Generation of antibodies for the diagnosis of acute and chronic Chagas Disease using phage display technology

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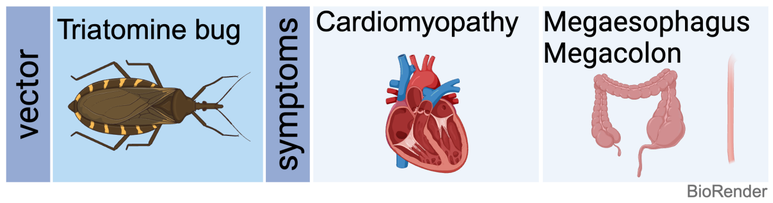
Background

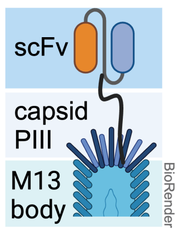
What is Chagas Disease?

- Caused by the parasite *Trypanosoma cruzi*
- 30% of individuals develop life-limiting cardiac and gastrointestinal complications
- <1% of infections are identified and treated
- No antigen detection assay exists



What is M13 Phage Display?

- Method for generating single-chain variable fragments (scFv's)
- Relies on bacteriophage's ability to express extracellular proteins linked to a capsid protein



Objectives

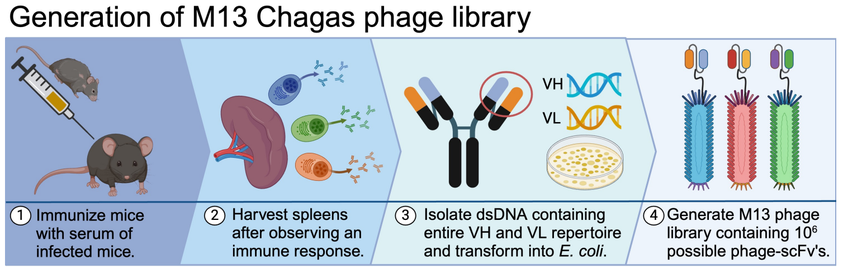
Aim 1: Identification of circulating antigens by using a non-directed approach to immunization followed by an antigen-identifying microarray.

Aim 2: Generation of antibodies (Abs) specific for *T. cruzi* using M13 phage display technology.

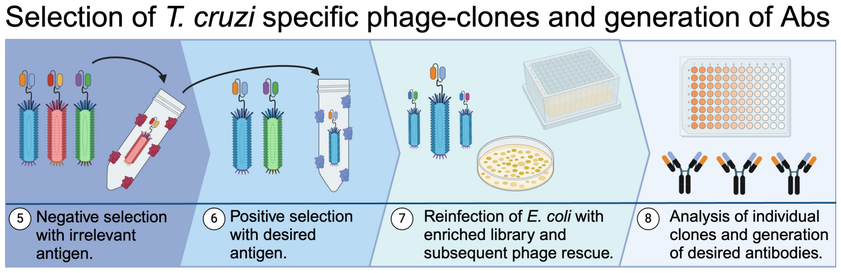
Aim 3: Demonstrate detection of acute and chronic *T. cruzi* infection in mice by ELISA.

Methods

Generation of M13 Chagas phage library



Selection of *T. cruzi* specific phage-clones and generation of Abs



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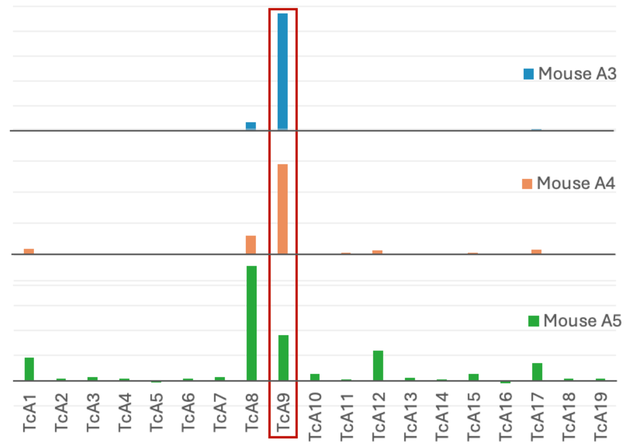
Results

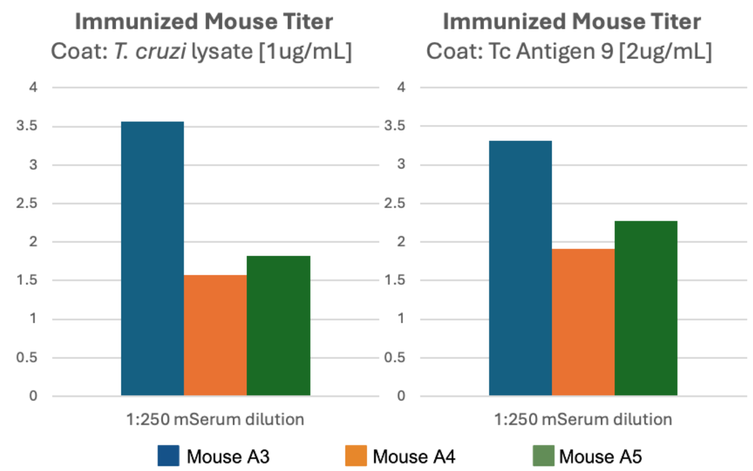
- Mice immunized with *T. cruzi* infected serum.
- Microarray panel of serum using 19 known *T. cruzi* associated proteins.

