

Common Patterns and Unique Threats in Antimicrobial Resistance as Revealed by Global Gonococcal Surveillance, 2014–2022

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The rapid emergence of antimicrobial-resistant strains of *Neisseria gonorrhoeae* threatens treatment options and control efforts. The Uniformed Services University Gonococcal Reference Laboratory and Repository of the Global Emerging Infections Surveillance Program receives isolates from several geographically distinct regions worldwide. We analyzed 962 isolates collected during 2014–2022 for genomic and phenotypic antimicrobial resistance. Resistance to antimicrobial drugs previously used for gonococcal infections was high, but of most concern were increases of resistance to cur-

rently used antibiotic drugs, such as extended-spectrum cephalosporins and the alternative antibiotic treatment gentamicin. The percentage of isolates with reduced susceptibility to ceftriaxone was 3.6%, to cefixime was 2.5%, and to gentamicin was 15.0%. Although isolates were collected from populations of limited diversity, 706 (73.4%) of isolates demonstrated novel multiantigen sequence types, and 225 (23.4%) had novel multilocus sequence types. Continued surveillance of *N. gonorrhoeae* is essential to monitoring the prevalence and spread of resistant organisms worldwide.

Neisseria gonorrhoeae infections cause substantial illness globally, and control is challenged by increasing antimicrobial resistance. The World Health Organization (WHO) reported 82.4 million new *N. gonorrhoeae* infections worldwide among persons 15–49 years of age (1). In the United States, an

estimated 1.5 million new cases of gonorrhea are reported each year (2).

Gonococcal urogenital tract infections can cause severe complications, especially in women, who are often asymptomatic and go undiagnosed. Untreated cervical infections can cause upper genital tract

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disease, such as pelvic inflammatory disease, chronic pelvic pain, and ectopic pregnancy, and also increases the risk for tubal infertility. Urethral infections in men can ascend to cause epididymitis or orchitis; however, unlike cervical infections, urethral infections are usually symptomatic. The resulting discharge and dysuria increase the likelihood that male patients will seek testing and treatment.

Effective infection control is challenged by underdiagnosis of asymptomatic infections, lack of point-of-care diagnostics, and increasing persistent antimicrobial resistance. *N. gonorrhoeae* has developed resistance to all antibiotic drugs that have been used for routine treatment because of its ability to readily acquire genes through horizontal gene transfer or spontaneous mutations. The prevalence of antimicrobial resistance (AMR) within *N. gonorrhoeae* strains has steadily increased across the antibiotic era, necessitating frequent changes in treatment recommendations. The initial emergence of high-level penicillin and tetracycline resistance was followed by the introduction of fluoroquinolones for gonorrhea treatment in the mid-1980s, which were subsequently removed from treatment in 2007 (3). Dual therapy using extended-spectrum cephalosporins (ESCs) and azithromycin then became the primary recommended therapy for a decade. Azithromycin was removed in 2021 because of increasing resistance, leaving only ESCs for first-line treatment of gonorrhea. Globally, ceftriaxone is the sole remaining primary therapy for first-line treatment of gonorrhea in most guidelines (4–6). However, isolates with reduced susceptibility to ceftriaxone have proliferated worldwide, and multidrug-resistant, ceftriaxone-resistant strains have been reported in several countries (7–10), threatening simple outpatient therapy.

Because of the threat of untreatable gonorrhea, *N. gonorrhoeae* is classified by the Centers for Disease Control and Prevention as an urgent threat (11) and by WHO as a high-priority pathogen (12) for which new treatments are critically needed. Global rates of *N. gonorrhoeae* infections have been reported since 1992 through the WHO Enhanced Gonococcal Antimicrobial Surveillance Programme (EGASP). Data for 2017–2018 from 73 countries demonstrated resistance to ESCs of 0%–22%, azithromycin resistance of 0%–60%, and ciprofloxacin resistance of 0%–100% (13). Although several countries report AMR data to the EGASP, *N. gonorrhoeae* surveillance data from many global regions, such as Central America, Eastern Europe, Southeast Asia, sub-Saharan Africa, and the Eastern Mediterranean, remain scarce.

