Adaptation of Influenza A/Kansas/14/2017 (H3N2) to BALB/c Mice

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Introduction

Influenza viruses can infect a human host as part of their infectious life cycle and have been responsible for large-scale pandemics and illnesses. However, not all strains of influenza that infect humans are capable of being transmitted from person-to-person[1].

Mice are used in animal research due to their short reproduction time, low cost, and general adaptability when it comes to developing models for the study of viruses. However, certain viruses, such as the Influenza A/Kansas,14/2017 (H3N2) strain utilized in this study, may not readily infect mice and cause disease. Serial passaging of the virus in mice allows the virus to adapt to cause disease in mice in order to use these mouse-adapted viruses to evaluate antiviral drugs and vaccines. The purpose of this study was to adapt an influenza virus strain to be more virulent in BALB/c mice and enhance the virus' ability to infect these mice.

Mice were infected via the intranasal route and observed for signs of disease. Mice were euthanized and virus was recovered from the lungs of infected mice. The lung tissue was homogenized, and the virus isolated from lung tissue was used to infect the next set of mice. Serial passaging increased virulence of the virus and increased mortality in mice was observed after passaging. Mannan was initially used to aid in adaptation by enhancing influenza virus replication through inhibiting host defense collectins but removed for later passages[2].

Materials and Methods

Animals:

Eight-week-old female BALB/c mice

- Autoclaved bedding and water

Virus:

- Influenza A/Kansas/14/2017 (H3N2)

Serial Passaging:

- Groups of 5 animals for each passaging study
- Virus serially-passaged until consistent mortality is observed in mice
- Necropsy mice three days after infection to collect lung samples
- Infect mice via the intranasal route with 90 µL of MP7
- An intranasal treatment (75 µL saline) was used to increase infection severity.

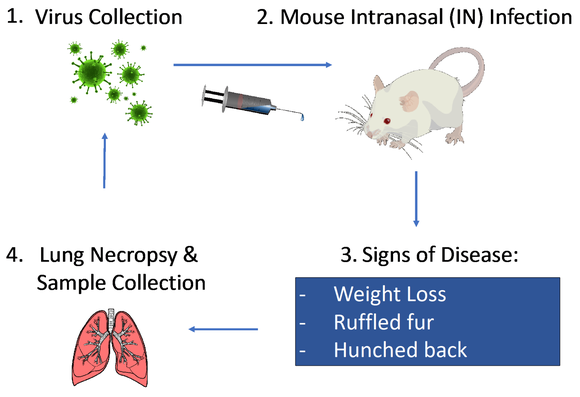

Figure 1. Serial passaging of influenza virus in mice. Mice were infected by intranasal instillation of virus. After three days, the mice are euthanized, and lung tissue homogenized to isolate the virus. The supernatant containing influenza virus is used to infect subsequent groups of mice. This process is repeated several times over to yield several passages of virus.

Results

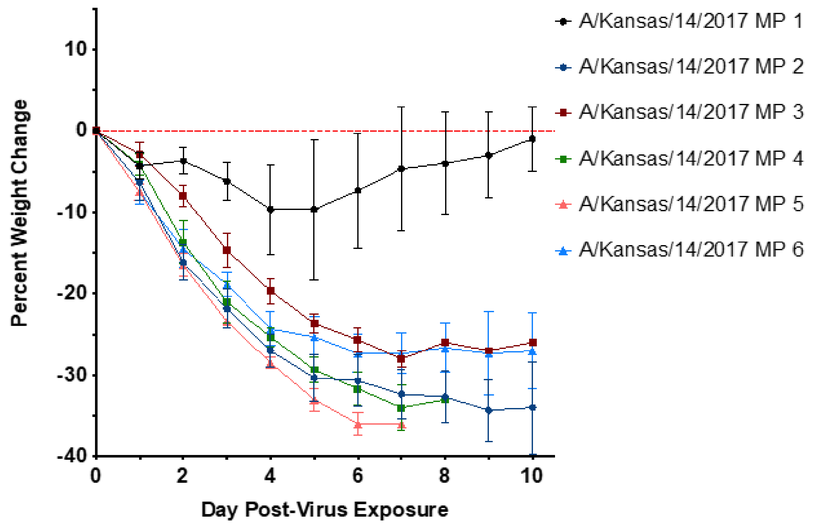

Figure 2. Percent weight change of BALB/c mice infected with influenza A/Kansas/14/2017 MP1-MP6 (with mannan). Mannan was introduced to as a binding agent to aid in virulence and allow virus easier entry into the respiratory tract and lungs. More severe weight loss was observed after serial passaging of the virus.

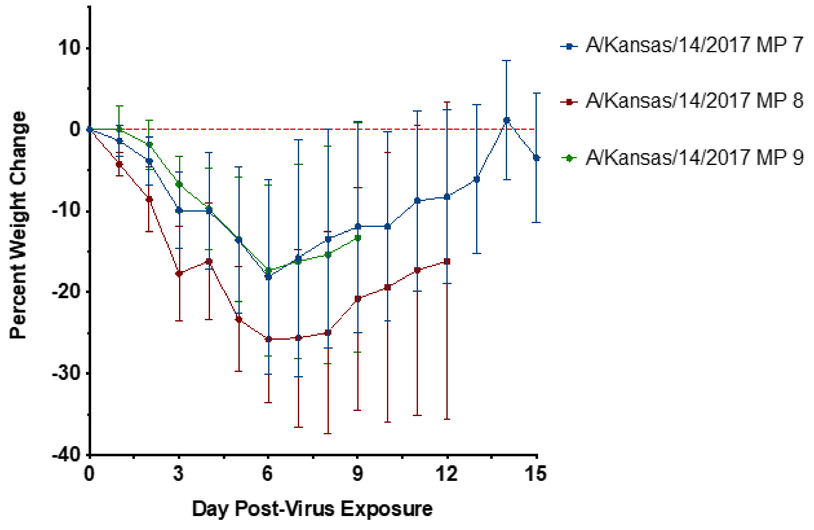

Figure 3. Percent weight change of BALB/c mice infected with influenza A/Kansas/14/2017 MP7-MP9 (without mannan). Mice were infected without the use of mannan and an intranasal treatment (75 µL saline) was completed on days 1 and 2 post-infection. Without the use of mannan, the virus caused significant weight loss in infected mice. Mortality was also observed (1 of 3 mice) in each infected group .

Conclusions and Future Studies

Conclusions:

- Serial passaging increased the lethality of the virus
- Weight loss was observed in mice infected without the use of mannan

Future Studies:

- Titration of lung tissue samples to determine concentration of virus in homogenate
- Continue passaging until the virus can consistently cause mortality (≥90%)
- Complete sequencing of mouse-adapted virus stock
- Evaluate antiviral therapies or vaccines using mouse-adapted virus

References

1. Bouvier, N. M. & Lowen, A. C. Animal Models for Influenza Virus Pathogenesis and Transmission. *Viruses* **2**, 1530–1563 (2010).
2. Smee, D. F., Wandersee, M. K., Wong, M. H., Bailey, K. W., & Sidwell, R. W. (2004). Treatment of mannan-enhanced influenza B virus infections in mice with oseltamivir, ribavirin and viramidine. Antiviral chemistry & chemotherapy, 15(5), 261–268.

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