

The Performance of Immunoassays to Measure Antibodies to the C Antigen Pgp3 in Different Epidemiological Settings for Trachoma

Sarah G. Minn,^{1*} Andre W. Nute,² Eshetu Sata,³ Zerihyn Tadesse,³ Ambahon Chernet,³ Mahteme Haile,⁴ Tade Zerew,⁴ Dana a Bethea,¹ Christian Laurent,¹ E. Kell Callahan,² Scott D. Nash,² and Diana L. Martin¹

¹Duke University Medical Center, Durham, NC, USA; ²Centers for Disease Control and Prevention, Atlanta, GA, USA; ³University of Gondar, Gondar, Ethiopia; ⁴Amhara Health Institute, Bahir Dar, Ethiopia

*Corresponding Author: Programs to eliminate trachoma as a public health problem, use prevalence of the clinical sign trachomatous inflammation—

participants or access to identifying information and were determined to be not engaged in human subjects research.

Study sites. Population-based prevalence impact and surveillance surveys were conducted in four districts (Alefa, Andabet, Dera, and Woreta Tona) in the Amhara region of Ethiopia as previously described.¹⁵ Briefly, a multistage cluster-random survey was conducted in all four districts. All individuals aged 1 year and older in the selected households were invited to participate in the survey. Dried blood spots from only 1 to 9-year-olds ($N = 2195$) were tested by each assessor described below. Of these individuals, 1,055 (48.1%)

as 0.899 (0.873–0.926). Samples with discordant results between the MBA and other assays fell closer to the MBA cut-off value for ELISA and LFA-late (median MFI-bg of 729 and 516, respectively) than LFA-gold (median MFI-bg of 7).

DISCUSSION

Population-based serological surveys can give an indication of exposure to a pathogen that can be modeled to estimate transmission in that population. We have been investigating whether and how antibody testing can provide

information about transmission of ocular Ct in children to help trachoma programs monitor endemic or pre-ious endemic populations.

Each test platform has advantages. MBA has good repro-

ELISA results here show good population-level agreement with MBA. Both of these tests also provide semi-quantitative data that may be useful for some analyses of antibody levels¹⁸ that the LFA does not provide. But the generally low cost, lack of instrument requirements, and ease of training and use make the LFA an appealing option for post-validation surveillance where funding may be scarce or nonexistent.

The current study shows optimization of a rapid lateral flow-based test (Pgp3-LFA) in response to population-level serological data that contrasted greatly with other tests for this sample set. These data—particularly the LFA-gold panel in Figure 2—highlight the subjectivity of tests using chromogenic readouts rated by a person. For this study, the technician documented if they considered bands to be faint, very faint, or even fainter on a worksheet during the study. Although these notations were outside of protocol, they proved helpful in understanding the issues at hand, as these samples were routinely negative by other tests and most likely represented overcalling of positive tests. We posited that this occurred

because pinning DBS on the LFA can lead to a pink smear. As a result, the red test line that appears with a colloidal gold developing reagent often needs to be differentiated from a light pink background. The test line for strongly positive specimens are easy to differentiate from the background, but for weakly positive specimens this can become challenging. For low-prevalence populations, the tendency to overcall tests

16. Arnold BF, Scobie HM, Priest JW, Lammie PJ, 2018. Integrated serologic surveillance of population immunity and disease transmission. *Epidemiol. Infect.* **146**: 1188–1194.
17. Wiegand RE, Cooley G, Goodhead B, Bannietts N, Kohlhoff S, Ginn S, Martin DL, 2018. Latent class modeling to compare testing platforms for detection of antibodies against the C-terminus of Plasmodium falciparum antigen Pgp3. *Syst. Res. Pharm.* **8**: 4232.
18. Arnold BF, van der Laan MJ, Hubbard AE, Steel C, Kibofcik J, Hamlin KL, Moss DM, Newman TB, Priest JW, Lammie PJ, 2017. Measuring changes in transmission of neglected tropical diseases, malaria, and enteric pathogens from quantitative antibody levels. *PLoS Negl. Trop. Dis.* **11**: e0005616.
19. Corran P, Coleman P, Riley E, Drakele C, 2007. Serology: a robust indicator of malaria transmission intensity. *PLoS Med.* **4**: 575–582.
20. Drakele C et al., 2005. Estimating medium- and long-term trends in malaria transmission by using serological markers of malaria exposure. *PLoS Med.* **2**: e119.