Supplemental Methods

Stool NGS: Clean and inhibitor-free DNA including adequate *H. pylori* DNA from fecal samples appropriate for real-time PCR and NGS was obtained using the SuperFecal method developed by American Molecular Laboratories. Briefly, approximately 0.5 g of solid or semisolid fecal samples were homogenized with SuperFecal lysis buffer. After centrifugation, the supernatant was applied onto a SuperFecal column device and washed twice with SuperFecal wash buffers I and II. The DNA was then eluted from the column and DNA quality and quantity were measured using a Nanodrop spectrophotometer (Thermo Fisher Scientific). Purified fecal DNA was either kept at 4°C temporarily for immediate downstream analysis or stored at –20°C for longer-term storage. DNA mutations within the 6 H pylori genes associated with most cases of antibiotic resistance to the most common antibiotics used to treat the infection: 23S rRNA (clarithromycin), gyrA (fluoroquinolones), 16S rRNA (tetracycline), pbp1 (amoxicillin), rpoB (rifabutin), and rdxA (metronidazole) (5) were analyzed and determined by PyloriAR as described previously. (9)

Gastric biopsy culture: The selective *H. pylori* media used for isolation has a Brucella agar base with added vancomycin, trimethoprim, polymyxin B, and vitamin K1. Plates are incubated at 35 degrees C in a microaerophilic atmosphere. The agar dilution method is used for susceptibility testing. The antimicrobial is added to agar in various concentrations depending upon levels attainable in serum. A standardized suspension of the organism is applied to the agar plates that are incubated for 72 hours at 35 degrees C. Complete inhibition of all but 1 colony or a very fine residual haze represents the endpoint.