Development of Murine Model for Enterovirus D68 in AG-129 Mice

UtahStateUniversity
DEPARTMENT OF BIOLOGY

Institute for Antiviral Research

John McClatchy, Joseph Evans, Brett Hurst, Bart Tarbet

UtahStateUniversity
DEPARTMENT OF BIOLOGY

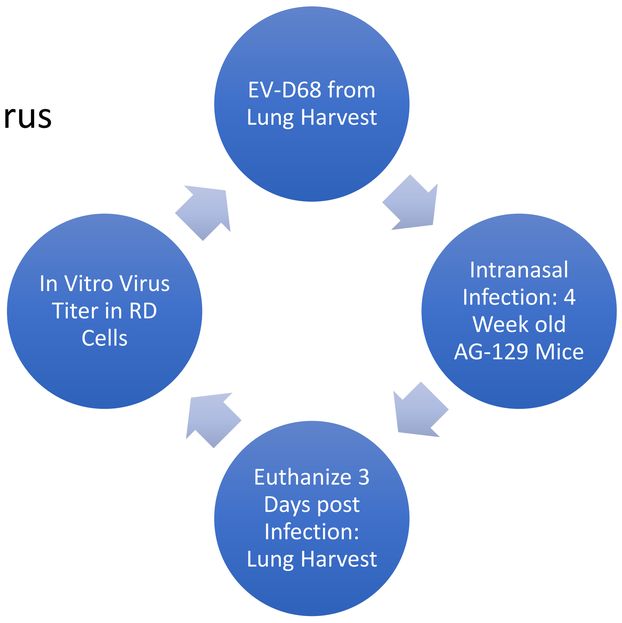
Introduction

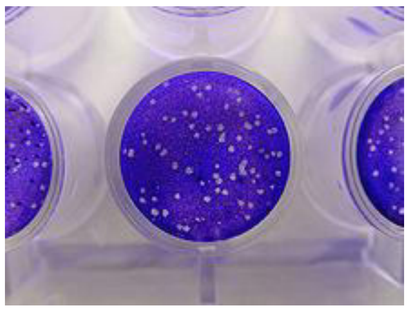
Enterovirus D68 (EV-D68) is an emerging picornavirus virus which typically causes respiratory disease. In 2014, a nationwide outbreak of EV-D68 occurred, with a portion of these cases associated with neurological disease. At the time of this outbreak, no animal models existed for Enterovirus D68, making it difficult to characterize pathology and test potential therapeutics. To address this, we developed a mouse model of EV-d68 infection in AG-129 mice (immuno-compromised mice).

Methods (Design?)

Serial Passaging of Enterovirus D68 in Mice

Serial passaging was first utilized in order to adapt Enterovirus D68 to a murine host. Enterovirus D68 was passaged 30 times.





Plaque Purification of EV-D68

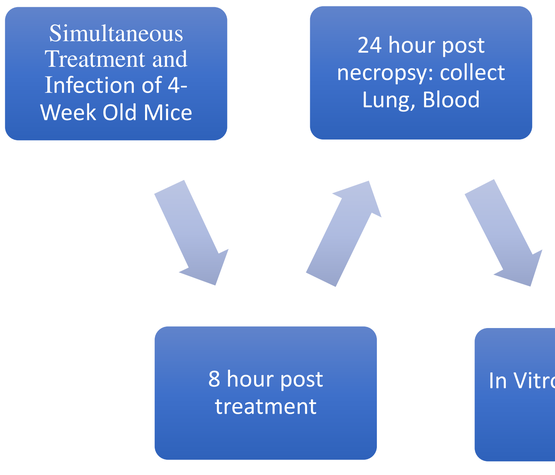
Virus was harvested from a single plaque. This allowed us to work with the progeny of a single virion of our adapted stock, hereby known as mp30PP (mouse passage 30 plaque purified)

Confirmation of Viral Strain via PCR

To exclude the possibility that MP30PP was a contaminant, the stock was confirmed with real time reverse transcriptase PCR.

Comparison of Mouse Passages

6 groups of mice were infected, with either MP 0 (Mouse Passage 0), MP 10, MP 20, MP 30, MP30PP, or placebo (uninfected). 3 days post infection mice were euthanized, lungs, liver, kidney spleen and blood were taken and tittered in RD cells.

