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The Influence of HIV Status on the Burden and Clinical Manifestations of Gastrointestinal Pathogens in Yangon, Myanmar

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Abstract. The impact of HIV infection on the burden of gastrointestinal pathogens in Myanmar is poorly defined. Stools of 103 HIV-infected and 105 HIV-uninfected adult outpatients at a tertiary referral hospital in Yangon were examined microscopically. Stool antigen tests for *Helicobacter pylori* infection were positive in 63/103 (61%) HIV-infected and 61/105 (58%) HIV-uninfected patients ($P = 0.65$). Soil-transmitted helminth infections were much less common, occurring in 9/103 (9%) HIV-infected and 13/103 (13%) HIV-uninfected patients ($P = 0.50$). One HIV-uninfected patient had *Giardia duodenalis*, but there were no cases of *Strongyloides stercoralis*, *Entamoeba histolytica*, *Capillaria philippinensis*, *Isospora*, *Cyclospora*, or *Schistosoma* infection in the entire cohort. Despite the high prevalence of *H. pylori*, only 1/208 (0.5%) had ever received eradication, compared with 159/208 (76%) who had ever been dewormed. *Helicobacter pylori* appears to be an underappreciated pathogen in Myanmar. Its strong association with gastric cancer and peptic ulcer disease necessitates a more aggressive approach to its management.

The few published studies to examine the burden of gastrointestinal parasites in Myanmar report a prevalence that is one of the highest in Asia.[1,2] HIV is also an important infection in the country, affecting an estimated 0.8% (0.6–0.9%) of adults aged 15–49 years.[3] HIV-infected patients in Myanmar frequently present with advanced disease, at which time, the main aim is to initiate life-saving antiretroviral therapy (ART) and to identify life-threatening conditions such as tuberculosis; gastrointestinal pathogens are seen as less of a priority.[4]

However, gastrointestinal parasites are common in HIV-infected patients, and regular screening is an important component of HIV management,[5,6] especially as the morbidity—and even mortality—from these infections is underestimated.[7] There are even data to suggest that antihelminthic therapy may have a favorable effect on HIV progression.[8]

The clinical significance of another important gastrointestinal pathogen in Myanmar—*Helicobacter pylori*—is enormous. The infection's local prevalence approaches 70%,[9] and the country's age-standardized death rate for gastric cancer is 11.1/100,000/year, while that of peptic ulcer disease is 7.1/100,000/year. This compares with a rate of 17.4/100,000/year for HIV infection.[10] There are conflicting data about the impact of HIV infection on *H. pylori* colonization,[11] but it is important to identify any interaction in a country such as Myanmar where both infections are common.

This study determined the prevalence of gastrointestinal parasites and *H. pylori* infection in outpatients at a tertiary referral hospital in Yangon, Myanmar's largest city. It aimed to establish whether a patient's HIV status had any impact on the prevalence of these infections or the symptoms that they caused.

This cross-sectional study was performed at Insein General Hospital, a 500-bed teaching hospital between March 1 and May 31, 2019. Adults (aged ≥ 18 years) attending the hospital's ART and general medical outpatient department for any reason were eligible for inclusion. With the study's limited budget, it was planned that data from 100 HIV-infected and 100 unmatched HIV-uninfected patients would be evaluated. Written consent was obtained before enrollment. The only exclusion criterion was the use of antibiotic therapy in the prior 4 weeks which would have vitiated the results of the *H. pylori* stool antigen testing (SAT). Participants completed a dedicated pro forma addressing their demographics, socioeconomic status, and their access to water, sanitation, and hygiene to determine the relative contributions of these factors to the prevalence of gastrointestinal pathogens. The pro forma also asked about current gastrointestinal symptoms, a history of *H. pylori*–related disease, and any prior deworming or *H. pylori* therapy. All patients had their hemoglobin level and mean corpuscular volume (MCV) determined; HIV-infected patients had their most recent CD4 T-cell count recorded and documentation as to whether–or not–they were receiving ART.

