Project Title: Identification of novel SARS-CoV-2 S2 neutralizing epitopes

Preferred Scholar On-Site Project Dates: Long-term (9-12 months) - May 1, 2024 – January 31, 2025

Project Site: Birmingham, Alabama (University of Alabama, Birmingham)

Project Overview:

The Spike (S) of coronaviruses is the major target for antibody mediated protection from infection, however the S1 component of Spike, which includes the Receptor Binding Domain is undergoes substantial mutation as SARS-CoV-2 evolves, compromising the effectiveness of vaccines. In contrast, the S2 component of Spike is highly conserved across coronaviruses including SARS-CoV-2, SARS-CoV, and MERS-CoV, making it an attractive target for developing universal coronavirus vaccines. This project will utilize a variety of custom S2 proteins as probes to isolate S2 specific B cells from individuals with or without HIV for the generation of monoclonal antibodies that will be characterized for universal coronavirus activity.

Project Summary:

This project will utilize pre-existing plasma and peripheral blood mononuclear cell (PBMC) samples, established flow cytometry and molecular biology methodologies, and a custom panel of B cell probes that represent multiple regions of SARS-CoV-2 S2 to isolate and generate monoclonal antibodies to define novel epitopes that may inform future universal coronavirus vaccine development. The scholar will develop experience with systems serology, molecular biology, and anti-viral assays. Related manuscripts from the Kobie lab include: PMC9302814, PMC7904445, PMC8212047

This project is designed to be a long-term project consisting of 9 months of bench-based research conducted in the Kobie laboratory. The detailed timeline is presented below.

Major RAMP scholar study objectives and endpoints:

1) Learn ELISA and SARS-CoV-2 pseudovirus neutralization assay (wk 0- wk 3)
2) Screen plasma samples for 1) IgG binding antibodies to various S2 probes and 2) SARS-CoV-2 neutralization activity. (wk 4- wk 8)
3) Stain and flow sort S2-specific B cells from PBMC samples using S2 probe 1 and 2 (wk 8-10)
4) Learn molecular techniques for recombinant mAb generation (wk 10-12)
5) Generate S2-specific mAbs (wk 12-20)
6) Screen S2-specific mAbs for reactivity by ELISA and neutralization by pseudovirus assay (wk 20-22)
7) Repeat steps 3-7 for S2 probe 3 and 4 (wk 23-32).
8) Analyze results, prepare RAMP power point and poster presentation, and written report (wk 32-36).

Capacity, skill building, and career development components:

1) Learn and implement flow cytometry, molecular biology, and protein engineering techniques.
2) Weekly physician shadowing in outpatient HIV clinic and in-patient infectious diseases service.
3) Attend weekly immunology and virology seminar series.

Regulatory requirements for the project and plans for completing them:

All regulatory requirements, including IRB approval for the project have already been completed.

Expected Deliverables:

1) PowerPoint slide presentation summarizing project and findings
2) Poster summarizing project and findings for HVTN meeting and additional scientific conference
3) Results that will be incorporated into a future manuscript

Project Contact Person(s) (Name, Email):

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