The Sexually Transmitted Infection (STI) National Strategic Plan for the United States (2021–2025) recognizes the need to improve STI prevention at the local, state, and federal levels. The plan also recommends that specific groups, such as the military and fraternal organizations, include services that address men's sexual health and their role in transmitting STIs (14). Military service members are at high risk for STI because of social demographics including age; however, factors such as increased alcohol consumption, diversification of sexual networks, and infrequent condom use also exacerbate risk in military populations (15). In addition, sexual assault, which carries an inherent risk for STI, has been reported in 1.0% of men and 4.9% of women in military service (16).

In alignment with the National Action Plan for Combating Antibiotic-Resistant Bacteria (14), and to monitor this urgent, ever-changing AMR threat, the Uniformed Services University (USU), in collaboration with the Armed Forces Health Surveillance Division's Global Emerging Infections Surveillance (GEIS) Branch, established the USU Gonococcal Reference Laboratory and Repository (GC Repository) within the USU Department of Microbiology and Immunology (Bethesda, Maryland, USA). This report analyzes trends in the susceptibility of *N. gonorrhoeae* isolates from different geographic regions to 8 different antibiotic drugs during 2014–2022 as part of the GEIS STI surveillance program. We also report the distribution of key alleles on the basis of genomic analysis to help define the prevalence of specific AMR determinants in different geographic regions. The investigators have adhered to the policies for protection of human subjects as prescribed in AR 70-25.

Methods

The GEIS STI initiative was established in 2010 to improve the health of the US armed forces and support force health protection decision-making. The GC Repository was established in 2014 to serve as a central entity for confirmatory testing and both phenotypic and genotypic characterization (17–19). As part of the surveillance program, a proficiency testing program was also established for quality assurance of partner laboratory methods for *N. gonorrhoeae* AMR testing.

Sampling Methods

We collected samples from persons enrolled in clinical care or public health surveillance activities during 2014–2022, which included military populations,

civilians, and high-risk populations from 5 geographic regions. We gram-stained from urethral, vaginal, cervical, pharyngeal, or rectal swab samples, plated them on selective media such as modified Thayer-Martin agar, and incubated for 24 hours at 37°C in 5% CO₂ or in a candle jar. We froze isolates of presumptive *N. gonorrhoeae* in 25% glycerol and tryptic soy broth and shipped to the GC Repository. We assessed AMR using Etest (bioMérieux, <https://www.biomerieux.com>) (Appendix, <https://wwwnc.cdc.gov/EID/article/30/14/24-0296-App1.pdf>) and performed agar dilution to confirm MICs for isolates with reduced susceptibility to azithromycin, ceftriaxone, cefixime, and gentamicin.

Reference Laboratory Testing

As of December 2023, the GC Repository received a total of 1,244 presumptive isolates from 6 countries: Thailand (n = 557), the Philippines (n = 35), Ghana (n = 73), Peru (n = 237), Kenya (n = 211), and Georgia (n = 95). We confirmed isolates by culture on modified Thayer-Martin agar, Gram staining, oxidase test positivity, superoxol test positivity, and API NH biochemical test (bioMérieux). We used detection of the *porA* pseudogene to resolve inconclusive API NH test results (Appendix). We determined MICs for all 962 isolates (Appendix).

Whole-Genome Sequencing and Bioinformatic Analysis

We sent *N. gonorrhoeae* isolates to the Walter Reed Army Institute of Research’s Multidrug-Resistant Organism Repository and Surveillance Network (Silver Spring, Maryland, USA) for whole-genome sequencing (Appendix) and genotypic characterization. Multilocus sequence typing (MLST) was performed in silico using the *N. gonorrhoeae* scheme curated by Maiden (20). We performed additional in silico molecular typing using *N. gonorrhoeae* multiantigen sequence typing (NG-MAST) and *N. gonorrhoeae* sequence typing

for antimicrobial resistance (NG-STAR) with ngmaster version 1.0.0 (21) (Appendix).

