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Characterization of urinary *Escherichia coli* isolates in HIV-seropositive women with asymptomatic bacteriuria in Western Ghana

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Abstract

Background Asymptomatic bacteriuria (ASB), a precursor of urinary tract infection (UTI), is highly prevalent among people living with HIV (PLHIV). In this study, we characterized urinary *Escherichia coli* isolated from PLHIV with asymptomatic bacteriuria.

Methods Urine samples from women living with HIV (WLHIV) were cultured, and bacteria isolates were identified using MALDI TOF biotyper. Antimicrobial susceptibility test was performed on isolates and genes encoding virulence factors and integrases were screened using Multiplex PCR and gel imaging. Patients' predictor variables like WHO disease staging, Highly Active Antiretroviral Therapy (HAART) duration and viral load were extracted from patient folders using a structured questionnaire.

Results The mean age of our 400 participants was 43.5 ± 12 years, 78.5% had viral loads < 1000 copies/ml, 53.8% had been on HAART for 1–5 years and 62.5% were classified as having WHO stage III disease. 85 of 400 (21.2%) urine samples cultured positive, and the most prevalent organism was *E. coli* (69.4%, 59/85), all being multidrug-resistant. None of the patients' predictor variables reached statistical significance; WHO disease stage (OR = 0.953, 95%CI = 0.683–1.330, $p = 0.777$), age (OR = 0.1.207, 95%CI = 0.971–1.499, $p = 0.090$), HAART duration (OR = 1.233, 95%CI = 0.923–1.648, $p = 0.156$, and viral load (OR = 0.915, 95%CI = 0.774–1.082, $p = 0.298$). Lower resistance rates were recorded for meropenem and fosfomycin (1.7%), nitrofurantoin (6.8%, 4/59) and amikacin (8.5%, 5/59). The most common virulence genes identified were *chuA* (66.1%, 39/59), *papC* (57.6%, 34/59) and *cnf1* (50.8%, 30/59). Integrons identified were *intI* 42% (25/59) and *intII* 20% (12/59).

Conclusion ASB is common in our HIV- seropositive participants, indicating that WLHIV are at a higher risk of developing complications from UTIs. Though fosfomycin may be useful in managing UTIs in this population group,

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the high prevalence of multidrug-resistant, *E. coli* harbouring diverse virulence genes, warrants surveillance of ASB among this vulnerable group.

Keywords Asymptomatic bacteriuria, Urinary *Escherichia coli*, Virulence genes, Integrons, HIV seropositive women, Ghana, Antimicrobial resistance

Introduction

Urinary tract infections (UTIs) are one of the commonest bacterial infections affecting people globally. A study in 2022 estimated the burden of UTI from 1990 to 2019 and revealed that 404.6 million people had UTIs globally and nearly 236, 786 people died resulting in 5.2 million disability-adjusted-life-years (DALYs) [1]. UTIs account for high antibiotic usage across all age groups, high healthcare expenditure and a major cause of morbidity [2, 3]. It is a major public health concern particularly amongst women where more than half the population of women have at least one episode of UTI during their lifetime [4]. UTI is usually preceded by asymptomatic bacteriuria (ASB) [5]. ASB can be defined as the presence of significant bacterial counts ($\geq 10^5$ CFU/mL) of the same strain in two consecutive urines taken 24 h apart in the absence of clinical symptoms of UTI [6]. Amongst the etiologic agents, *Escherichia coli* remains the predominant uropathogen accounting for over 80% of the infections [7]. The pathogenic subset known as uropathogenic *E. coli*, possesses virulence genes such as *papC*, *iutA*, *hlyA*, *cnf1* among others, which are expressed to enhance adhesion, invasion and evasion of host defenses [8]. *E. coli* isolates are increasingly resistant to fluoroquinolones, trimethoprim-sulfamethoxazole and several classes of antibiotics due to the large spread of extended spectrum β-lactamases and plasmid mediated resistance genes [9].

The risk of ASB is high among sickle cell disease patients [10, 11], and has also been reported among pregnant women where untreated cases have progressed to UTI [12]. In HIV patients, UTI remain a major problem due to multidrug resistant pathogens [13]. Among PLHIV, women may be disproportionately affected by UTIs for anatomical reasons, such as a short urethra and proximity to the anal region, making it easy for bacteria to ascend to the urinary tract [14]. In sub-Saharan Africa, women account for the majority (60%) of PLHIV [15]. It is therefore important to understand the burden and risk factors associated with ASB in this population for targeted infection control and clinical outcomes. In Ghana, approximately 210,000 women aged ≥ 15 years were living with HIV as of 2019, and this accounts for 65.6% of the total HIV cases among adults in Ghana [16]. While there have been studies in Ghana on opportunistic infections in PLHIV [17–19], the burden of ASB in Women Living with HIV (WLHIV) remains unknown. Furthermore, data on virulence genes in Multi-Drug Resistant (MDR) uropathogens in this population group has not been adequately described. In this study, we determined the prevalence and risk factors associated with asymptomatic bacteriuria in women with HIV. Additionally, antimicrobial susceptibility of *E. coli* isolates were profiled together with virulence and integrase genes.

