



Preclinical evaluation of vaccines and antivirals in RSV challenge SCID mouse models

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Executive Summary

Presently, there is no targeted treatment available for human respiratory syncytial virus (hRSV) infection other than the supportive care to mitigate the signs of and symptoms. Antiviral drug development and RSV vaccine is expected to play a critical role in overall reduction of RSV infections globally. Continued effort in RSV research published in high profile scientific journals has invigorated efforts by larger and smaller pharmaceutical and biotechnology companies to develop antivirals and RSV vaccines. Preclinical development of vaccines is hindered by the lack of clinically relevant rodent models. More importantly, the safety of new vaccines, preclinical pharmacological, safety and toxicology studies should be conducted prior to initiating the clinical studies. Aragen will enable successful IND-enabling preclinical RSV therapeutic development studies by providing appropriate study designs, right animal models, and confirming effective immune response by performing *in vivo* and *in vitro* assays. Our scientists have and will continue to establish the identity, purity, safety, and potency of the vaccine or antivirals. In this white paper, we present several case studies performed in clinically relevant mouse and rat models to assess the critical characteristics of potential RSV therapeutics. In summary, this article includes in-house as well as client-sponsored study data established over several years using SCID mouse models. These studies generated clinically pertinent information for proper screening of new vaccines and antivirals. Aragen will continue to support your RSV therapeutic development effort by providing reliable and effective preclinical services in this important infectious disease area.

Introduction

As per Centers for Disease Control and Prevention (CDC), human respiratory syncytial virus (hRSV) is a common respiratory virus that usually causes mild, cold-like symptoms (1). RSV and its chimeric strains affect infants, adults, elderly worldwide and cause significant morbidity. Aragen continues to serve pharmaceutical, biotechnology and SMEs in their preclinical efficacy and safety studies. Appropriate in vivo rodent models enable development of anti-RSV antibodies, small molecules, and vaccines for the treatment of respiratory disease.

RSV is a leading cause of infant hospitalization. RSV infection causes cold-like symptoms and often progresses to bronchiolitis and pneumonia in infants. The cold-like symptoms seen in RSV infected patients appears to be like common flu in infants. However, infants and older adults exhibit symptoms differently. In infants, the RSV infection might occur at age 2 and remain unnoticed until infant exhibits severe RSV symptoms whereas in adults RSV symptoms are milder and less easy to identify. There are no targeted antivirals or vaccines readily available to treat or prevent RSV infection. Several pharmaceutical and biotechnology companies are striving to develop effective RSV vaccines.

Treatment

Typically, RSV infections go away on their own in a week or two. Virus is contagious, spreads from infected people to others living in close contact with infected persons or areas. There are few steps taken to relieve symptoms, such as managing fever and pain, giving enough fluids, and contacting healthcare provider if the symptoms persist. There is no specific treatment for RSV infection, though efforts are on to develop vaccines and antivirals. Palivizumab is a monoclonal antibody recommended by the American Academy of Pediatrics (AAP) is prescribed to prevent severe RSV illness in certain infants and children.

Present Status of RSV Vaccines

Unfortunately, no vaccine is available against RSV for more than 50 years of effort. Main hurdle for developing RSV vaccine appears to be its multiple mechanisms of evading immunity, hence it reinfects people throughout life with relatively low genetic variation compared to retro (RNA) viruses. The recent availability of structure and antigenic content of the fusion (F) glycoprotein in its metastable untriggered prefusion form (pre-F) and the stable rearranged post fusion form (post-F) gave an impetus to develop vaccine strategies (2). Hence, novel live-attenuated and chimeric virus vaccine candidates and other novel approaches to deliver vaccine antigens have been developed as vaccine products to fight RSV infection (2). Fight against RSV starts with maternal or infant immunization. RSV vaccine should reduce elderly mortality, which is still high. In both populations, vaccine safety is highly critical (3). Palivizumab currently used for treatment of high-risk infants with RSV infection. Developing RSV mice models to measure the RSV-specific neutralizing hu-MoAbs after RSV infection is crucial for early detection and therapeutic intervention (4) in infants.