The participants were provided with a sealed stool container and asked to provide a specimen. The fresh specimen was tested within hours using a monoclonal antibody–based SAT (BIONEXIA® *H. pylori* Ag, BioMérieux, Marcy-l'Étoile, France; reported sensitivity: 86.1% [95% CI: 71.3–93.9%] and specificity: 93.7% [95% CI: 86.9–97.1%])[12] by study doctors at the hospital. If participants tested positive, they were treated—according to the hospital policy—with sequential combination therapy of rabeprazole, amoxycillin, clarithromycin, and tinidazole. If repeat SAT was positive, they were offered second-line levofloxacin-based therapy.

Stools were also transported to the microbiology laboratory where experienced microbiologists examined the specimen for the presence of intestinal parasites on the same day. Stools were examined for hookworm, *Ascaris lumbricoides*, *Trichuris trichiura*, *Strongyloides stercoralis*, *Giardia duodenalis*, *Entamoeba histolytica*, *Capillaria philippinensis*, and *Cryptosporidium*, *Microsporidium*, *Isospora*, *Cyclospora*, and *Schistosoma* species using the formalin–ether concentration technique

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‡ Professor David Cooper has sadly passed away during the preparation of this report, but contributed significantly to the work.

and modified Ziehl–Neelsen staining, where appropriate. All positive results and 10% of the negative results were reviewed by a senior microbiologist (W. P. P. A.). All microbiologists were blinded to the HIV status of the patient. Positive stool results were reported to the study doctors who prescribed any necessary treatment.

Data were de-identified, entered into an electronic database, and analyzed with statistical software (Stata 14, StataCorp., College Station, TX). Groups were analyzed using the Kruskal–Wallis and chi-squared tests, where appropriate. The Human Research Ethics Committee of the University of Medicine 2, Myanmar (24/ERC-1 [10–2017]), provided ethical approval for the study.

The study enrolled 208 patients; 142 (68%) lived in a city, most commonly in Yangon (98/208, 47%), while 66 (32%) lived in a rural area, most commonly the Ayeyarwady delta region (51/208, 25%).

There were 105 HIV-uninfected patients and 103 HIV-infected patients, 87 (85%) of whom were receiving ART. The HIV-infected patients had a median (interquartile range) CD4 T-cell count of 514 (372–712) cells/mm^3, 7/103 (7%) had a CD4 T-cell count < 200 cells/mm^3. Patients with HIV infection were younger and had a lower body mass index than HIV-uninfected patients, but did not have a higher prevalence of gastrointestinal pathogens. Indeed, gastrointestinal symptoms were less common among the HIV-infected patients (Table 1).

There were 22/208 (11%) patients in the cohort with evidence of soil-transmitted helminth (STH) infection; of these, 14 (7%) were infected with *T. trichiura*, 5 (2%) with hookworm, and 3 (1%) with *A. lumbricoides*; one patient was infected with both *T. trichiura* and *A. lumbricoides* (Figure 1). HIV-infected patients were no more likely to have STH infection than HIV-uninfected patients (9/103 [9%] versus 13/103 [13%], $P = 0.50$), although they were more likely to have been dewormed previously (88/103 [85%] versus 71/105 [68%], $P = 0.002$).

TABLE 1

Demographic, socioeconomic, clinical, and laboratory characteristics of the patients, stratified by HIV status

