

Marked Disparity Between *Helicobacter pylori* Seroprevalence and Active Infection in Nigeria: Implications for Test-Result Interpretation and Clinical Decision-Making

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Somtochukwu Nebo¹, Christian Aneke Chinedu¹, Luke Ugwu Jideofor¹

¹Department of Medical Laboratory Science, Faculty of Allied Health Sciences, Enugu State University of Science and Technology, Nigeria

Correspondence: Somtochukwu Nebo, eduanke042@gmail.com

Abstract

Background: *Helicobacter pylori* infection remains an important global health problem and is particularly prevalent in Africa. However, estimates of infection prevalence vary substantially according to the diagnostic method used. Serological testing detects antibodies generated by exposure to *H. pylori* but may not reliably distinguish current from previous infection, whereas stool antigen testing provides evidence of current infection. This distinction is particularly important in high-prevalence settings where serological positivity may substantially exceed active-infection positivity. This study is aimed at comparing the prevalence and concordance of *H. pylori* serum antibody positivity, stool antigen positivity and faecal occult blood positivity among adults in Nigeria, and to examine the implications of discordant test results for interpretation and management of *H. pylori* infection.

Methods: A cross-sectional study was conducted among 223 adults. Each participant was tested for *H. pylori* serum antibody, stool *H. pylori* antigen and faecal occult blood. The prevalence of positivity for each marker was determined. Associations between categorical test results were assessed using the chi-square test, with statistical significance set at $p < 0.05$.

Results: Serum *H. pylori* antibody positivity was substantially higher than stool antigen positivity, occurring in 169/223 participants (75.8%) compared with 23/223 (10.3%), respectively. Faecal occult blood was positive in 53/223 participants (23.8%). All participants with positive stool antigen results were also antibody-positive, demonstrating complete overlap between stool antigen positivity and seropositivity in this study. Antibody positivity was significantly associated with stool antigen positivity ($\chi^2=6.789$, $p=0.009$). Similarly, antibody positivity was significantly associated with faecal occult blood positivity ($\chi^2=20.517$, $p<0.001$). Stool antigen positivity was strongly associated with faecal occult blood positivity ($\chi^2=32.574$, $p<0.001$), with 17 of the 23-stool antigen-positive participants (73.9%) also having occult

blood in stool.

Conclusions: The study demonstrates a marked disparity between *H. pylori* seropositivity and stool antigen positivity in this Nigerian population. The substantially higher seroprevalence should not be interpreted as equivalent to current active infection. The findings support careful selection and interpretation of *H. pylori* diagnostic methods in high-prevalence settings and reinforce the importance of active-infection tests when the clinical objective is to identify current infection. The observed association between stool antigen positivity and faecal occult blood also warrants further investigation into the relationship between active *H. pylori* infection and gastrointestinal mucosal bleeding.

Keywords: *Helicobacter Pylori*, Seroprevalence, Stool Antigen, Active Infection, Faecal Occult Blood, Diagnostic Testing, Nigeria, Concordance

1. INTRODUCTION

Helicobacter pylori is the most prevalent chronic bacterial infection globally, affecting approximately 50% of the world's population and up to 70.1% in Africa [1]. In Nigeria, prevalence estimates range from 85% to 87.7%, thus, making her one of the countries with the highest burden of *H. pylori* infection worldwide [2]. The infection is primarily transmitted through person-to-person contact, and it is believed to be acquired during childhood and persists for life, if not treated. The exact mechanisms of transmission are not fully understood, but several routes have been proposed, including Oral-Oral transmission, faecal-oral transmission, saliva, and gastrointestinal secretions. Chronic infection can lead to gastritis, peptic ulcer disease, mucosa-associated lymphoid tissue lymphoma, and gastric cancer [3].