Materials and methods

Study site and population

We conducted a cross-sectional study on asymptomatic bacteriuria by randomly sampling WLHIV attending the antiretroviral treatment (ART) clinic at St. Martin de Porres Hospital, Eikwe, in the Western Region of Ghana, from March 1 st to July 30th, 2021. The St. Martin de Porres Hospital is a faith-based institution managed by the Catholic Diocese of Sekondi-Takoradi and is a member of the Christian Health Association, Ghana (CHAG). It functions as a district hospital within the Ellembelle district, serving as a major referral centre for the people of Nzema and neighbouring towns in the Ivory Coast. The hospital has a capacity of 200 beds with an annual Outpatient department (OPD) attendance exceeding 70,000 patients and over 13,700 yearly admissions. Its ART Clinic serves approximately 960 active patients with an average monthly attendance of approximately 200 patients. All HIV-seropositive women attending the HIV clinic were eligible for inclusion, except those under 15 years of age, those with complaints of UTI symptoms and those with recent antibiotic exposure other than trimethoprim-sulfamethoxazole.

Specimen collection and laboratory analysis

Midstream urine samples were obtained from each participant and processed within 2 h of collection at the Bacteriology Department of the hospital. In events where a delay in processing (> 2 h) was anticipated, urine samples were kept at 2–8 °C but not later than 6 h before processing. The urine samples were cultured on cysteine lactose electrolyte-deficient agar (Oxoid, Ltd., Basingstoke, UK) using a 10 µl standard loop and aerobically incubated at 37 °C. The plates were inspected after 18–24 h and those with bacterial counts of 1×10^5 CFU/mL were considered significant. Conventional biochemical tests (Simmons citrate, triple sugar iron slant, indole, urea slant) were used for the preliminary identification of organisms. Lactose-fermenting (yellow) colonies were sub-cultured onto eosin methylene blue (EMB) agar (Techno PharmaChem, India) and incubated aerobically at 37 °C. *E. coli* was differentiated from other lactose fermenters of

Enterobacterales by the production of a metallic green sheen on EMB agar. The final identities of isolates were confirmed via MALDI-TOF Biotyper (Bruker Daltonics, Bremen, Germany) at the Department of Medical Microbiology of the University of Ghana Medical School (UGMS). During processing, a single colony was picked from a 24-hour culture with sterile applicator and applied onto a target plate overlaid with 1 µL of α-cyano-4-hydroxycinnamic acid (HCCA) matrix (50% acetonitrile, 2.5% trifluoroacetic acid), and air-dried before analysis. Calibration was done using the Bruker Bacterial Test Standard according to the manufacturer's instructions. Identification scores ≥ 2.0 were accepted as reliable species-level identifications.

Antimicrobial susceptibility testing was performed using the Kirby-Bauer disc diffusion method with the following antibiotics discs; trimethoprim-sulfamethoxazole (cotrimoxazole) (25 µg), nitrofurantoin (300 µg), nalidixic acid (30 µg), cefuroxime (30 µg), ampicillin (10 µg), fosfomycin (200 µg), tetracycline (30 µg), ciprofloxacin (5 µg), gentamicin (10 µg), cefepime (30 µg), amikacin (30 µg), meropenem (30 µg), ceftriaxone (30 µg), and amoxiclav (30 µg) (Oxoid Ltd. Basingstoke, UK). Interpretation of zone sizes were done according to the Clinical and Laboratory Standards Institute (CLSI) guidelines (2021) [20].

The efficacy of the antibiotic discs and growth promotion potential of the culture media were ascertained with American Type Culture Collection (ATCC) control strains (*E. coli* ATCC 25922), and the results were interpreted via the CLSI 2021 guidelines [20].

Table 1 Primer sequences and amplicon sizes for the virulence and integrase genes

Gene	Primer Sequences [5′ – 3′]	Amplicon Sizes [bp]	Reference
Virulence Genes			
kpsMT II	**F**: GCGCATTTGCTGATACTGTTG	272	[22]
	R: CATCCAGACGATAAGCATGAGCA		
cnf1	**F**: AAG ATG GAG TTT CCT ATG CAG GAG	498	[23]
	R: CAT TCA GAG TCC TGC CCT CAT TAT T		
papC	**F**: GTGGCAGTATGAGTAATGACCGTTA	200	[22]
	R: ATATCCTTTCTGAGGGATGCAATA		
usp	**F**: ACATTCACGGCAAGCCTCAG	440	[24]
	R: AGCGAGTTCCTGGTGAAAGC		
iutA	**F**: ggctggacatcatgggaactgg	300	[25]
	R: cgtcgggaacgggtagaatcg		
chuA	**F**: gacgaaccaacggtcaggat	279	[26]
	R: tgccgccagtaccaaagaca		
Integrase Genes			
Int I	**F**: gggtcaaggatctggatttcg	483	[27]
	R: acatgggtgtaaatcatcgtc		
Int II	**F**: cacggatatgcgacaaaaaggt	788	[27]
	R: gtagcaaacgagtgacgaaatg		