Results

Of the 1,244 frozen suspensions of presumptive *N. gonorrhoeae* from 5 geographic regions received by the GC Repository, 962 (77.3%) were confirmed as *N. gonorrhoeae* isolates. Among isolates for which the type of sample was recorded, most came from urethral swab samples taken from men. Limited, inconsistent demographic data were available to the partner laboratories involved in public health surveillance.

Antimicrobial Susceptibility Testing

We compiled phenotypic AMR data for all 962 isolates (Table 1). Benzylpenicillin resistance was most commonly observed (917/962 [95.3%]), followed by tetracycline (902/962 [93.7%]) and ciprofloxacin (882/962 [91.7%]). Resistance to those antibiotic drugs varied among sites; ≈50% resistance to each of those antibiotic drugs was observed in Georgia, whereas other sites exhibited up to 90% resistance. Elevated MICs (IR≥1; R≥2) to azithromycin was found in 10 isolates, 8 of which had azithromycin MICs of 1 and 1.5 µg/mL (2 from Georgia, 2 from Peru, and 4 from Thailand); 2 isolates had MICs >256 µg/mL (Kenya). Among isolates from Kenya, 5 exhibited reduced susceptibility to the ESCs: 1 for cefixime, 2 for ceftriaxone, and 2 for both cefixime and ceftriaxone. Similarly, 11 Georgia isolates exhibited reduced susceptibility to ESCs. We observed that 84% (809/962) of the isolates were susceptible to gentamicin (S≤4; IR = 8–16; R≥32). Of the remaining isolates, 7 (0.7%) had MICs of 16 µg/mL (4 from Peru, 2 from Georgia, and 1 from Ghana). All 962 isolates were susceptible to spectinomycin. Multidrug resistance was common among all international collection sites. The frequency of resistance to any 3 antibiotic drugs ranged from 11% (Ghana) to 92% (Peru).

Table 1. Summary of phenotypic antimicrobial resistance in study of common patterns and unique threats in antimicrobial resistance as demonstrated by global gonococcal surveillance*

Region	Isolates with reduced susceptibility or resistance, no. (%)						
	Tetracycline	Benzylpenicillin	Ciprofloxacin	Azithromycin	Cefixime	Ceftriaxone	Gentamicin
Thailand, n = 516	500 (96.9)	502 (97.3)	500 (97)	4 (0.77)	2 (0.4)	16 (3.1)	31 (6.0)
Ghana, n = 19	19 (100)	19 (100)	17 (89.5)	0	1 (5.3)	0	7 (36.8)
Peru, n = 208	195 (93.7)	205 (98.5)	186 (89.4)	2 (0.96)	7 (3.4)	3 (1.4)	63 (30.3)
Nairobi, Kenya, n = 27	27 (100)	26 (96.3)	23 (85.1)	0	0	0	1 (3.7)
Kisumu, Kenya, n = 110	108 (98.2)	105 (95.5)	106 (96.4)	2 (1.8)	3 (2.72)	4 (3.63)	26 (23.6)
Uganda, n = 10	9 (90)	10 (100)	9 (90)	0	0	0	2 (20)
Georgia, n = 72	44 (61.1)	50 (69.4)	41 (56.9)	2 (2.8)	11 (15.2)	11 (15.2)	16 (22.2)
Total, N = 962	902 (93.7)	917 (95.3)	882 (91.7)	10 (1.02)	24 (2.5)	34 (3.6)	146 (15.2)