In spite of the high prevalence of *H. pylori* infection, there is a striking discordance among its diagnostic methods. The gold standard remains invasive endoscopic biopsy with histology or culture. However, due to the challenges surrounding the endoscopic biopsy for either histology or culture, with regards to invasiveness and high cost, second alternatives such as the urea breath test (UBT) and stool antigen test are usually chosen as the first-line non-invasive diagnostic test methods for detecting *H. pylori* since they have shown high sensitivity and specificity for active infection. The rapid immunochromatographic stool antigen tests have been proven to show sensitivity >87% and specificity >93% when compared to UBT or histology, although antigen composition across communities and detection method used may result in outcome differences. Conversely, *H. pylori* testing using serum antibody method is strongly discouraged since it cannot differentiate between current and past infection. This is so because antibodies can remain in circulation for a long period of time after the antigen causing its production has long been killed. Unfortunately, reports indicate its continued usage in Nigeria, due to cost and accessibility of gold standard and alternative methods [4].

In addition, a report has shown that stool antigen detection has shown a direct relationship with active gastrointestinal bleeding, unlike positive antibody result, which shows cumulative exposure, rather than active infection [4][5]. Understanding these variations in the usefulness and limitations of these diagnostic methods in the laboratory diagnosis of *H. pylori* is of particular importance in sub-Saharan African communities where there are no standardized African guidelines equivalent to the Maastricht VI/Florence Consensus Report. The 2025 IARC Working Group Report emphasizes that population-based *H. pylori* screen-and-treat strategies should use accurate non-invasive tests tailored to local prevalence and healthcare capacity [6].

In Nigeria, where the use of serological antibody testing is common but stool antigen testing is poorly utilized, understanding the relationship between these methods is of immense importance in informing *H*

pylori screening policy for improved treatment outcome and maintaining antibiotic stewardship. This study is aimed at evaluating the prevalence and relationships among rapid stool antigen, serum antibody, and faecal occult blood tests in adults in Enugu State, Nigeria. The findings will provide evidence-based guidelines and result interpretations for non-invasive screening strategies for *H. pylori* infection in resource-limited settings.

2. MATERIALS AND METHODS

2.1 Study Area

This study was conducted in Abakpa Nike, a sub-urban city of Enugu State. Enugu state is located in the South-Eastern geopolitical zone of Nigeria. Enugu State lies approximately between latitudes 5°56'N and 7°06'N and longitudes 6°53'E and 7°55'E. The state experiences a tropical rainforest climate characterised by two major seasons: the rainy season (April–October) and the dry season (November–March). Average annual temperatures range between 22°C and 31°C. Abakpa sub-urban was chosen for this study because of its settlement advantages and geographical location: standard of living is relatively cheaper for middle income earners, and it shares boundaries with all the three senatorial districts that make up Enugu State (Enugu North, Enugu West, and Enugu East).

The Abakpa district is thickly populated, with many households living in overcrowded conditions and relying primarily on untreated borehole water as their major source of domestic water supply. Although most of them have access to water-system-based toilet facilities, open defecation is still practiced by some residents, particularly a few of those living close to streams and rivers. They either defecate directly or evacuate their feces into these water bodies. Of particular concern is that some commercial vegetable vendors use water from these contaminated sources to wash vegetables and fruits or to rinse and clean bitter leaves before sale. These collectively, make Abakpa a good public health catchment area for this study.

2.2 Study Design and Setting

This research employed a cross-sectional descriptive study design, and was conducted from February 7, 2026 to June 18, 2026, in Enugu State, Nigeria. Participants consisted of randomly selected 223 individuals (185 females and 38 males), aged 18-65 years, residing in Abakpa, and who were not on antibiotics, proton pump inhibitors, and bismuth preparations, at least 4 weeks before testing. Also included were those who do not have history of gastrointestinal bleeding or haemorrhoids, and women who were on menstrual flow. Excluded in the study were those who did not meet up with the inclusion criteria. Samples of participants were collected and tested serologically for *H pylori* stool antigen, *H pylori* serum antibody, and Stool occult blood. Participants recruitment, preparation, sample collection, testing procedures, data handling, analysis and reporting were done in accordance with provisional standards.

2.3 Ethical Approval

Ethical Approval for the study was obtained from the Research and Ethics committee of the Enugu State Ministry of Health. Research details were made known to the participants, and their informed consents were obtained before enrolment.

