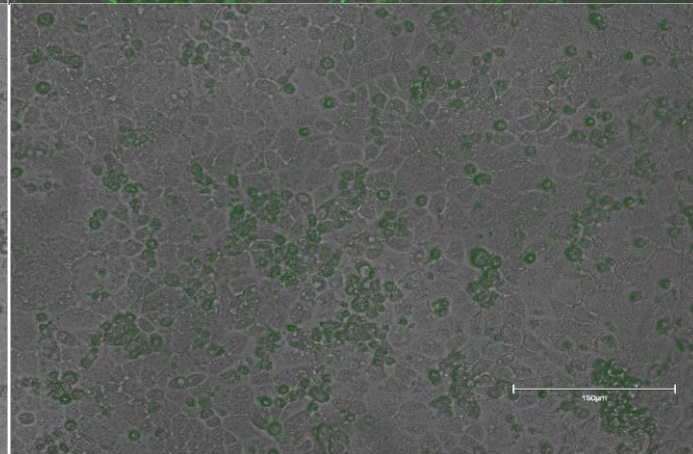
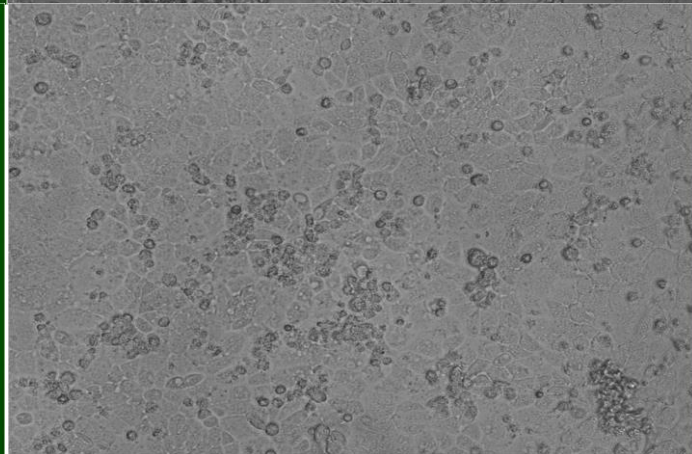
PBS



