Dynamics of Helicobacter pylori-Specific Immunoglobulin G for 2 Years after Successful Eradication of Helicobacter pylori Infection in an American Indian and Alaska Native Population

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Helicobacter pylori antibodies were measured over 24 months in American Indian and Alaska Native persons who cleared their infections. Two months after treatment, 82% of H. pylori-negative persons remained seropositive. While there were declines in H. pylori antibodies for 12 months, after 24 months 71% of persons remained seropositive.

Helicobacter pylori eradication can be difficult, as reported rates of resistance to the commonly used antibiotics are 6% to 50% for clarithromycin and 12% to 61% for metronidazole (4). The eradication rates of resistance to the antibiotics are 6% to 12% to 16% for metronidazole (4, 5, 9). Tests to confirm treatment success, such as measuring H. pylori antibodies, are becoming clinically important. However, few serological studies have followed patients for longer than 12 months after eradication, and none have examined H. pylori antibodies over time in an Alaska Native (AN) or American Indian (AI) population. We measured H. pylori antibodies in AN/AI persons for 24 months after treatment. This study was approved by both the Centers for Disease Control and Prevention and Alaska Area Institutional Review Boards and the South Central Foundation Board of Directors.

H. pylori-infected persons were treated with an antibiotic regimen at the discretion of their medical providers. Blood was drawn and a urea breath test (UBT; Meretek Diagnostics, Inc., Nashville, TN) administered 2 months after treatment. Those who tested negative by UBT were enrolled in the 2-year follow-up study (7). Persons in the follow-up study were tested by UBT and had blood drawn 4, 6, 12, and 24 months after treatment. If a participant tested positive by UBT during follow-up, they were discontinued from the study.

Sera were tested for H. pylori-specific immunoglobulin G (IgG) by an in-house enzyme-linked immunosorbent assay (ELISA). This ELISA used 10 μg/ml of the high-molecular-weight cell-associated proteins described by Evans et al. as antigen (provided by Ezem, Inc., Westbury, NY) (2). Sera were diluted 1:200 and added to the plates. Antibodies were detected using alkaline phosphatase-labeled anti-human IgG (Sigma Chemical Co., St. Louis, MO) and p-nitrophenyl phosphate diluted in diethanolamine buffer (Kirkegaard & Perry Laboratories, Inc., Gaithersburg, MD). Optical density (OD) was measured at a 410-nm wavelength. To ensure assay reproducibility, a negative control serum and low- and highly positive control sera were tested on every plate (intra-assay variation, 9%, 5%, and 3%, respectively; interassay variation, 18%, 17%, and 11%, respectively).

Sera were positive for H. pylori-specific IgG if the OD was >0.5, negative if it was <0.3, and indeterminate if it was 0.3 to 0.5. We determined the cutoff values after repeated examination of 254 sera collected from Alaskan adults and children as part of a previous survey of H. pylori infection in Alaska (CDC, unpublished data). Using these cutoff values, the positive and negative predictive values of the ELISA optimized with sera from this unpublished survey were 89% and 93%.

Among 128 persons treated for an H. pylori infection who had sera available for testing, 90 (70%) eradicated their infection, as evidenced by a negative UBT. Among persons with H. pylori eradication, 79/90 (88%) had a decline in H. pylori-specific IgG between enrollment and 2 months after, compared to 22/38 (58%) persons who failed treatment. There was a decline in the mean H. pylori-specific IgG ODs in persons with H. pylori eradication but not in those without eradication, and persons with H. pylori eradication had smaller amounts of H. pylori antibody at 2 months than did those in whom H. pylori treatment failed (P < 0.0001). Despite the decline in H. pylori-specific IgG, 74 (82%) persons remained seropositive 2 months after eradication. Thus, the predictive value of 2-month positive serology identifying persons who failed treatment was only 30% (95% confidence interval [95% CI], 21 to 39%). The predictive value of 2-month negative serology identifying persons with successful treatment was 79% (95% CI, 57 to 100%).

A total of 104 AN/AI participants were enrolled in the follow-up study. There were declines in H. pylori-specific IgG between enrollment and 2 months, 2 and 4 months, 4 and 6 months, and 6 and 12 months (P < 0.0002 for all four time intervals). Further declines were not seen after 12 months (P = 0.29 [12 months versus 24 months]). For all participants, the mean H. pylori-specific IgG declined 43%, from 1.13 OD units (95% CI, 1.04 to 1.23) at enrollment to 0.64 OD units (95% CI, 0.55 to 0.73) 24 months after the start of treatment. The percentages of decline were similar regardless of age and H. pylori-specific IgG OD at enrollment (Fig. 1).

Two months after H. pylori treatment, 13% (11/86) of per-
sons whose *H. pylori* infection was eradicated converted from seropositive for *H. pylori* antibodies to seronegative; this increased to 29% (18/63) 2 years after treatment. No persons with large amounts of antibody (>1.5 OD units) at enrollment became seronegative over 24 months, compared to 92% of those with smaller amounts of antibody (<0.7 OD units) (Table 1).

This is the first study investigating *H. pylori* antibodies over time in an AI/AN population. We found that antibody dynamics posttreatment do not differ in this population compared with that in populations studied previously (3, 6, 8, 10). While the majority of studies followed patients for ≤12 months, our study followed participants for 24 months. In persons whose *H. pylori* infection was eradicated, we found a continuous decline in antibodies for 12 months, at which point the antibody decline ceased. We determined that less than one-third of persons became seronegative during the 24-month study period, similar to a study published by Cutler et al. that followed persons for >12 months (1).

The results of this study show that single IgG measurements should not be used to determine *H. pylori* treatment outcomes. In addition, antibodies remain circulating long after successful treatment, and therefore *H. pylori*-specific IgG antibodies should not be used to diagnose active infection in persons previously treated for an *H. pylori* infection. We have also been able to show that after treatment for an *H. pylori* infection, AI/AN persons have an antibody response that is similar to that for persons from other parts of the world.

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