Preclinical RSV models for vaccine development

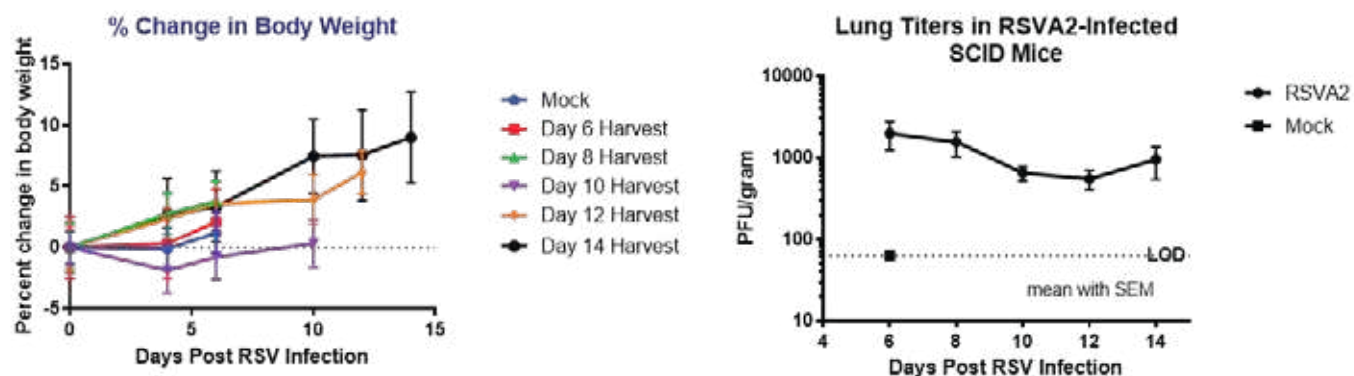
The RSV mouse model is the preferred choice for most preclinical for most immunological studies, ranging from simple vaccine testing to the intricate assessment of fundamental immunopathogenic responses. An important preclinical study showed the generation of RSV-specific neutralizing hu-MoAbs after intravenous injection of immunoglobulins and Palivizumab, a 'humanized' anti-RSV-F MoAb (4). In last few years, neonatal mice (aged <7 days at time of initial infection) model is being validated to understand RSV infection in infants (4). Researchers showed that downregulating IL-4R α allows for the survival of Th1 cells in the presence of Th2 cytokines such as IL-4/IL-13. This finding demonstrates the importance of the age of initial infection of RSV in determining disease outcome. Therefore, the key to a successful vaccine lies in administration of immunomodulatory agents at the time of vaccination (4).

Highlights of RSV preclinical Services at Aragen

- Experts with several years of expertise, delivered over 100 successful RSV studies (30-150 animals/study)
- A wide range of readouts available with regular updates throughout study
- Flexible and customizable assays in RSV models. We have tested small molecules, large molecule, and vaccines for past and existing clients/ partners
- Willing and able to develop models as per client specification with customized studies

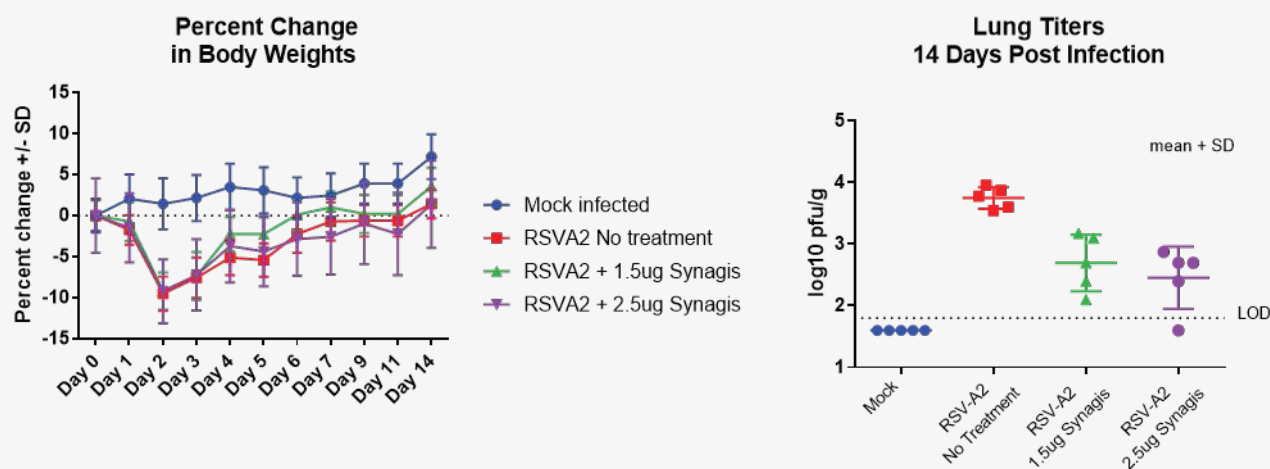
Case Study 1: SCID mouse model to measure RSV-A2 replication