*MICs interpreted according to Clinical and Laboratory Standards Institute criteria when available (22). CLSI resistance breakpoints used for penicillin (I>0.06; R≥2.0 µg/mL), tetracycline (I>0.25; R≥2.0 µg/mL), and ciprofloxacin (I>0.06; R≥1.0 µg/mL) (22). Gonococcal Isolate Surveillance Project breakpoints used for azithromycin (I≥1; R≥2.0 µg/mL), cefixime (I>0.06; R≥0.25 µg/mL), and ceftriaxone (I>0.06; R≥0.125 µg/mL) (23,24), because CLSI has not established criteria for resistance to those antimicrobial drugs. Gentamicin breakpoints (I≥8–16 µg/mL; R I≥32.0 µg/mL) were determined according to research published by the Centers for Disease Control and Prevention (25).

Table 2. Presence of antimicrobial-resistant genetic determinants in study of common patterns and unique threats in antimicrobial resistance as demonstrated by global gonococcal surveillance*

Region	No. (%) isolates								
	Tetracycline resistance		Benzylpenicillin resistance		Ciprofloxacin resistance		Azithromycin resistance	Cefixime and ceftriaxone resistance	
	V57	<i>tetM</i>	β -lactams	<i>ponA</i> _{L421P}	<i>gyrA</i>	<i>parC</i>	<i>mtrD</i>	<i>Mtr</i>	<i>penA</i>
Thailand, n = 516	506 (98)	449 (87)	<i>bla</i> _{TEM-1} , 327 (63.4); <i>bla</i> _{TEM-135} , 77 (15)	80 (15.5)	S91, D95, 318 (61.6)	D86, 217 (42); S87, 218 (42.2); E91, 21 (4); S88, 8 (1.5)	<i>MtrD</i> S821A K823E, 7 (1.35)	Internal stop codon, 27 (5.3)	I312M V316T G545S, 2 (0.38)
Ghana, n = 19	19 (100)	15 (79)	<i>bla</i> _{TEM-1} , 13 (68.4)	16 (84.2)	S91, D95, 17 (89.5)	D86, 3 (17.6); S87, 12 (70.6); E91, 1 (5.9)	0	G45D, 7 (36.8)	I312M V316T G545S, 1 (5.26)
Peru, n = 208	208 (100)	87 (41.8)	<i>bla</i> _{TEM-1} , 111 (53.3); <i>bla</i> _{TEM-135} , 13 (6.25); <i>bla</i> _{TEM-22} , 1 (0.48)	120 (57.6)	S91; D95, 188 (90.3)	D86, 52 (28); S87, 46 (24.7)	<i>MtrD</i> mosaic 2, <i>MtrR</i> mosaic 2, 2 (0.96)	G45D, 21 (10)	I312M V316T G545S, 17 (8.17)
Nairobi, Kenya, n = 27	27 (100)	23 (85.2)	<i>bla</i> _{TEM-1} , 22 (81.5)	12 (44.4)	S91, D95, 21 (77.7)	D86, 1 (4.76)	0	0	0
Kisumu, Kenya, n = 110	110 (100)	99 (90)	<i>bla</i> _{TEM-1} , 32 (29); <i>bla</i> _{TEM-135} , 3 (2.72); <i>bla</i> _{TEM-239} , 5 (4.54)	66 (60)	S91, D95, 106 (96.3)	D86, 10 (9); S87, 9 (8.2); E91, 65 (59)	23s rDNA A2045G, 2 (1.8)	A39, 84 (76.5); G45, 2 (1.8); D79, 11 (10); M197, 1 (0.9)	A501, F504, A516 N512, 4 (3.6)
Uganda, n = 10	10 (100)	10 (100)	<i>bla</i> _{TEM-1} , 8 (80); <i>bla</i> _{TEM-135} , 1 (10); <i>bla</i> _{TEM-239} , 1 (10)	5 (55)	S91, D95, 10 (100)	D86, 4 (40); S87, 2 (20); E91, 4 (40)	0	A39, 8 (80); D79, 2 (20)	F504, 10 (100); A516, 10 (100)
Georgia, n = 72	47 (65.3)	14 (19.4)	<i>bla</i> _{TEM-1} , 11 (15.3)	40 (55.5)	S91, D95, 41 (57)	D86, 12 (19.5); S87, 31 (75.6); E91, 12 (29.3)	<i>MtrD</i> mosaic 2, <i>MtrR</i> mosaic 2, 2 (2.7); <i>MtrD</i> S821A K823E, 5 (6.9)	A39, 28 (38.9); G45, 6 (8.3); D79, 14 (19.4); M197, 2 (2.7)	I312M V316T G545S, 9 (12.5%)
Total, N = 962	928 (96.4)	697 (72.5)	<i>bla</i> _{TEM-1} , 524 (54.5); <i>bla</i> _{TEM-135} , 94 (9.77); <i>bla</i> _{TEM-239} , 6 (0.62); <i>bla</i> _{TEM-22} , 1 (0.10)	339 (35.2)	S91, D95, 701 (72.9)	D86, 299 (31); S87, 318 (33); E91, 103 (10.7)		A39, 641 (66.6); G45D, 36 (3.74)	