Molecular analysis

DNA was extracted from overnight cultures of *E. coli* isolates plated on nutrient agar using a modified version of the boiling lysis method described by Ribeiro Junior et al. (2016) [21]. Three well-isolated colonies were suspended in 200 µl of double-distilled water and heated at 98 °C for 10 min. The suspension was then refrigerated for 10 min at −20 °C and centrifuged at 1,350 rpm for 5 min. The supernatant (DNA template) was aliquoted into 2 m Eppendorf tubes and stored at −20 °C until further use. Multiplex Polymerase Chain Reaction (PCR) was performed for the following genes: iron capture systems (*iutA*, *chuA)*, adhesin *(PapC)*, capsules (*kpsMTII)*, toxins (*cnf1*, *usp)* and integrons (*intI* and *intII)* with primers described in Table 1. The PCR was run with three primer sets: Set 1 (*iutA*, *kpsMTII*, *USP*, *cnf1*), Set 2 (*PapC*, *chuA*), and Set 3 (*intI*, *intII*). Each PCR reaction had 1 µl of 25 Pmol primer, 6.25 µl of OneTaq Quick-Load 2× Master Mix with Standard Buffer, 3.25 µl of nuclease-free water and 1.5 µl of DNA template, resulting in a final reaction volume of 15 µl. Nuclease-free water was used as a negative control. Characterized *E. coli* isolates with integrase genes were obtained from the microbiology department of UGMS for positive controls. However, no positive controls were available for the virulence genes at the time of the study. Amplification was performed via an Applied Biosystems Thermal Cycler (Thermo Fisher Scientific, USA). The amplicons were loaded onto a 2% agarose gel containing SYBR red and electrophoresed at 100v for one hour. The gel was observed under UV light and band sizes were compared to a 100 bp DNA ladder (See supplementary file S2 for cycling conditions for integrase and virulence genes).

Data management and statistical analysis

Sociodemographic and clinical data including age, WHO disease stage, HAART duration and viral load were collected via questionnaires developed specifically for this study (See supplementary file, S1). Information on each patient's most recent viral load results, WHO disease stage and HAART duration were obtained directly from patients' folders and verified against ART registers to ensure accuracy. Viral load results were categorized into < 1000copies/mL (viral suppression) and ≥ 1000 copies/mL (non-suppression) according to WHO criteria [28]. Data was recorded in Microsoft Excel files for preliminary analysis, and was later exported to the Statistical Package for Social Sciences (SPSS) (Version 29) for further analysis. Descriptive statistics were used for categorical variables, and associations were tested by the chi-square test. Odds ratios (ORs) and 95% confidence intervals (CIs) were derived from binary logistic regression analysis. P values < 0.05 were considered significant.

Table 2 Distribution of participants by demographics and clinical information

Variables	Frequency	Percentage (%)
Age groups (years)		
15 to 25	19	4.8
26 to 35	79	19.8
36 to 45	135	33.8
46 to 55	114	28.5
56 to 65	41	10.3
> 65	12	3.0
WHO disease stage		
Stage 1	38	9.5
Stage 2	76	19.0
Stage 3	250	62.5
Stage 4	36	9.0
HAART duration (years)		
< 1	17	4.1
1 to 5	215	53.8
6 to 10	110	26.8
> 10	58	14.1
Viral load (copies/mL)		
< 1000	314	78.5
≥ 1000	86	21.5

Results

Demographics and clinical characteristics

A total of 400 HIV seropositive women were enrolled in the study. The mean age of the participants was 43.54 years ± 12.03. A majority (78.5%, 314/400) of the participants had viral loads less than 1000 copies/mL. Majority (53.8%, 215/400) of the participants had been on highly active antiretroviral therapy for 1–5 years, and majority of the participants (62.5%, 250/400) were classified as having WHO stage 3 disease. (Table 2).

Table 2 represents the distribution of demographics and clinical information among study participants. Age groups were categorized into ten-year intervals. The disease stage follows WHO's classification of HIV infection. HAART duration is the number of years the participants have been on antiretroviral therapy and have been categorized into 4 (< 1, 1 to 5, 6 to 10 and > 10). Viral load was dichotomized using WHO classification (< 1000 = viral suppression and ≥ 1000 = non-suppression).

Prevalence of ASB and distribution among demographics and clinical characteristics

The overall prevalence of ASB among the study population was 21.2% (85/400). The prevalence of bacteriuria was apparently high among participants with a viral load < 1000 copies/mL 22.6% (71/314) those in the 15–25 years age group 31.6%(6/19), those in WHO disease stage 1 category 23.7% (9/37) and women with a HAART duration of less than 1 year, 47.1%(8/17). None of the predictor variables reached statistical significance; WHO disease stage (Wald $X^2(1) = 0.08$, $p = 0.777$), Age(Wald $X^2(1) = 2.882$, $p = 0.09$), HAART duration (Wald $X^2(1) = 2.02$, $p = 0.16$), and Viral load (Wald $X^2(1) = 1.08$, $p = 0.30$) (Table 3).

Table 3 represents the number and percentage of participants with positive and negative cultures within

Table 3 Association between demographics, clinical characteristics and positive culture

Variables	Positive culture (*n*, %)	Negative culture (*n*, %)	Wald X^2	OR	95% CI	*p* value
Age groups (years)			2.882	1.207	0.971–1.499	0.090
15 to 25	6 (31.6)	13 (68.4)				
26 to 35	10 (12.6)	69(87.3)				
36 to 45	30 (22.2)	105 (77.8)				
46 to 55	32 (28.1)	82 (71.9)				
56 to 65	12(29.3)	29(70.7)				
> 65	2 (16.7)	10(83.3)				
WHO disease stage			0.080	0.953	0.683–1.330	0.777
Stage 1	9 (23.7)	29 (76.3)				
Stage 2	14 (18.4)	62(81.6)				
Stage 3	55(22)	195 (78)				
Stage 4	7 (19.4)	29 (80.6)				
HAART duration (years)			2.017	1.233	0.923–1.648	0.156
< 1	8(47.1)	9 (52.9)				
1 to 5	38(17.6)	177(82.3)				
6 to 10	27(24.5)	83(75.5)				
> 10	15(25.9)	43(74.1)				
Viral load (copies/mL)			1.082	0.915	0.774–1.082	0.298
< 1000	71(22.6)	243(77.4)				
≥ 1000	14(16.3)	72(83.7)				

Wald X^2 Wald Chi-square test, *OR* odds ratio, *CI* confidence interval, *p*-value significance

each predictor variable category. ORs and 95%CIs were derived from binary logistic regression analyses. Overall *p* -values were obtained from the chi-square test of association. An OR < 1 indicates a reduced likelihood of positive culture. The variables with 95% CIs that excludes 1 and $p < 0.05$ indicates statistical significance.