2.4 Sample Collection

Participants were recruited and advised to abstain from taking NSAIDs for 4 weeks before test, and red meat/vitamin C supplements, for 7 days before test. Labelled stool sample bottles were shared to participants and were advised to note down their numbers. Furthermore, they were trained on the processes of stool sample collection as follows: For solid stool samples, participants were advised to collect a small

sample (~50 mg or a 1/4 pea size) from 3 different interesting sites on the faeces, using the applicator stick provided, and transfer into the sample bottle provided. For liquid stool, they were advised to gently mix the stool sample, and aspirate using a dropper provided, and transfer roughly 80 µL (or 2 drops) into the sample bottle.

On the blood sample collection day, participants were asked to collect stool as instructed, and return it to my team, avoiding contamination with urine, water, and other materials. Participants who returned their stool samples were interviewed for their sociodemographic information, after which their blood samples were collected. The collected blood samples were tied together with the stool samples, and sent within two hours of collection to the Medical Microbiology Laboratory of Enugu State University of Science and Technology Teaching Hospital for laboratory testing. Results were collated, tabulated, and analysed using statistical science for social sciences (SPSS) version 27, and the final report was made afterwards.

2.5 *H. pylori* Stool Antigen Detection

Stool antigen testing was performed using the Abbott Clearview *H. pylori* Antigen Test Device (Abbott Laboratories, Chicago, IL, USA), a rapid qualitative immunochromatographic assay based on monoclonal antibodies. The test device and the faecal sample were brought to room temperature [15–30°C] before testing. Approximately 50 mg of solid stool or 80 µL of liquid stool was collected, using the provided applicator and transferred into the sample buffer. The mixture was shaken vigorously to ensure homogenization. Three drops [80–120 µL] of the extracted specimen were applied to the sample well of the test device. The mixture migrates chromatographically along the length of the test strip contained within the device and interacts with the reagents on the strip. Results were read and interpreted within 10–15 minutes. A positive result was indicated by two coloured lines appearing in both the test and control regions. A negative result was indicated by a single-coloured line in the control region only. Absence of a control line rendered the test invalid.

2.6 *H. pylori* Serum Antibody Detection

Serum antibody testing was performed using the Abbott Clearview *H. pylori* Antibody Test Device [Abbott Laboratories, Chicago, IL, USA], a rapid qualitative immunochromatographic assay for the detection of IgG, IgM, and IgA antibodies to *H. pylori* in human serum or plasma. Venous blood was collected aseptically and serum was separated within 2 hours of collection to avoid haemolysis. The test device and serum sample were brought to room temperature before use. Approximately 10–75 µL of serum was transferred to the sample well of the test device using the provided dropper. The serum migrates chromatographically along the length of the test strip contained within the device and interacts with the reagents on the strip. Results were read and interpreted within 10–15 minutes. A positive result was indicated by two coloured lines appearing in both the test and control regions. A negative result was indicated by a single-coloured line in the control region only. Absence of a control line rendered the test invalid.

2.7 Faecal Occult Blood Testing

Faecal occult blood testing was performed using the AccuQuik Faecal Occult Test Kit (AdvaCare Pharma USA), a rapid qualitative immunochromatographic assay. The test device and faecal blood specimen were brought to room temperature [15–30°C] before testing. Approximately 50 mg of solid stool or 80 µL of liquid stool was collected, using the provided applicator and transferred into the sample buffer. The mixture was shaken vigorously to ensure homogenization. Three drops [80–120 µL] of the homogenized mixture were applied to the sample well of the test device. The mixture migrates chromatographically along the length of the test strip contained within the device and interacts with the reagents on the strip.

Results were read and interpreted within 3-5 minutes. A positive result was indicated by two coloured lines appearing in both the test and control regions. A negative result was indicated by a single-coloured line in the control region only. Absence of a control line rendered the test invalid.

Note:

The reaction with intact red cells probably from lower GIT bleeding occurs very slowly while the reaction with digested blood (as in ulcer conditions) is rapid. Therefore, the result of this test was read within 3-5 minutes.