*Percentages calculated on total number of *Neisseria gonorrhoeae* isolates confirmed for the country.

Molecular Determinants of AMR and Genomic Characterization

All 676 isolates with high-level tetracycline resistance (Tet^R) (MIC >8 μ g/mL) (676/962 [97.7%]) isolates harbored the *tetM* gene and the *rpsJ* V57M mutation, whereas isolates with MICs of 0.5–3 μ g/mL did not carry the *tetM* gene but had the *rpsJ* V57M mutation (Table 2). Among the 917 benzylpenicillin-resistant isolates carrying β -lactamase-producing plasmids, 4

different β -lactamase resistance genes were detected; *bla*_{TEM-1} was detected in 57.1% of isolates and *bla*_{TEM-135} was detected in 10.3% of isolates. One isolate from Peru harbored the *bla*_{TEM-22} plasmid. The *bla*_{TEM-239} plasmid was present in 6 isolates from East Africa (1 from Uganda and 5 from Kenya).

In contrast, the number of isolates harboring chromosomally mediated determinants of AMR varied widely. Mutations in the *mtrR* gene (G45D) were

present in 3.7% of isolates, and mutations in the *mtr* promoter region (−35Adel) were present in 10% percent of isolates, whereas the A39THTH mutation was more prevalent (66.6% of isolates). Overall, we identified MtrR disruptions in 12% of isolates. The *ponAL421P* mutation was found in 35.2% of isolates, whereas *porB* mutations A121N, G120K, and A121D were less common and found in 2.2% of isolates (A121N), 8.6% of isolates (G120K), and 3.8% of isolates (A121D). All isolates with reduced susceptibility or resistance to ciprofloxacin (MICs 1 to >32 µg/mL) harbored S91F and D95G/A/N mutations in *gyrA*. Mutations in *parC* (D86, S87, or E91K) were found in 74.8% of isolates. We found the *fusA* A563V mutation, which confers reduced susceptibility to gentamicin, in 1 isolate from Peru (26,27).

Isolates with reduced susceptibility to azithromycin harbored myriad chromosomal resistance determinants. Mosaic *mtrD* and *mtrR* alleles were found in 4 isolates (2 from Georgia and 2 from Peru). One of those Georgia isolates also carried the *penA* mosaic allele XXXIV. One isolate from Peru carried the *mtrR* mosaic allele but lacked the *mtrD* mosaic allele. We found that 7 isolates from Thailand harbored *mtrD* S821A K823E mutations associated with azithromycin resistance (28), but only 4 of the 7 isolates had reduced susceptibility to azithromycin (MICs ≥1 µg/mL). The 23s rDNA A2045G mutation was present in 2 isolates from Kenya (MIC >256 µg/mL). Examination of ESC resistance determinants showed that 32 of the 962 isolates carried mosaic *penA* alleles. We detected 4 mosaic *penA* alleles: XXXIV (24 isolates [2.5%]), 166 (2 isolates [0.2%]), and 217 (5 isolates [0.5%]); 1 isolate (0.1%) had a novel allele. Of those 32 isolates, 29 had reduced susceptibility to cefixime and 11 had

reduced susceptibility to both cefixime and ceftriaxone. We found that 5 isolates with reduced susceptibility to both ceftriaxone and cefixime did not carry a mosaic *penA* allele.