Identification of circulating *T. cruzi* antigens.

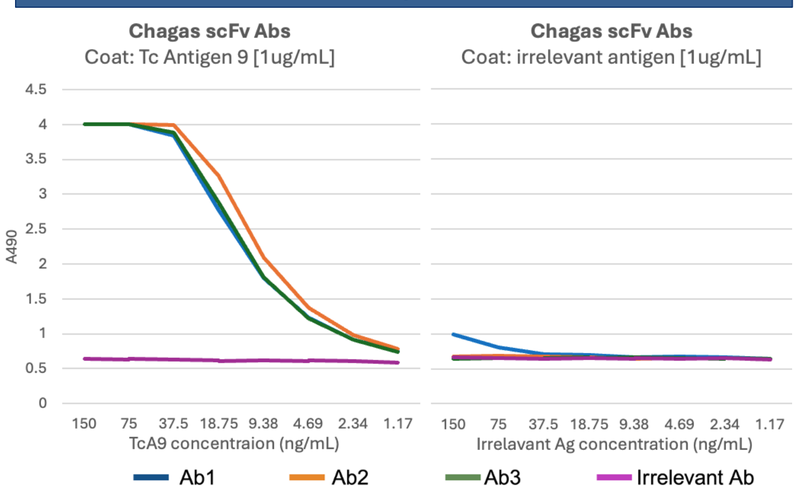


- ELISAs measuring Ab titer of mice comparing crude *T. cruzi* lysate to TcA9.

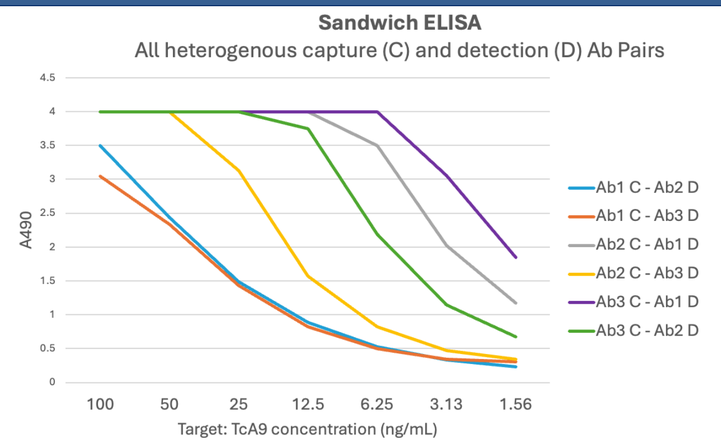
Immunized mice generated anti-TcA9 Abs.



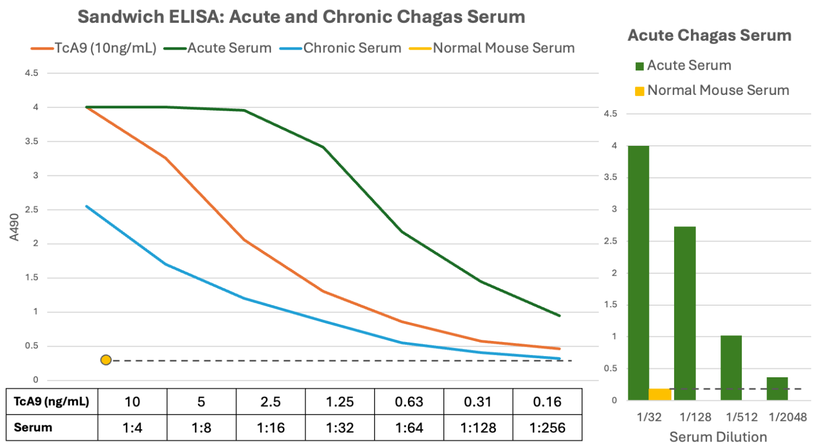
Generation of three specific anti-TcA9 Abs.



Identification of best capture and detection Abs.



Detection of acute and chronic *T. cruzi* infection.



TcA9 (ng/mL)	10	5	2.5	1.25	0.63	0.31	0.16
Serum	1:4	1:8	1:16	1:32	1:64	1:128	1:256

Conclusion

- Our Abs detect acute and chronic *T. cruzi* infection in mice at serum dilutions of **1:2000** and **1:128** respectively.
- We anticipate these Abs can be used for point-of-care antigen detection assays, addressing the need for earlier detection and intervention of Chagas disease.

References

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