Poster # 73

Binding Kinetics of RSV Pre-fusogenic F Nanoparticle Vaccine to Palivizumab and Serum Polyclonal Antibody

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BACKGROUND

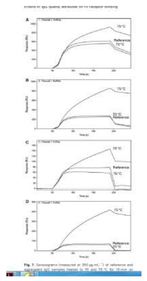
Respiratory syncytial virus (RSV) is one of the most common causes of acute lower respiratory tract infections in infants and young children worldwide. RSV Pre-fusogenic F nanoparticle vaccine has been shown to be safe and immunogenic in clinical studies in young women and older adults. ResVax (Pre-fusogenic RSV F nanoparticle vaccine) is currently being assessed in the Prepare™ (Phase 3) trial for the protection of infants via maternal immunization in healthy third trimester pregnant women. It has been demonstrated in animal and human studies to induce anti-RSV F antibodies competitive with palivizumab (Synagis®) for binding to antigenic site II. High antibody binding affinity to neutralizing epitopes would likely contribute to a protective immune response, a desirable characteristic of an RSV vaccine response. In this study, we characterize Pre-fusogenic F nanoparticle vaccine binding kinetics.

OBJECTIVES

- To determine if there is protein aggregation following accelerated stability conditions.
- To compare palivizumab binding kinetics from Pre-F and Post-F vs Pre-fusogenic vaccine.
- To test immune sera for RSV F antigenic site II peptide binding avidity to Pre-fusogenic vaccine.

METHODS

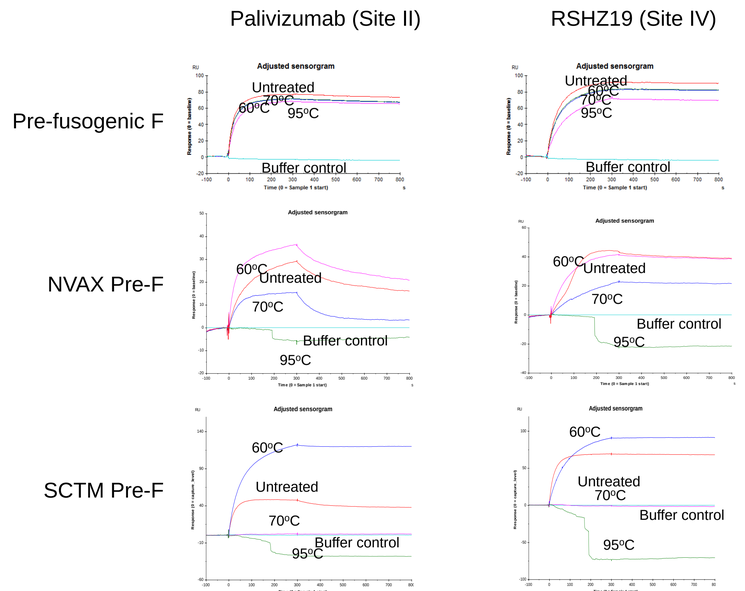
- Methods were based on stress conditions using heat treatment of recombinant IgG near the Tm, which results in changes in FcR binding. This is due to loss of binding sites or protein aggregate formation as reported by Geuijen et al. 2017 (right figure). A similar approach was applied to RSV F constructs heat stressed near the Tm and effect on antibody binding.



Geuijen et al. Openbio 2017

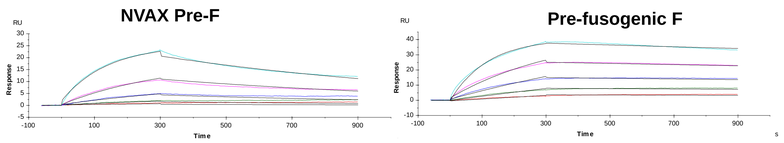
- Binding affinity of F constructs to site II (palivizumab) and site IV (RSHZ19) neutralizing antibodies were analyzed using Biacore T200 surface plasmon resonance (SPR).
- Serum binding avidity (k_{off}) site II peptide were measured by injecting antibodies across a site II peptide surface. All sensorgrams were double referenced.

RESULTS



RSV F protein	Tm1 (ºC)	Tm2 (ºC)
Pre-fusogenic F	92.1	105
NVAX Pre-F	64.9	79.1
SC-TM Pre-F	52.5	64.2
Post-F	54.6	93.1

Fig. 1 Sensorgrams of untreated, 60ºC, 70ºC and 95ºC treated RSV F constructs binding to Palivizumab or RSHZ19. Tm of RSV F constructs are shown in the Table.



Analyte	ka (1/Ms)	kd (1/s)	KD (nM)
NVAX Pre-F	2.51E+04	1.00E-03	40.0
Pre-fusogenic F	2.78E+04	1.63E-04	5.9

Fig. 2 Palivizumab binding kinetics to NVAX Pre-F and Pre-fusogenic F. Binding affinity of RSV F constructs are shown in the Table.

RESULTS

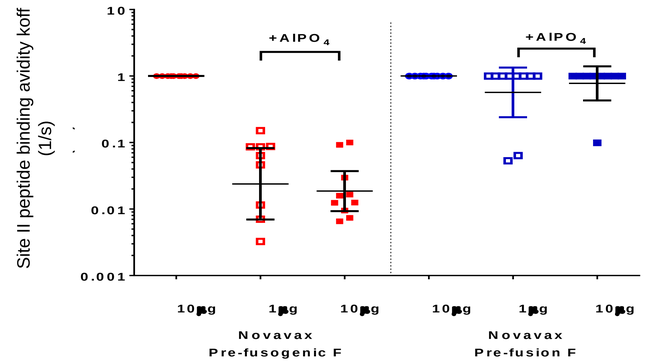


Fig. 3 Site II peptide binding avidity koff rate (1/s) sera from mice immunized with pre-fusogenic F or pre-fusion F with and without AlPO4 adjuvant. A biotinylated site II antigenic peptide was captured on a streptavidin sensor chip. Day 35 mouse serum (n=10) was diluted at 1:20 in HBS-EP+ buffer were injected through peptide surface for 4 minutes and followed by HBS-EP+ buffer for10 minute. Using a 1:1 fit model the koff rate determined.

CONCLUSIONS

- RSV Pre-fusogenic F nanoparticle vaccine is not aggregated even following treatment at elevated temperatures.
- Pre-F and Post-F constructs form protein aggregates when heat treated near Tm.
- Palivizumab has >6-fold higher affinity (KD) to RSV Pre-fusogenic F vaccine than Pre-F.
- RSV Pre-fusogenic F with $AlPO_4$ adjuvant induced higher affinity serum antibody against site II peptide in mice than Pre-F antigen.

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Conflict of Interest Disclosure: HL, JHT, MGX, NP, MM, JW ,SB, GG, and GS are employees of Novavax, Inc. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter of materials discussed in the poster apart from those disclosed.