2.8 Statistical Analysis

Data were analysed using descriptive and inferential statistics. Categorical variables were summarized using frequencies and percentages. Association/relationships between variables were examined using Pearson's chi-square test. Statistical significance was defined as: $p < 0.05$.

3. RESULTS

3.1 Participant Characteristics

A total of 223 participants aged 18-65 were enrolled, 185 females (83.0%) and 38 males (17.0%). The mean age was approximately 38.7 ± 13.9 years, with a median age of 38 years.

Table 1.0 Overall prevalence of *H. pylori* antibody, stool antigen and faecal occult blood positivity

Marker	Positive, n (%)	Negative, n (%)	Total
Serum <i>H. pylori</i> antibody	169 (75.8)	54 (24.2)	223
Stool <i>H. pylori</i> antigen	23 (10.3)	200 (89.7)	223
Faecal occult blood	53 (23.8)	170 (76.2)	223

Table 1 shows that serum antibody positivity was substantially higher than stool antigen positivity. Approximately three-quarters of participants were seropositive, whereas only about one in ten had detectable stool *H. pylori* antigen. Faecal occult blood was detected in approximately one-quarter of participants.

Table 2. Cross-tabulation of serum antibody and stool antigen results

Serum Antibody	Positive Stool Antigen	Negative Stool Antigen	Total
Positive	23	146	169
Negative	0	54	54
Total	23	200	223

Table 2 above shows that in every stool antigen-positive participant was also antibody-positive, while most antibody-positive participants were stool antigen-negative. The association between the two tests was statistically significant ($\chi^2=6.789$, $p=0.009$), but their positivity rates differed markedly.

Tables showing relationship between *H. pylori* markers and faecal occult blood

Table 3.1 Relationship between stool antigen and faecal occult blood

Stool Antigen	Positive Occult Blood	Negative Occult Blood	Total
Positive	17	6	23
Negative	36	164	200

Total 53 170 223

Table 3.1 shows that faecal occult blood was considerably more common among participants with positive stool antigen results. The association between stool antigen and occult blood was statistically significant ($\chi^2=32.574$, $p<0.001$).

Table 3.2. Association between serum *H. pylori* antibody and faecal occult blood status

Serum antibody status	Occult blood positive, n (%)	Occult blood negative, n (%)	Total, n (%)
Positive	53 (23.8)	116 (52.0)	169 (75.8)
Negative	0 (0.0)	54 (24.2)	54 (24.2)
Total	53 (23.8)	170 (76.2)	223 (100.0)

Statistical test: $\chi^2 = 20.517$, $df = 1$, $p < 0.001$.

Table 3.2 shows that all 53 participants with positive faecal occult blood results were also positive for serum *H. pylori* antibodies (53/53, 100%). Conversely, 116 of the 169 antibody-positive participants (68.6%) had negative occult blood results. The association between serum antibody status and faecal occult blood was statistically significant ($\chi^2 = 20.517$, $p < 0.001$).

4. DISCUSSION

The major findings of this study was the striking discordance identified, between serum *Helicobacter pylori* antibody positivity and stool antigen positivity. Among the 223 participants investigated, 169 (75.8%) were positive for serum antibodies, whereas only 23 (10.3%) were positive for stool antigen, representing an absolute difference of 65.5 % points. This difference highlights important variations between evidence of previous exposure to *H. pylori* and evidence of detectable infection at the time of testing. Thus, the result of *H. pylori* prevalence depends on the diagnostic method employed, and this should be considered while interpreting such results.

The high positivity observed in this study is consistent with established data on the high burden of *H. pylori* reported in Africa. Previous and current studies have reported high seroprevalence among African adults, including Nigerian populations, making *H. pylori* infection a major public health concern across Africa [4][7][8]. A study done in Nigeria, by Adeniyi et al. reported seropositivity rates of 88% among dyspeptic patients and 80% among healthy adults, supporting the serological evidence of high prevalence of *H. pylori* exposure in Nigerian populations [9]. Other studies conducted in Africa substantiate the fact that *H. pylori* prevalence depends basically on the population studied and the diagnostic method used. For instance, a systematic review from East Africa reported a pooled prevalence of 60.79% when serology was used compared with 34.64% when stool antigen testing was used [10]. These reports are in agreement with the findings of this study, although a little variation in the number of stool antigen positivity was observed, which could be as a result of the sensitivity of test kits used, differences in population, or level of infection among the participants. These findings further reinforce the importance of diagnostic methods when interpreting *H. pylori* test results.

