Enhanced immunogenicity conferred by TNFR superfamily members ligation *in vivo*

Lavanya Mahadevan

Supervisor: Prof. Muthumani Karuppiah M.S., PhD

University of Pennsylvania

Background:

- Tumor Necrosis Factor superfamily ligands (TNRSFL) are co stimulatory molecules that have been reported to be involved in T cell activation and have been used as adjuvant in several vaccination studies.
- GITRL and TWEAK are members of TNFSFL that modulates natural and acquired immune response. It was concluded from a study that GITRL (Glucocorticoid induced TNF receptor ligand) induced signaling is mediated by ERK1/2, which then triggers the activation of the transcription factor NF-κB. NF-κB controls the expression of several pro-inflammatory mediators such as chemokines and cytokines.
- TWEAK (TNF like weak inducer of apoptosis) shares a common feature with GITRL that it plays a vital role in the signaling pathways that involves an activation of NF-κB. It also acts as a switch from the innate to the adaptive immune response.

Aim of the study

To evaluate the immune modulatory effects produced when the DNA plasmids encoding GITRL and TWEAK were used as molecular adjuvants, in combination with HIV-1 Tier 2 (ZM53M.PB12) DNA vaccine.

Methods:

-Ten groups of female Balb/c mice were immunized thrice intramuscularly with electroporation.

-Various concentrations of human full length TWEAK & GITRL expressing vectors (T & G resp.) were co-vaccinated with HIV-1 DNA vaccine.

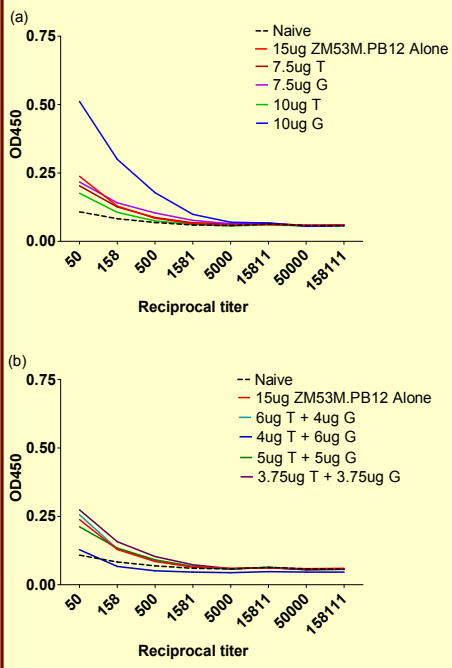
-One week following each immunization, the mice were bled and the sera was used to perform ELISA. One week following booster dose 2, the mice were sacrificed and the spleens were isolated and processed to perform ELI Spot assay.

Conclusion

1. A 2 to 2.5 fold increase in cellular response was observed in 7.5µg T and 10µg G co vaccinated groups.

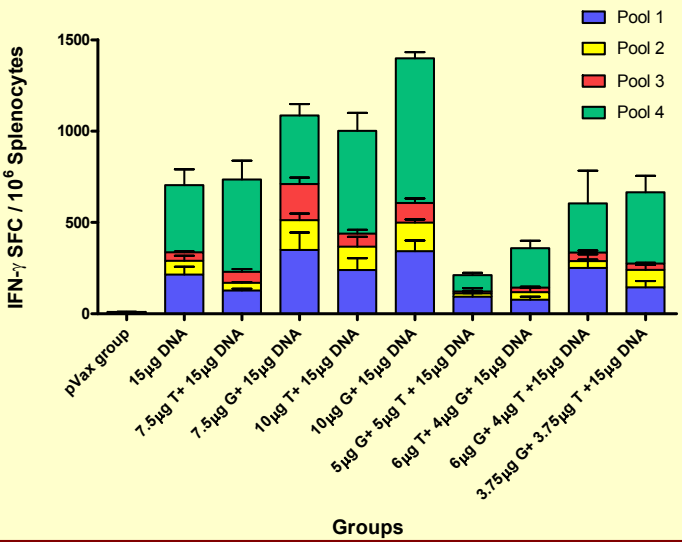
2. The humoral response was found to be significantly higher in 10µg G co vaccinated group, suggesting 10µg GITRL as a potential adjuvant for triggering a higher immune response against HIV-1 Tier 2 antigen.

Result 1-Humoral response, ELISA data:



The above figure shows the humoral responses obtained when 'G' and 'T' were used at various doses (a) separately; (b) in combination along with HIV-1 DNA vaccine. It can be noted from (a) that a higher level of response is obtained from 10µg G co-vaccinated group. A statistical test (paired t-test) performed upon the same revealed a significance with a p value of 0.022 ($p<0.05$)

Result 2-Cellular response, ELI Spot data:



The figure shows the responses obtained from all the ten groups of animals that were used for the experiment. Pool 1, Pool 2, Pool 3 and Pool 4 refer to the peptide pools (that were used to stimulate the splenocytes) of 15mer peptides that were specific against the HIV-1 antigen used in the study. A 2 and 2.5 fold increase in the level of cellular response was conferred by the 7.5µg T & 10µg G co-immunized group of animals when compared to the DNA (parent vaccine) vaccinated group.

Acknowledgements

I would like to thank my supervisor and my dear colleagues for having supported me throughout this thesis.

Contact: swaanya@gmail.com