Isolated uropathogens

Eighty-five (85) uropathogens were isolated and identified from urine samples that yielded significant microbial growth. The top three pathogens were *Escherichia coli* (69.4%, 59/85), *Enterococcus faecalis* (8.2%, 7/85), and *Klebsiella pneumoniae* (7.0%, 6/85) (Fig. 1).

The bar chart shows the percentage distribution of bacterial isolates recovered from 85 positive urine cultures. *Escherichia coli* was the predominant uropathogen, accounting for 69.4% (59/85) of isolates, followed by *Enterococcus faecalis* 8.2% (7/85), *Klebsiella pneumoniae* 7% (6/85), *Proteus mirabilis* 7% (6/85), and *Staphylococcus haemolyticus* 3.5% (3/85). Other Uropathogens isolated included *Enterobacter bugandensis*1.17% (1/85). *Enterobacter cloacae* 1.17% (1/85)., *Acinetobacter nosocomialis*1.17% (1/85), and *Enterobacter asburiae* 1.17% (1/85).

Distribution of virulence among *E. coli* isolates

The most widely distributed virulence genes, as shown in Fig. 2, were *chuA*, with a prevalence of 66.1%, followed by *papC*, *cnf1*, *kpsMTII* and *iutA*, with prevalence rates of 57.6%(34/59), 50.8%(30/59), 45.8%(27/59) and 35.6%(21/59), respectively.

The bar chart illustrates the prevalence of selected virulence genes detected in *E. coli* isolates recovered from urine samples. The most prevalent gene was *chuA* (66.1%), followed by *papC* (57.6%), *cnf1* (50.8%), *kpsMTII* (45.8%), and *iutA* (35.6%). Percentages were calculated from the total number of *E. coli* isolates tested ($n = 59$).

Distribution of Integrase genes among *E. coli* isolates

The prevalence of the *intI* integrase gene was 42.4% (25/59), while *intII* was detected in 20.3% (12/59) of isolates. No isolate carried multiple integrase gene (Fig. 3).

The bar chart shows the prevalence of *int1* and *intII* integrase genes detected in the urinary *E. coli* isolates obtained. The *int1* gene was present in 42.4% (25/59) of isolates, whereas *intII* was detected in 20.3% (12/59). None of the isolates carried more than one integrase gene.