Variable	HIV infected ($n = 103$)	HIV uninfected* ($n = 105$)	P-value
Demographic characteristics			
Age (years)	41 (35–49)	58 (44–68)	0.0001
Male gender, *n* (%)	44 (43)	37 (35)	0.27
Rural residence, *n* (%)	35 (34%)	31 (30%)	0.52
Socioeconomic characteristics			
Number of people presently living in household	4 (3–5)	4 (3–6)	0.03
Number of siblings	5 (3–7)	5 (4–7)	0.68
Employed†	54/101 (53%)	28/56 (50%)	0.68
Completed high school	42 (51%)	40 (49%)	0.70
Private flushing toilet	21 (20%)	13 (12%)	0.12
Drinks purified drinking water	72 (70%)	66 (63%)	0.28
Municipal water supply	26 (25%)	29 (28%)	0.70
Water from underground aquifer (well)	75 (73%)	71 (68%)	0.41
Household income > 300,000MMK/month‡	27 (26%)	26 (25%)	0.81
Relevant medical history			
Ever dewormed	88 (85%)	71 (68%)	0.002
Dewormed last year	28 (27%)	17 (16%)	0.054
Prior *Helicobacter* eradication	1 (1%)	0 (0%)	0.50
Active peptic ulcer disease	0	0	–
Past peptic ulcer disease	1 (1%)	2 (2%)	1
Past or present stomach cancer	0	0	–
Family history of stomach cancer	0 (0%)	3 (3%)	0.25
On antiretroviral therapy	87 (84%)	–	–
Current clinical findings			
Current dyspepsia	21 (20%)	42 (40%)	0.002
Current abdominal pain	11 (11%)	18 (17%)	0.18
Current diarrhea	4 (4%)	5 (5%)	1
Current melena	0	1	1
Any gastrointestinal symptoms currently	30 (29%)	49 (47%)	0.009
Body mass index (kg/m^2)	21.2 (18.9–24.1)	24.2 (21.4–27.6)	0.0001
Laboratory findings			
Helicobacter pylori positive	63 (61%)	61 (58%)	0.65
Any parasites	53 (51%)	60 (57%)	0.41
Number of different parasites	1 (0–1)	1 (0–1)	0.65
Ascaris lumbricoides	2 (2%)	2/103 (2%)	1
Trichuris trichiura	6 (6%)	8/103 (8%)	0.78
Hookworm	2 (2%)	3/103 (3%)	1
All soil-transmitted helminths	9 (9%)	13/103 (13%)	0.50
Microsporidium	22 (21%)	15 (14%)	0.18
Cryptosporidium	41 (40%)	48 (46%)	0.39
Hemoglobin (g/dL)	12.5 (11.5–13.7)	12.4 (11.5–13.5)	0.36
Mean corpuscular volume (fL)	86 (79–94)	84 (77–87)	0.0001
CD4 count (cells/mm^3)	514 (372–712)	–	–

Numbers represent absolute number (%) or median (interquartile range).

* In two HIV-uninfected patients, there was insufficient stool to perform an adequate examination for parasites.

† Only including those aged < 60 years.

‡ Median household income in Yangon is 300,000 Myanmar Kyat (approximately USD200)/month.

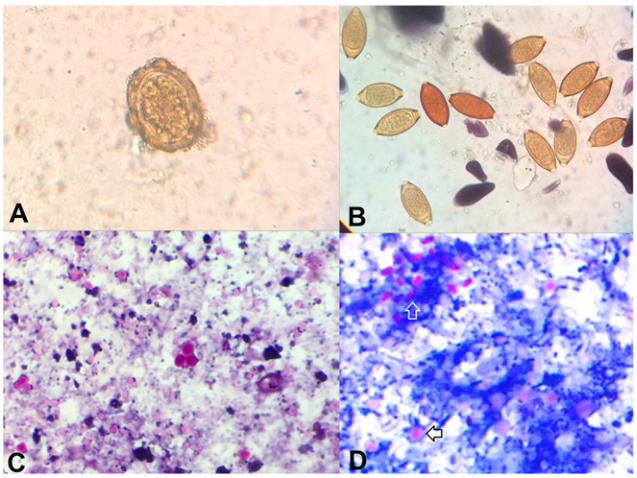

FIGURE 1. Stool microscopy findings in four of the patients. (**A**) *Ascaris lumbricoides* ovum. (**B**) Ova of *Trichuris trichiura.* (**C**) Oocysts of *Cryptosporidium* species. (**D**) Oocysts of *Cryptosporidium* (black arrow) and *Microsporidium* species (white arrow). This figure appears in color at www.ajtmh.org.

Helicobacter pylori infection was more common in the cohort with 124/208 (60%), having a positive SAT. But HIV-infected patients were no more likely to have a positive SAT than HIV-uninfected patients (63/103 [61%] versus 61/105 [58%], $P = 0.65$). Similarly, there was no difference in the demographics, socioeconomic indicators, symptoms, or laboratory findings of the *H. pylori*–positive patients compared with those who were negative.