To measure the replication of RSV-A2 in immunocompromised mouse model, we intranasally injected placebo group with vehicle and test groups with RSV-A2 (1x10⁵ pfu), monitored body weight for over 14 days in different groups of mice before harvesting lung tissues for RSV measurement on days 6, 8, 10, 12 and 14 days. We observed that RSV-A2 replication was prolonged in SCID mice over 14 days compared to placebo (mock) group.



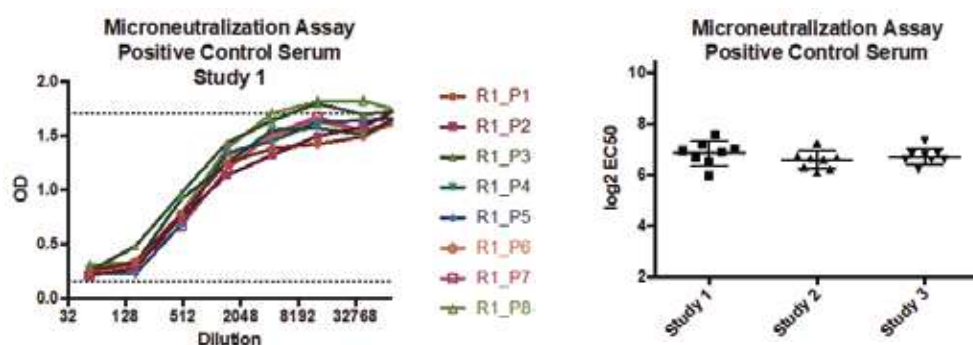
Case Study 2: Effect of Synagis® on RSV-A2 replication in SCID mouse model

Female SCID mice, 6 weeks old were inoculated intranasally with RSV-A2 (1x10⁵ pfu), treated with Synagis® IP on days 2, 4 and lung tissues harvested on day 14 for RSV burden analysis. We observed that RSV-A2 replication was prolonged. However, the administered doses of Synagis® provided partial protection against RSV infection.



Case Study 3: Vaccine development- RSV neutralization assays.

Aragen participated in a WHO collaborative study to establish the 1st International Standard for antiserum to RSV. A positive control standard run on every plate in microneutralization assay. Options for microneutralization or plaque reduction assay was adapted to develop in- house or client's protocols.



Bibliography

Note: References 6-9 pertain to Aragen generated case studies.

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About the Author

Dr. Nagendra has held progressive leadership roles in pre-clinical and clinical oncology research and development. He worked at academic centers like-University of Kansas, Cedars-Sinai Medical Center, Mercer University Medical Center, Vanderbilt University Medical center, Anderson Cancer Institute. He led research teams on brain tumor and breast cancer biology. He had extensively published in peer-reviewed journals and secured US, EU and Japan patents. He directed the Human Tissue banking and biorepository and New Animal Facility. Later he worked in pharmaceutical companies such as Dr. Reddy's Labs, Scintilla BioMarc, PPD/ Thermo Fisher Scientific with focus on in vitro diagnostics, clinical pharmacology and toxicology, medicinal chemistry aspects of clinical drug development. He is presently a Global Senior Director of Scientific Affairs at Aragen Life sciences, USA.

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