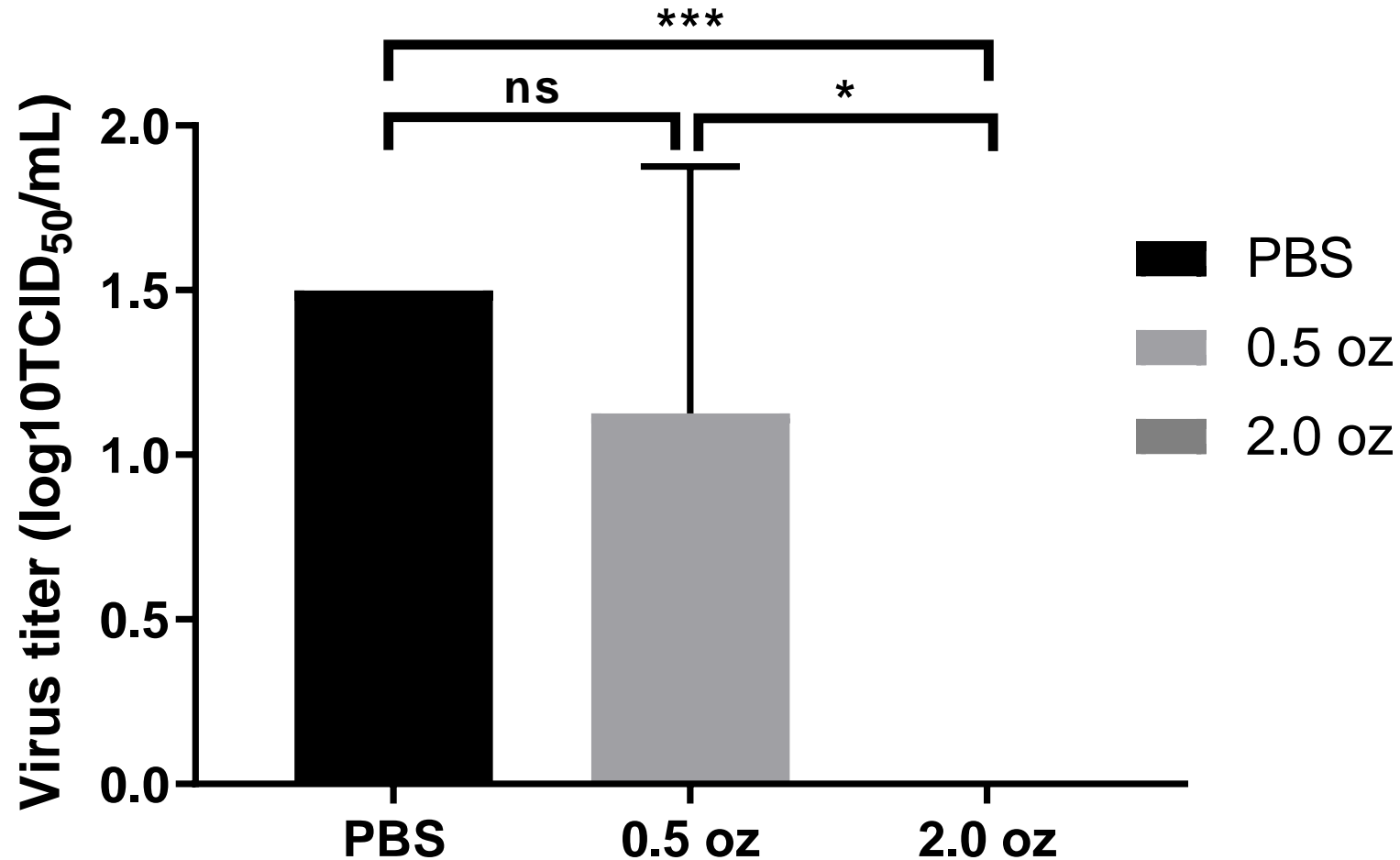
0.5 oz



2.0 oz



EIV treated with PBS or different concentrations of XB



The 10^4 TCID₅₀ of equine influenza virus (EIV) was treated with PBS or different concentrations of XB (0.5 oz/gallon and 2.0 oz/gallon) for 10 min at RT via unintentional spray 5 times, and the samples were collected and titrated on MDCK cells.

Result: Due to the reactivity between our universal anti-NP antibody and EIV, the viral titer of treated-EIV was lower than expected, but a similar trend of decrease in viral titer was also observed in EIV.

Anti-NP

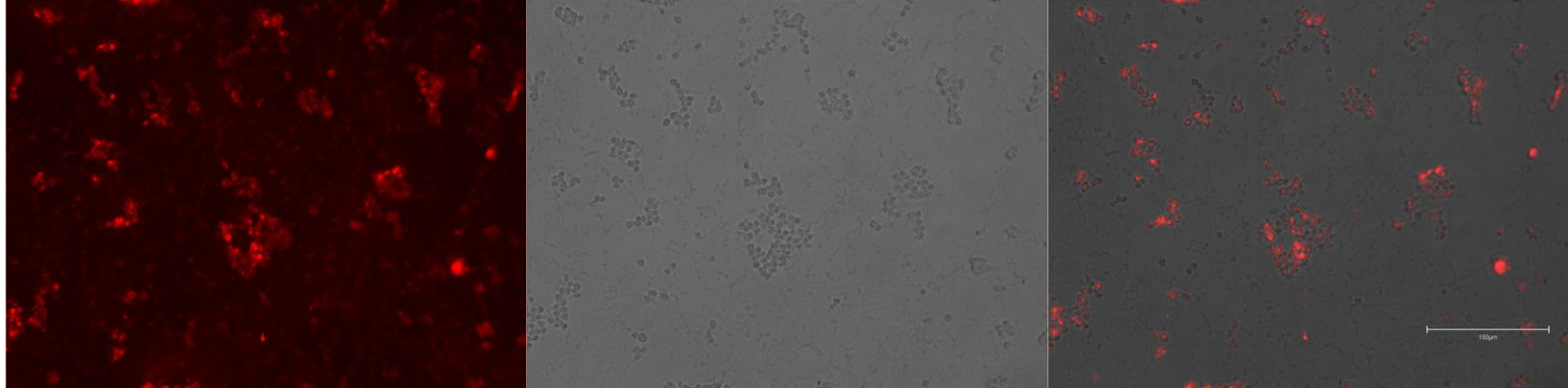
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PBS



0.5 oz



2.0 oz