The observed variations between antibody and stool antigen positivity in this study is a possibility because the two tests measure different aspects of *H. pylori* infection. Serum antibody testing detects the host immune response to the organism, whereas stool antigen testing detects bacterial antigen shed into the gastrointestinal tract and therefore provides evidence more closely related to the presence of infection at the time of testing. Current international consensus guidance recognizes that *H. pylori*-specific IgG serology cannot reliably distinguish active infection from previous infection and is consequently unsuitable

for determining eradication success [11]. In contrast, validated monoclonal stool antigen assays are considered appropriate non-invasive tests for detecting infection and confirming eradication [11]. The present findings therefore provide an important epidemiological caution: serological prevalence should not automatically be interpreted as the prevalence of active *H. pylori* infection.

Furthermore, an additional finding of interest was the relationship between *H. pylori* markers and faecal occult blood. This study identified that all the 53 participants with positive faecal occult blood results were also serum antibody-positive, producing a statistically significant association between antibody status and occult blood positivity ($\chi^2=20.517$, $p<0.001$). More importantly, 17 of the 23-stool antigen-positive participants (73.9%) were also positive for faecal occult blood, and the association between stool antigen and occult blood was statistically significant. The latter finding is particularly noteworthy because it links evidence of detectable *H. pylori* antigen with evidence of gastrointestinal blood loss. *H. pylori* is strongly associated with gastritis and peptic ulcer disease, and infection has an established relationship with ulcer-related gastrointestinal bleeding [12]. However, Faecal occult blood is a non-specific marker of gastrointestinal bleeding and cannot establish either the anatomical source or the cause of blood loss. Positive results can come from peptic ulcer disease, mucosal inflammation or other gastrointestinal lesions, and *H. pylori* may represent only one of several possible contributing factors. This fact is particularly important in interpreting faecal occult blood result, in relationship with *H. pylori* infection, since other factors different from *H. pylori* can give positive faecal occult blood.

An important strength of this study is that all three investigations were performed in the same participants. That gave the opportunity for direct within-person comparison of serological evidence, stool antigen detection and faecal occult blood rather than comparing prevalence estimates obtained from different populations. The study therefore provides a clear demonstration of the extent to which apparently high *H. pylori* prevalence can differ according to the diagnostic approach used. The Nigerian setting also adds value because locally generated evidence addressing the interpretation of different *H. pylori* diagnostic approaches remain relatively limited, despite the high burden of infection reported in the country and across Africa [13][14].

Limitations of the study

The study had several limitations. The study did not include an independent reference standard such as histological examination, culture, rapid urease testing, urea breath testing or molecular detection. Therefore, the present findings should not be interpreted as a formal diagnostic-accuracy comparison, and sensitivity, specificity, positive predictive value or negative predictive value cannot be reliably assigned to either antibody or stool antigen testing. Again, faecal occult blood is non-specific and cannot be attributed to *H. pylori* without further clinical, histological, or endoscopic evaluations.

CONCLUSION

In conclusion, this study demonstrates a striking discordance between serum *H. pylori* antibody positivity and stool antigen positivity in a Nigerian adult population. These findings indicate that serological positivity and evidence of current detectable infection should not be treated as interchangeable measures of *H. pylori* prevalence. In settings where the clinical objective is to identify current infection, greater emphasis should be placed on validated tests that detect active infection.

Recommendation

Future Nigerian studies incorporating urea breath testing, histology, rapid urease testing, culture and/or molecular detection would provide a stronger reference framework for determining the true prevalence of active infection and for clarifying the biological and diagnostic basis of the observed serological–antigen

discordance.

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