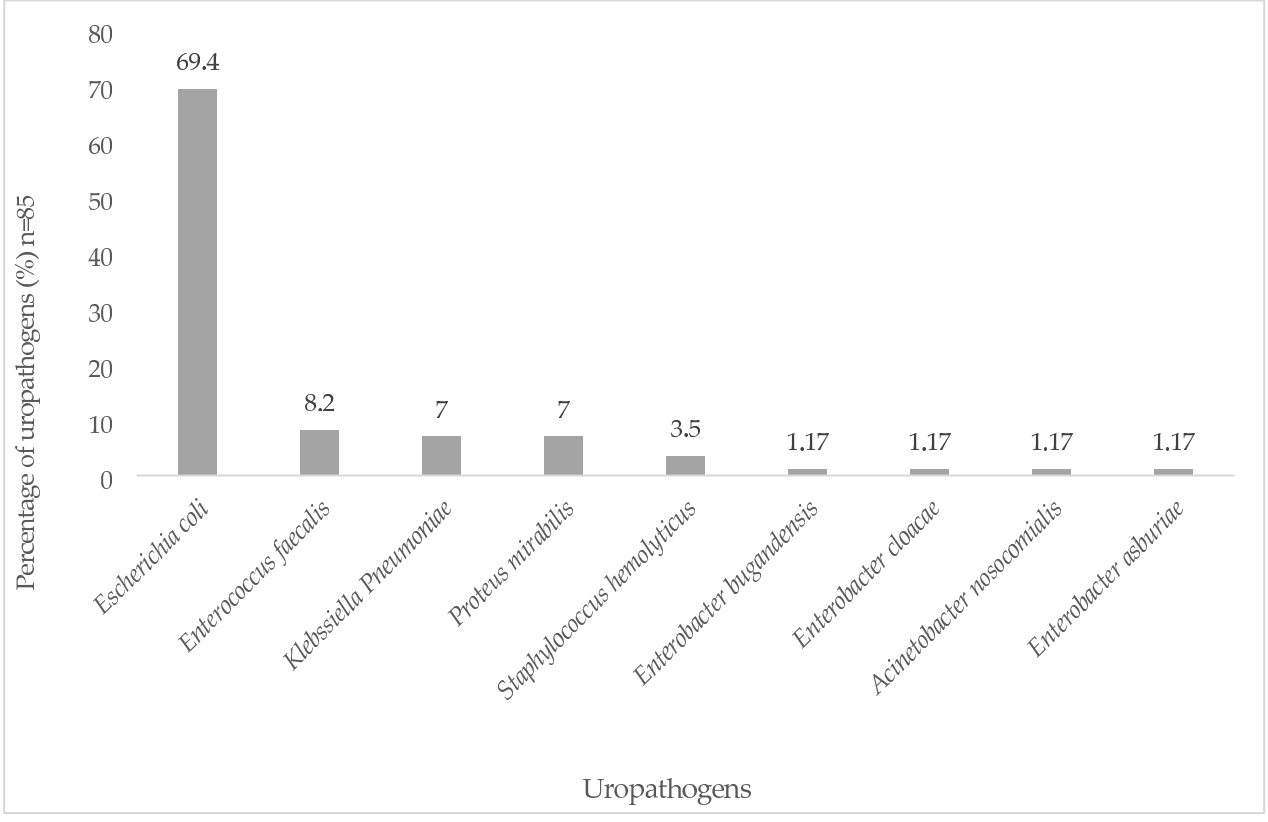

Fig. 1 Uropathogens isolated from women living with HIV at the St. Martin de Porres Hospital-Eikwe, Ghana, March 1 – July 30, 2021

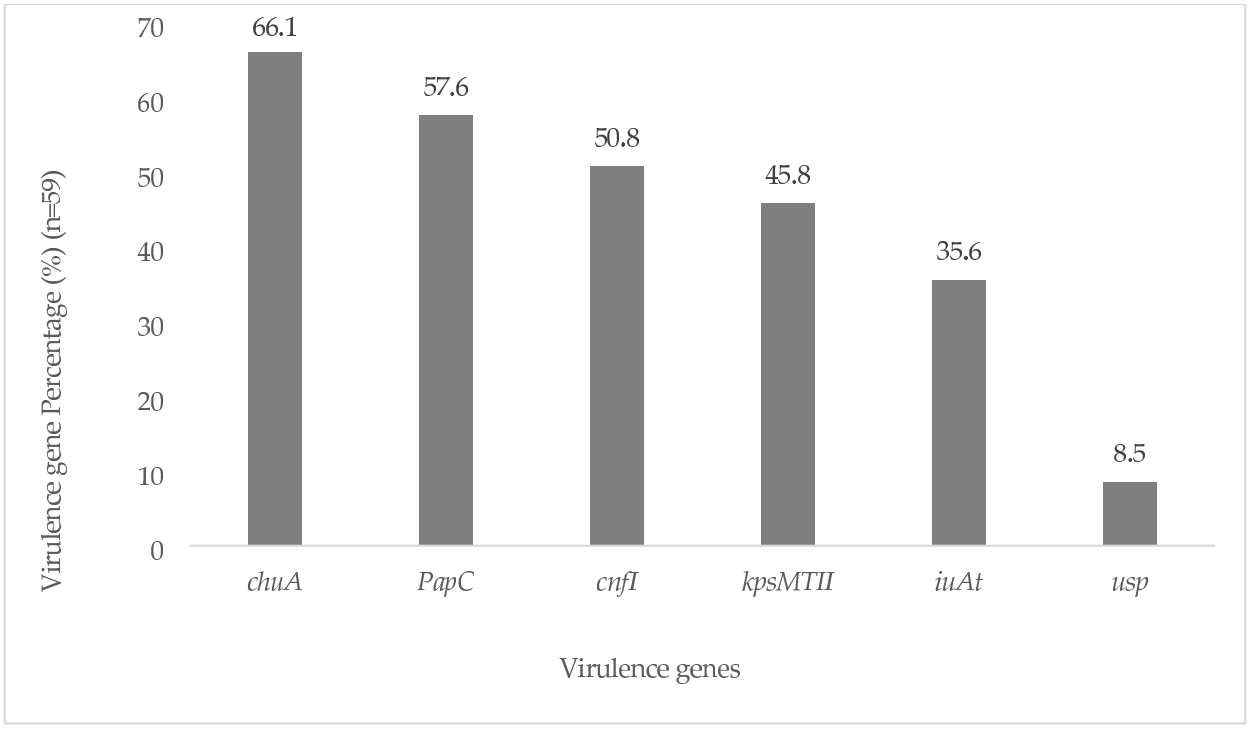

Fig. 2 Distribution of virulence genes in *E. coli* isolates from women living with HIV at St. Martin de Porres Hospital-Eikwe, Ghana March 1 – July 30, 2021