We monitored the type of *porB1* allele present, which encodes the major outer membrane porin (PorB). *N. gonorrhoeae* strains express 1 of 2 *porB1* alleles. The *porB1A* allele is associated with strains that cause disseminated infection, whereas strains with *porB1B* more frequently cause localized infections (29). Although *porB1B* strains are usually more common, the frequencies of *porB1A* and *porB1B* alleles were similar among the 962 isolates, except for Thailand, Georgia, and Peru isolates. Thailand isolates cultured before 2016 (n = 88) carried *porB1A* more frequently (81/88 [92.0%]) than *porB1B* (7/88 [8.0%]). After 2016, *porB1B* strains were isolated more often in Thailand. Among isolates from Georgia, only 5 (6.9%) isolates expressed *porB1A*, and in Peru, 62 (47.3%) of 131 *porB1A*-expressing isolates were collected during 2014–2017, compared with 14 isolates (18.2%) of 77 *porB1A* strains collected during 2018–2022.

Molecular typing identified 98 NG-MAST, 198 MLST, and 199 NG-STAR sequence types (STs) among the 962 isolates (Figure 1). We found that 706 isolates belonged to a novel NG-MAST ST. The most common defined NG-MAST STs were ST6211 (n = 36), ST8058 (n = 21), ST2318 (n = 14), ST5573 (n = 12), and ST681 (n = 10). We identified novel MLST STs in 225 of 962 isolates. The most common MLST STs were ST1587 (n = 133), 1588 (n = 80), 7363 (n = 55), 8756 (n = 55), 8143 (n = 44), and 7827 (n = 40). Those isolates were all ciprofloxacin resistant. Using NG-STAR, we identified 173 novel types. The most common defined NG-STAR STs were ST719 (n = 69), ST271 (n = 24),

