(1) Submission ID#1525328

Functional antibody assays for assessing efficacy of gonococcal vaccine candidates

---

Author(s)

Mary Gray, n/a

Research Specialist

University of Virginia

Keena Thomas, n/a

Research Specialist

University of Virginia

Lacie Werner, PhD

Graduate Student

University of Virginia

Alison Criss, PhD

Professor

University of Virginia

Background

Drug-resistant Neisseria gonorrhoeae is an international priority for vaccine development. Antibodies are a correlate of protection for vaccine efficacy against Neisseria meningitidis, but their contribution to protection against N. gonorrhoeae remains unclear. Assays are needed to evaluate the ability of vaccine-elicited antibodies to bind the surface of various N. gonorrhoeae strains and promote serum bactericidal activity and opsonophagocytic killing.

Aim/Methods

Serum was collected from BALB/c mice that were immunized with gonorrhea vaccine candidates or appropriate negative controls (e.g. adjuvant-only). Candidates included the meningococcal 4C-MenB vaccine, which is being evaluated for cross-protection against gonorrhea. IgG and IgM binding to intact N. gonorrhoeae was measured by imaging flow cytometry. Serum bactericidal activity (SBA) was evaluated using immunoglobulin-depleted normal human serum as the complement source and measured by enumerating bacterial colony-forming units (CFU). Opsonophagocytic killing activity (OPKA) was measured using primary human neutrophils as the phagocyte and complement C6-depleted human serum as the complement source, and enumerated by CFU. Purified IgG and IgM antibodies were used as positive controls in each assay for inter-experimental consistency. Assays were conducted in 96-well plates to minimize reagent use and promote throughput.

Results

Imaging flow cytometry demonstrated increases in specific IgG and IgM antibody binding to the surface of N.

gonorrhoeae compared with isotype controls. SBA was dependent on active complement and specific antibody, and the presence of the antigen on the bacterial surface. The reduction in CFU measured in the OPKA assay was dependent on bacterial opsonization with C6-depleted serum, specific antibody, and the presence of neutrophils. Phagocytosis of N. gonorrhoeae in the OPKA assay was observed using imaging flow cytometry. 4C-MenB vaccination elicited antibodies that bound to N. gonorrhoeae and stimulated SBA and OPKA.

Conclusions

A suite of assays for evaluating antibody-dependent functional activities has been developed for N. gonorrhoeae. The correlation between results from these assays and the ability of vaccine candidates to protect mice from experimental gonorrhea are being evaluated in conjunction with projects in the Gonorrhea Vaccine Consortium. These assays are being extended to evaluate if and how 4C-MenB vaccination protects humans against experimental urethral gonorrhea.