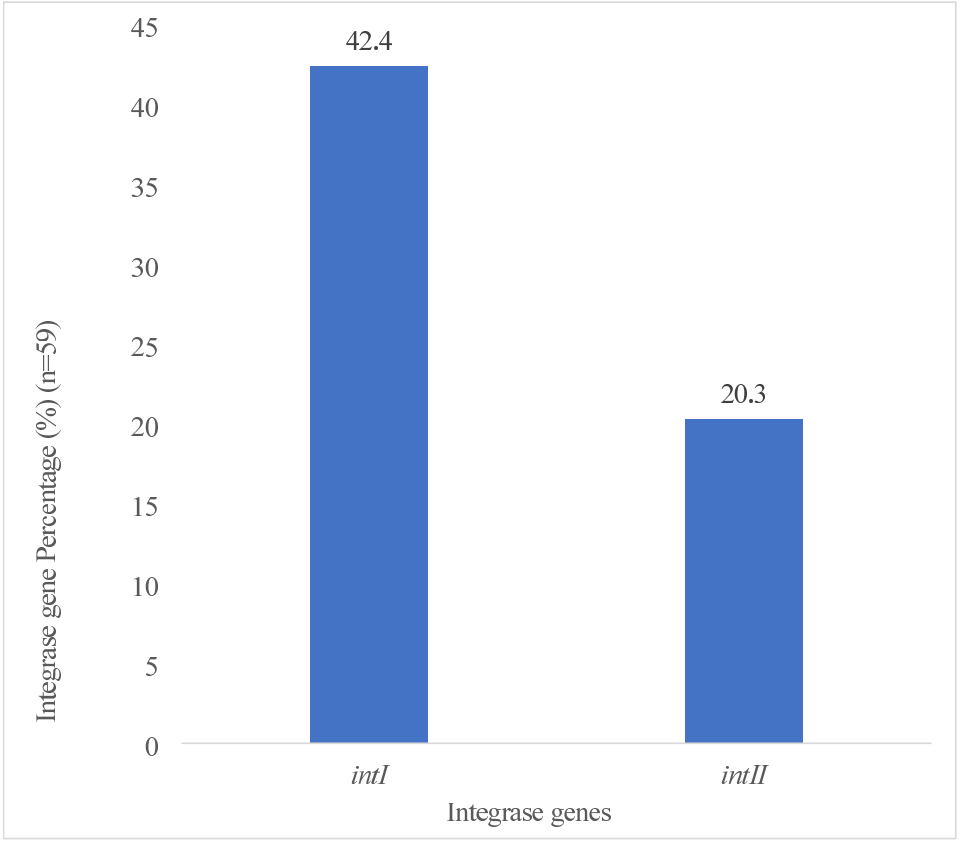

Fig. 3 Distribution of Integrase genes in *E. coli* isolates from women living with HIV at St. Martin de Porres Hospital-Eikwe, Ghana March 1 – July 30, 2021

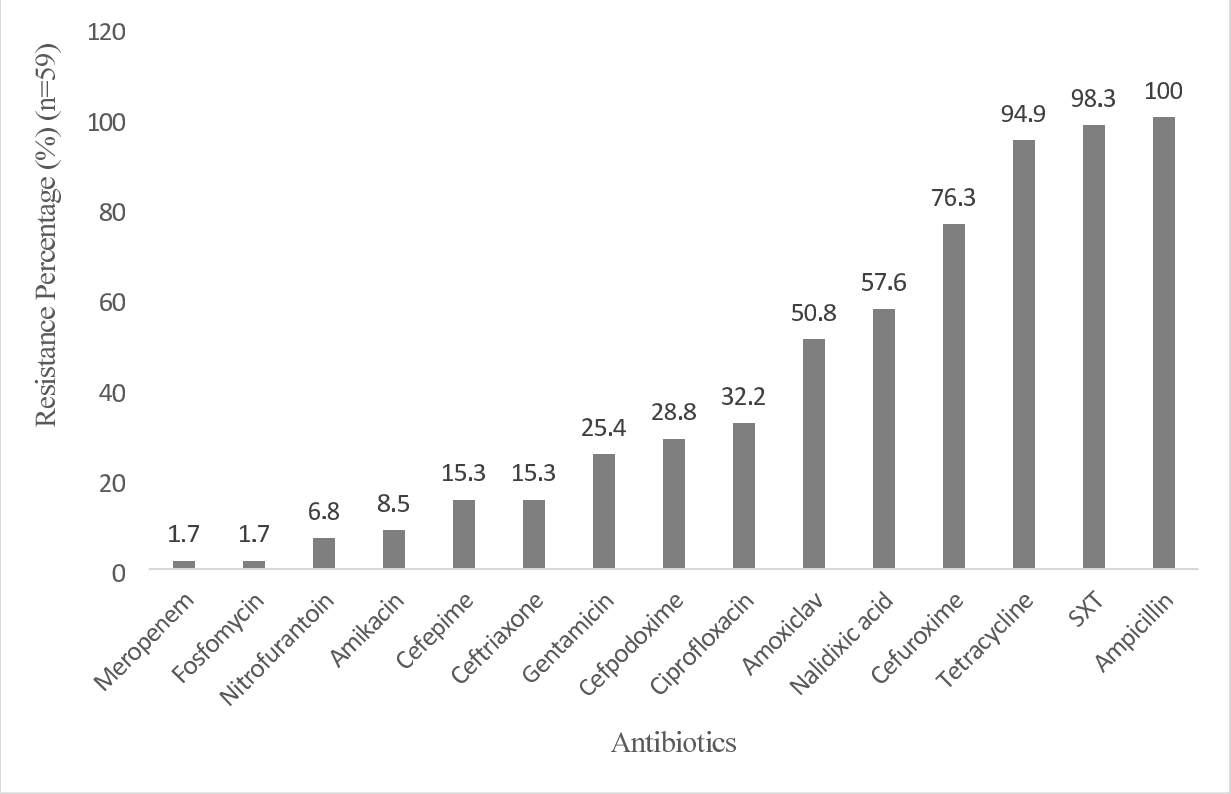

Fig. 4 Proportion of resistance of *E. coli* isolates from women living with HIV at St. Martin de Porres Hospital-Eikwe, Ghana March 1 – July 30, 2021

Proportion of resistance of *E. coli* isolates

The resistance pattern of *E. coli* isolates from the participants are shown in Fig. 4. All (100%) UPEC isolates were resistant to ampicillin. About 98.3% and 94.7% were resistant to sulfamethoxazole -trimethoprim and tetracycline respectively. The least resistance recorded was to meropenem and fosfomycin at 1.7%.