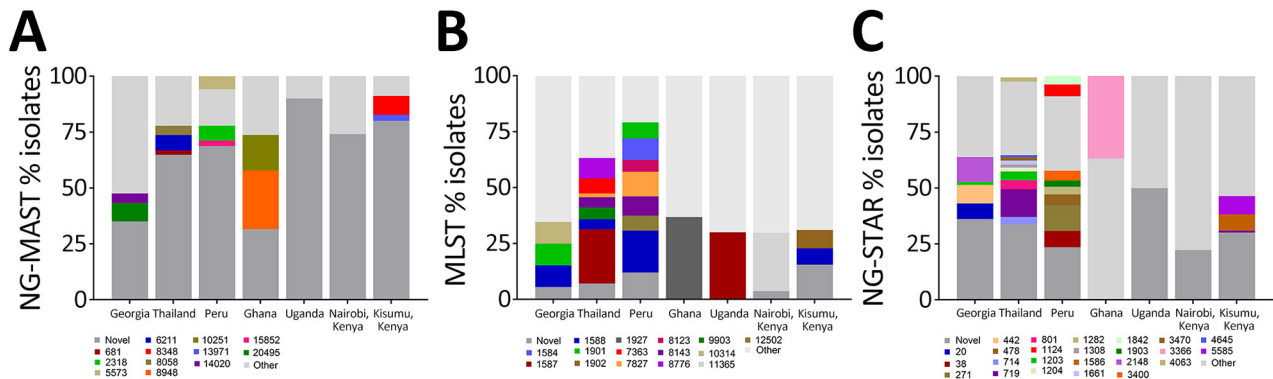


Figure 1. Distribution of most prevalent NG-MAST, MLST, and NG-STAR schemes in Global Emerging Infections Surveillance isolates of *Neisseria gonorrhoeae* received at Uniformed Services University, Bethesda, Maryland, USA, from sites outside the United States (n = 962) in study of common patterns and unique threats in antimicrobial resistance as demonstrated by global gonococcal surveillance. A) Percentage of isolates assigned to the most common NG-MAST types in each region. B) Percentage of isolates assigned to the most common MLST types in each region. C) Percentage of isolates assigned to the most common NG-STAR types in each region. MLST, multilocus sequence typing; NG-MAST, *N. gonorrhoeae* multiantigen sequence typing; NG-STAR, *N. gonorrhoeae* sequence typing for antimicrobial resistance.

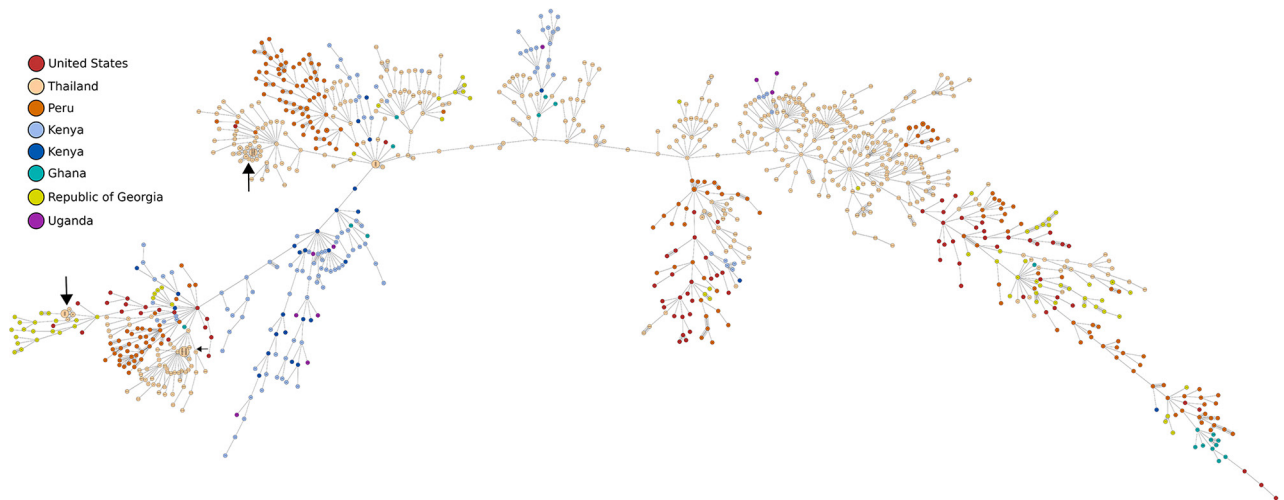


Figure 2. Minimum-spanning tree showing genome-based genetic relatedness of all *N. gonorrhoeae* isolates received at Uniformed Services University ($n = 1,044$), Bethesda, Maryland, USA, in study of common patterns and unique threats in antimicrobial resistance as demonstrated by global gonococcal surveillance. Tree was generated using core genome multilocus sequence typing. Each circle represents ≥ 1 isolates; isolates with 1–10 allelic differences are emphasized by gray shading around the lines and are considered highly genetically related with suspicion of nosocomial origin. Isolates are colored corresponding to their country of origin. Possible clonal isolates are shown with black arrows.

ST801 ($n = 23$), and ST1203 ($n = 22$). The distribution of NG-MAST, MLST, and NG-STAR STs also revealed that certain STs are specific to various regions (Figure 1). We generated a minimum-spanning tree on the basis of core genome MLST of all isolates, categorized by geographic location, to examine genomic diversity and possible clonal spread (Figure 2). Isolates from Thailand clustered into 4 major groups, and 3 appear to be clonal isolates (black arrows). Georgia isolates also clustered, but some were closely related to isolates from Thailand (≈ 300 core genome allele differences). Isolates from Peru grouped into 5 clusters.