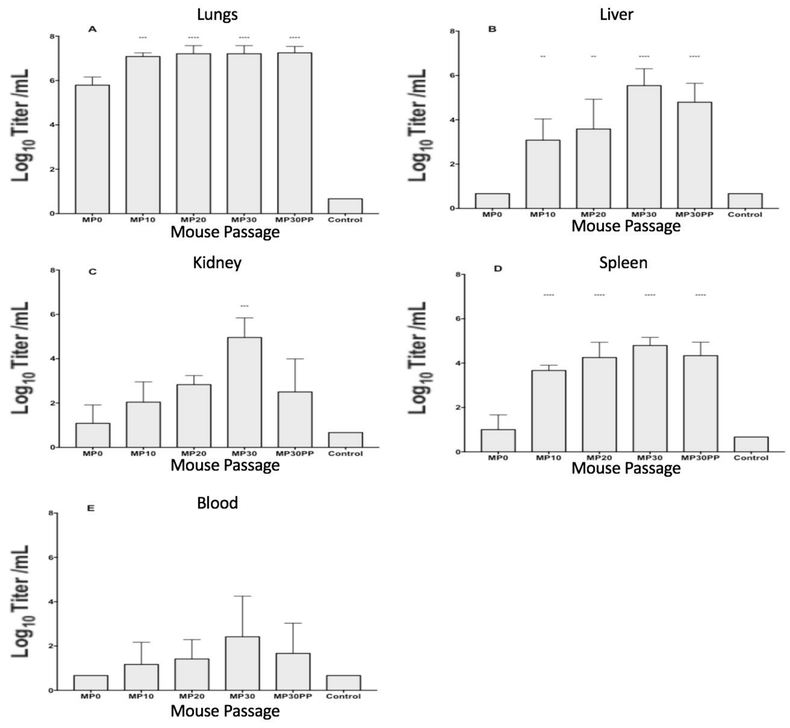


Establishment of a Positive Control

Mice were divided into six groups and infected with MP30PP. Each group was given either, Ribavirin 500 mg/kg, Ribavirin 300 mg/kg, Pleconaril 10mg/kg, Guandine 200 mg/kg, Enviroxime 200mg/kg, or MEM, at the time of infection and 8 hours post infection. Lung and blood were collected 24 hours post infection and tittered in RD cells.

Results

Comparison of Mouse Passages



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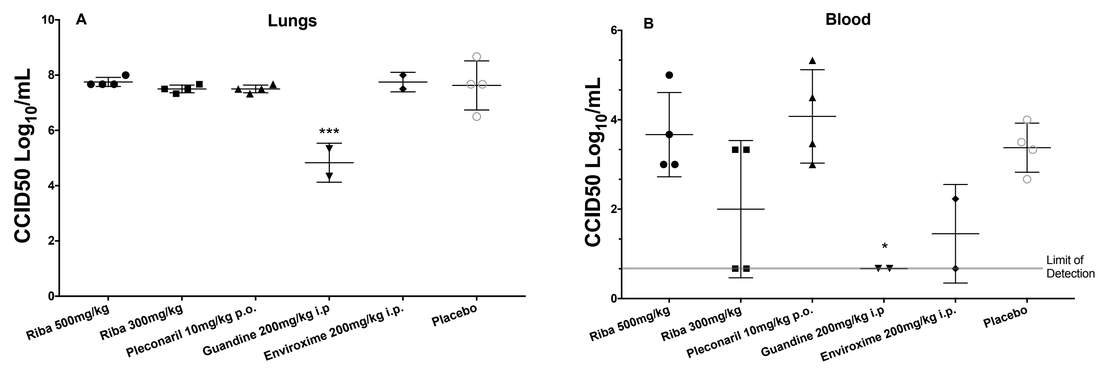

Conclusion

Qualitative Description Mouse Model

Infection with EV-D68 of four-week-old mice yields mild respiratory disease with, with titers in the lungs, liver, kidney, spleen, and blood. Infection of ten-day-old mice yields neurological disease, including flaccid paralysis.

Potential Future Utilizations of Model

- Screening of additional therapeutics against Enterovirus D68
- Determination of the mechanism of viral entry to the central nervous system
- Determination of the mechanism by which enterovirus D68 causes flaccid paralysis in mice

Acknowledgements

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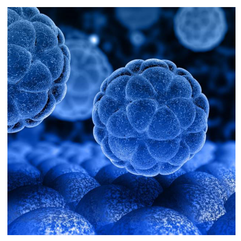

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