This bar chart shows the percentage resistance of urinary *E. coli* isolates to commonly tested antibiotics. All isolates (100%) were resistant to ampicillin, followed by high resistance to trimethoprim-sulfamethoxazole (98.3%), tetracycline (94.9%), and cefuroxime (76.3%). Moderate resistance was observed to nalidixic acid (57.6%), amoxiclav (32.2%), ciprofloxacin (28.8%), gentamicin (25.4%), cefpodoxime (25.4%), cefepime (15.3%), ceftriaxone (15.3%), and amikacin (8.5%). The lowest resistance rates were recorded for nitrofurantoin (6.8%), Fosfomycin (1.7%), and meropenem (1.7%).

Discussion

Asymptomatic bacteriuria (ASB) is reportedly common among PLHIV [29], and uropathogenic *E. coli* is the most prevalent organism. In this study, the prevalence of ASB among WLHIVs was greater than 21%. This finding indicates that WLHIV at our facility are at high risk of developing UTIs, as 30% of people with ASB may develop symptomatic UTIs if not treated [12]. The observed prevalence in our study was lower than the 57.8% and 51.8% reported in Nigeria and Poland, respectively [29, 30]. The comparatively lower prevalence in our study could be due to the inclusion of participants receiving prophylactic cotrimoxazole, which could reduce the risk of ASB. The highest prevalence of bacteriuria in our study was found among women in the 15–25 years age category. This finding is in agreement with reports by Karikari et al. (2020) who reported high bacteriuria (65.1%) among pregnant women aged 20–29 years [31]. A possible explanation could be that these individuals are more sexually active and that sex is a known risk factor for ASB and UTI [32, 33]. The participants assigned to the WHO stage 1 subgroup had the highest prevalence of bacteriuria. This is because women in this category are usually asymptomatic and may appear healthy and actively engage in sexual activities. However, the study revealed no association between WHO disease stage and ASB. This finding is similar to that of Iduoriyekemwen and colleagues (2012), who did not find any association between ASB and WHO disease stage in children with HIV/AIDS [34], but did not agree with others that established an association between ASB and WHO disease stages [35, 36]. These discrepancies may reflect differences in treatment protocols, treatment adherence, and the immune status of the participants.

In this study, the predominant uropathogen was *E. coli* similar to those from other regions of Ghana where the *E. coli* prevalence ranged from 36 to 63% [37–39]. Globally, *E. coli* is known to be the predominant cause of UTIs, including asymptomatic bacteriuria, and it represents the predominant gut flora [40]. The *E. coli* isolates in this study were highly resistant to ampicillin, trimethoprim-sulfamethoxazole, and tetracyclines, with resistance rates of 100%, 98.3%, and 94.9%, respectively. Similar findings of high antibiotic resistance among uropathogens have been reported in other regions of Ghana [41–43]. These findings suggest that these drugs may not be suitable for empirical treatment in our population. The high resistance to these drugs could be attributed to their affordability and accessibility, making it possible for self-medication, overuse and misuse, resulting in the development and spread of resistance. The least resistance was observed to fosfomycin, nitrofurantoin, and meropenem similar to other reports from Ghana, Nigeria, and Ethiopia [44–46]. The low resistance reported for these drugs could largely be the result of their infrequent use in Ghanaian healthcare settings. The Ghana Standard Treatment Guidelines (7th Edition, 2017) do not list these drugs among the first line drugs for empirical treatment of uncomplicated UTIs but rather; cefuroxime and ciprofloxacin . Nitrofurantoin is mainly reserved for lower UTIs whiles meropenem is a restricted antibiotic purposely for MDR infections [47]. Limited exposure to these drugs may account for the relatively low resistance observed. In addition, the route of administration for aminoglycosides makes self-medication and abuse difficult.

Frimodt-Møller and Bjerrum (2023), underscored that nitrofurantoin and fosfomycin can be used for empiric and targeted treatment of uncomplicated UTI due to their effectiveness and low likelihood of inducing resistance. Additionally, they recommended fosfomycin for the treatment of complicated UTI [48]. Although our findings corroborate the recommendation of Frimodt-Møller and Bjerrum (2023), this must be approached with caution as the empirical use of these drugs could accelerate the emergence of resistance and further limit treatment options. Their use should be guided by antibiotic susceptibility testing. Where this is not feasible, simple methods such as bacterioscopy can be performed to inform treatment decisions.

It is also imperative to restate that this study focussed on ASB whiles those recommendations apply to symptomatic uncomplicated and complicated UTIs. According to the Infectious Diseases Society of America [6] and the European Association of Urology [49], ASB treatment is not recommended in nonpregnant adults including people living with HIV, unless clinical indications such as pregnancy or planned urologic surgeries exist. Nonetheless, in women with advanced immunosuppression with low CD4 counts, ASB may progress to symptomatic infection easily, as several studies have shown lower CD4 counts are associated with high prevalence of bacteriuria and UTI [50, 51]. This calls for further longitudinal studies to define treatment thresholds in this population group.

In this study, all *E. coli* isolates displayed resistance to three or more antibiotics from different classes, indicating that they were multidrug resistant [52]. This may lead to treatment failure among this population using routinely recommended antibiotics.