Cryptosporidium oocysts were seen with acid-fast staining in the stools of 89/208 (43%), while *Microsporidia* oocysts were seen in 37/208 (18%) (Figure 1). However, most of these patients were asymptomatic and were no significant differences between patients who tested positive for these organisms and those who did not; most notably, there was no difference in HIV status (Supplemental Table 1). There was a single case of *G. duodenalis* in an HIV-uninfected patient. *Strongyloides stercoralis*, *E. histolytica*, *C. philippinensis*, and *Isospora*, *Cyclospora*, and *Schistosoma* species were not identified in any patients.

Despite the study's resource-limited, tropical setting, the prevalence of STH infection was relatively low. More than 76% of the cohort had previously taken deworming therapy (including 22% in the past year), suggesting that local awareness of these pathogens is high. In contrast, the prevalence and clinical significance of *H. pylori* infection appear underappreciated: Almost 60% of this cohort's unselected outpatients tested positive for *H. pylori*, which given the imperfect sensitivity of SAT is likely to underestimate the infection's true burden. However, only one patient in the cohort had ever received *H. pylori* eradication.

TABLE 2

Demographic, socioeconomic, clinical, and laboratory characteristics of the patients, stratified by infection with soil-transmitted helminths and *H. pylori*

Variable	STH positive ($n = 22$)	STH negative ($n = 184$)	*P*-value	*H. pylori* positive ($n = 124$)	*H. pylori* negative ($n = 84$)	*P*-value
Demographics						
Age (years)	48 (33–57)	48 (38–60)	0.35	47 (36–57)	50 (38–66)	0.15
Male gender, *n* (%)	6 (27%)	75 (41%)	0.22	38 (35%)	38 (45%)	0.13
Rural residence	11 (50%)	55 (30%)	0.06	41 (33%)	25 (30%)	0.66
Socioeconomic characteristics						
Number of people presently living in household	4 (3–5)	4 (3–6)	0.37	4 (3–5)	4 (2–5)	0.16
Number of siblings	5 (4–6)	5 (4–7)	0.46	5 (4–7)	5 (3–7)	0.92
Employed*	14/18 (78%)	67/138 (49%)	0.02	50/98 (51%)	32/59 (54%)	0.70
Completed high school	5 (23%)	77 (42%)	0.08	51 (41%)	31 (37%)	0.54
Private flushing toilet	3 (14%)	30 (16%)	0.75	18 (15%)	16 (19%)	0.38
Drinks purified drinking water	12 (55%)	124 (67%)	0.23	81 (65%)	57 (68%)	0.70
Municipal water	5 (23%)	49 (27%)	0.69	30 (24%)	25 (30%)	0.37
Water from underground aquifer (well)	15 (68%)	130 (71%)	0.81	90 (73%)	56 (67%)	0.36
Household income > 300,000MMK/month†	4 (18%)	48 (26%)	0.42	35 (28%)	18 (21%)	0.27
Relevant medical history						
Ever dewormed	18 (82%)	141 (77%)	0.58	94 (76%)	65 (77%)	0.79
Dewormed in last year	6 (27%)	39 (21%)	0.51	31 (25%)	14 (17%)	0.15
Prior *H. pylori* eradication	0	1 (0.5%)	1	1	0	1
On antiretroviral therapy	8/9 (89%)	79/94 (84%)	1	54/63 (86%)	33/40 (83%)	0.66
Current clinical findings						
Current dyspepsia	8 (36%)	53 (29%)	0.46	33 (27%)	30 (36%)	0.16
Current abdominal pain	2 (9%)	27 (15%)	0.48	14 (11%)	15 (18%)	0.18
Current diarrhea	0	9 (5%)	0.29	5 (4%)	4 (5%)	0.80
Current melena	0	1 (1%)	1	0	1	0.40
Any gastrointestinal symptoms currently	9 (41%)	68 (37%)	0.72	42 (34%)	37 (44%)	0.14
Body mass index (kg/m^2)	21.8 (20.0–24.2)	22.7 (19.8–26.3)	0.35	23.1 (19.9–27.6)	22.1 (20.0–24.7)	0.16
Laboratory findings						
Hemoglobin (g/dL)	12.1 (10.3–12.9)	12.7 (11.5–13.6)	0.08	12.4 (11.5–13.5)	12.6 (11.5–13.7)	0.76
Mean corpuscular volume (fL)	82 (74–87)	85 (78–90)	0.14	84 (79–89)	86 (78–90)	0.40
HIV positive	9 (41%)	94 (51%)	0.50	63 (51%)	40 (48%)	0.65
CD4 count (cells/mm^3)	567 (427–902)	508 (369–709)	0.30	549 (403–781)	493 (288–650)	0.06