Integrons play an important role in antibiotic resistance by capturing, integrating and expressing antibiotic resistance genes [53]. In this study, the carriage rates of the *intI* and *intII* genes were 42.4% and 20.3%, respectively. This agree with reports from Kenya, Ethiopia and an earlier study conducted among pregnant women in Ghana, where *IntI* was more common in the isolates [54–56]. Our study however reports a higher carriage of both *intI* and *intII* compared to findings of Forson and colleagues (2018) in Ghana, where the percentages of *E. coli* isolates harbouring the *intI* and *intII* genes were 12.2% and 2.4%, respectively [56]. No significant statistical associations were detected between integrase genes and resistance to trimethoprim-sulfamethoxazole (TMP-SMX), although 41.4% of the TMP-SMX-resistant isolates carried the *intI* gene. This is because TMP-SMX resistance genes (*sul* and *dhfr*) are predominantly carried on *intI* cassettes [57]. The findings here are similar to reports from Syria and Europe, where 54.7% and 57.6% of TMP-SMX-resistant UPEC isolates, respectively, carried *intI* genes [58]. The carriage rate in our study was lower than a report from Nigeria, where 72% of TMP-SMX resistant enterobacteria from the environment, carried *intI* gene [59]. It is also below the much higher rates reported in Korea (90%) and California (71.6%) [60, 61]. Over 40% of the *E. coli* isolates in this study were resistant to nalidixic acid, amoxicillin clavulanic acid, trimethoprim-sulfamethoxazole, cefuroxime, and tetracyclines, which also harbour *intI*. In other studies, a strong association was established between the *intI* gene and multidrug resistance, including ESBL production [58, 62].

The severity of UTIs is largely dependent on the array of virulence genes present and the levels at which they are expressed. In the current study, approximately 76.3% of the isolates harboured one or more virulence genes. *ChuA* was the most prevalent gene harboured by 66.1% of *E. coli* isolates. The *chuA* gene encodes a heme receptor protein and is ubiquitous in *E. coli*, assisting in the import of iron [63–65]. It aids in the formation of intracellular bacterial communities and displays biofilm-like properties that enable *E. coli* to resist immune responses through the development of a torpid pool of

bacterial cells inside uroepithelial cells [66, 67]. *PapC*, *cnf1*, *kpsMTII*, *iutA* and *usp* were prevalent in 57.6%, 50.8%, 45.8%, 35.6% and 8.5% of the isolates, respectively which is consistent with the findings of Forson and colleagues in Ghana [56]. Adherence is the first step in colonisation and the most important determinant of pathogenicity [68], which may explain the high prevalence of *papC*. The *papC* gene encodes a protein that is necessary for fimbriae P biogenesis regulation and has been reported as a precursor for pyelonephritis [69]. *Cnf1*, on the other hand, is a toxin that has been shown to induce apoptosis in human bladder cell lines [70] and has also been shown to decrease the phagocytic function of polymorphonuclear neutrophils [71]. Horcajada et al. (2005), revealed that *cnf1* is a good promoter of bacteraemia resulting from urinary tract infections [72]. Therefore, the combined prevalence (>50%) of these genes suggest that this HIV population group is at increased risk of infectious complications, with the possibility of developing bacteraemia of urologic origin.

Study limitation

Although the study population was representative of women attending our ART clinic, it may not be fully reflective of the larger population of WLHIV in Ghana. Bacteriuria may have also been underestimated due to the inclusion of participants on trimethoprim-sulfamethoxazole. Additionally, the diagnosis of ASB was based on a single-culture. This could have led to minor misclassification. Only a limited number of virulence genes were screened and the absence of other genes cannot be verified. Furthermore, the absence of positive controls for the virulence genes may affect the interpretation of negative results. However, positive controls for integrase genes were successfully amplified. This confirms the functionality of the PCR setup and minimizes the likelihood of technical failure.

Despite these limitations, the study provides an important insight into the burden of ASB, antimicrobial resistance, virulence and integrase gene carriage among WLHIV in Ghana.

Conclusions

This study revealed that asymptomatic bacteriuria is common among women living with HIV and age, viral load, WHO disease stage or HAART duration are not associated with *E. coli bacteriuria* in Western Ghana. The high prevalence of multidrug-resistant *E. coli*, coupled with the high carriage of virulence genes, possibly indicates that WLHIV are at high risk of developing invasive infections of urologic origin. Though fosfomycin may be useful in managing UTIs in HIV-seropositive individuals, regular monitoring of bacteriuria and antimicrobial susceptibility testing among this population group is warranted.

Supplementary Information

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Supplementary Material 1.

Supplementary Material 2.

Supplementary Material 3.

Supplementary Material 4.

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Authors' contributions

Conceptualization, JAO., AKL., and BE.; methodology, HKA., JAO., AKL., BE., EKM., NH., MMO., GRA., FO.; software, HKA., JAO., AKL., BE., EKM., NH., MMO., FO.; validation, JAO., AKL., and BE.; formal analysis, JAO., AKL., BE., EKM., NH., MMO., GRA., and FO.; investigation, HKA., EKM., NH., MMO., GRA., and FO.; resources, JAO., BE., PC., and EKM; data curation, HKA., JAO., AKL., and BE.; writing—original draft preparation, HKA., JAO., AKL., and BE.; writing—review and editing, HKA., JAO., AKL., BE., EKM., NH., MMO., GRA., FO., and PC.; visualization, HKA., JAO., AKL., and BE.; supervision, JAO., AKL., BE, and PC.; project administration, JAO., AKL., BE., and PC. All the authors have read and agreed to the published version of the manuscript.

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Data availability

The data used and/or analyzed during the current study are available in the paper.

Declarations

Ethics approval and consent to participate

The study was conducted according to the guidelines of the Declaration of Helsinki, and received ethical approval (CHS-Et/m.4.5.8/2020-21) from the Ethical and Protocol Review Committee of the College of Health Sciences, University of Ghana, and the Management of St. Martin de Porres Hospital, Eikwe. Written informed consent was obtained from participants seeking care at the ART clinic of St. Martin de Porres, Eikwe.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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