H. pylori = *Helicobacter pylori*; STH = soil-transmitted helminth. Numbers represent the absolute number (%) or median (interquartile range).
* Only including those aged < 60 years.
† Median household income in Yangon is 300,000 Myanmar Kyat (approximately USD200)/month.

Myanmar has the highest burden of *H. pylori* in Southeast Asia, with one recent study reporting a prevalence of 69%.[9] Meanwhile, gastric cancer is the country's sixth most common cancer and has a high attributable mortality.[10,13] *Helicobacter pylori* is simple to treat, and the risk of recurrence is relatively low.[14] Indeed, a recent study performed in Yangon showed that local first-line eradication regimens had a per-protocol efficacy that approached 95%.[15] However, the fact that only a single patient in the entire cohort had been previously treated for *H. pylori* suggests that local clinicians are not yet managing the infection as aggressively as they might.

The high prevalence of *Cryptosporidium* infection was surprising. Equally notable was the fact that there was no difference in the prevalence of the *Cryptosporidium* between immunocompetent and immunocompromised populations. The absence of symptoms in so many of the patients with *Cryptosporidium* ova seen in their stools was also noteworthy. Asymptomatic carriage of *Cryptosporidium* is certainly described,[16] although not at the rates seen in this series. It is hypothesized that some genotypes of *Cryptosporidium* may be less pathogenic and, thus, seen more commonly in asymptomatic patients.[17] However, the absence of molecular testing in the resource-poor setting of the study precluded us from either testing this hypothesis or from confirming or speciating the *Cryptosporidium* cases diagnosed with modified Ziehl–Neelsen staining. Microsporidia were also common, although again the prevalence was not higher in the HIV-infected patients and most patients were asymptomatic.

The study has many limitations. Gastrointestinal parasites were diagnosed using stool microscopy. Molecular techniques have increased sensitivity and may have assisted with speciation of the *Cryptosporidium* and *Microsporidium* cases.[7,17,18] Only adults attending the outpatients department at a single tertiary referral center were enrolled in the study. Although most of the cohort lived outside Yangon—more than 30% in a rural location—the results from this small sample are not necessarily representative of the country. Children—particularly in remote locations—are likely to have a higher burden of STH infection.[2] As antibiotic use in the prior 4 weeks is a contraindication to the use of SAT, this effectively excluded all HIV-infected patients with a CD4 T-cell count of < 350 cells/mm^3, a population for whom co-trimoxazole is recommended in Myanmar. This led to enrollment of patients with less immunodeficiency, tending to minimize the difference in the array of pathogens identified in the HIV-infected and HIV-uninfected participants. This also limits the generalizability of our findings to the wider local HIV-infected population.[4] The relatively small sample increases the likelihood of type 2 errors. Future studies should define the relationship between the gastrointestinal pathogens and clinically meaningful endpoints. They should also be powered to determine optimal strategies to identify and treat the infections in a timely and cost-effective manner.

Despite this, the findings do offer some insights into the present burden of gastrointestinal pathogens in Myanmar. There appears to be good local awareness of gastrointestinal parasites: most patients had had received deworming therapy previously, many within the previous 12 months. However, *H. pylori*, an infection with more significant health implications, remains relatively neglected in Myanmar, and indeed in low- and middle-income countries generally.[19] Although there has been progress against many infectious diseases at the national level in Myanmar,[20] *H. pylori* has not yet been targeted. These data suggest it is now time to redress this issue.

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Note: Supplemental table appears at www.ajtmh.org.

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