Discussion

Increasingly resistant *N. gonorrhoeae* infections present a major public health burden for civilian communities, military force health protection, and US military readiness. Surveillance programs incorporating specimen culture are critical for linking genotypic and phenotypic AMR data to enable AMR prediction. The WHO EGASP program provides data from 68 countries in 6 regions as of 2018 (30). The GEIS network fills some key gaps in surveillance, including Eastern Europe and East Africa, where *N. gonorrhoeae* AMR data remain scarce.

This study reports phenotypic and genotypic analyses of geographically and temporally diverse NG isolates collected through the GEIS STI surveillance program. Isolates from international sites displayed high frequencies of resistance to benzylpenicillin, tetracycline, and ciprofloxacin, ranging from

50% to 100%. Those data are similar to data from previously published literature. Investigators from Peru identified $\approx 95\%$ resistance to ciprofloxacin (31–33), whereas reduced susceptibility or resistance to penicillin was observed in 99.4% of isolates and to tetracycline in 94.5% of isolates (31). In Peru, 76% of isolates were reported to have reduced susceptibility to gentamicin (31). In comparison, however, our study identified reduced susceptibility to gentamicin in $\approx 15\%$ of isolates. Regional differences in AMR patterns can be driven by community-based factors, including limited access to care and lack of available diagnostics, leading to empiric treatment. Similarly, lack of access to recommended antibiotic drugs and readily available access to other over-the-counter antibiotic drugs in the absence of valid healthcare encounters can also drive selection for *N. gonorrhoeae* AMR. For example, ciprofloxacin is still used empirically to treat STIs in Peru and other countries in Latin America. In Uganda, cefixime therapy is recommended but not easily available (34).

In Georgia, the recommended treatment for *N. gonorrhoeae* remains 1 g ceftriaxone plus 2 g azithromycin. Isolates from Georgia displayed lower frequencies of resistance to penicillin, tetracycline, and ciprofloxacin ($\approx 50\%$) than did isolates from Africa and Asia ($>90\%$). However, isolates from Georgia were more likely to exhibit reduced susceptibility to ESCs ($\approx 15\%$) than were isolates from Asia and Africa ($\approx 3.6\%$). *N. gonorrhoeae* can develop resistance to antibiotic drugs within a few decades of introduction

(35). Earlier uptake of ESCs in Georgia might account for the decrease in susceptibility seen, compared with our isolates collected from the global south. Antimicrobial susceptibility among Georgia isolates might also be affected by population changes caused by neighboring political unrest. Several studies of STIs in migrants, refugees, and internally displaced persons observe that these populations might be at higher risk for sexual assault and STI (36). However, the potential association between migration and *N. gonorrhoeae* AMR requires further study (37).

Many multidrug-resistant *N. gonorrhoeae* isolates originate in Asia (1). However, isolates from Thailand tested at the GC Repository exhibited low overall frequencies of resistance to primary therapies such as cefixime (0.4%), ceftriaxone (3%), and azithromycin (0.77%). The findings are surprising given the regional history of resistant *N. gonorrhoeae*; however, other recent surveillance studies in Thailand have observed similar results (38). The GC Repository recently received 18 isolates collected from high-risk patients in Pattaya, Thailand, that exhibited higher frequencies of resistance to macrolides and ESCs.

Recently, the US Centers for Disease Control and Prevention published guidelines on preventive treatment for bacterial STIs using doxycycline postexposure prophylaxis (doxyPEP) (39). Multiple prospective studies observed a reduction in incident bacterial STIs among men who have sex with men who were taking doxyPEP (40–42). Those studies have largely focused on syphilis, but the effect on *N. gonorrhoeae* infection has been noted. For example, in South Africa, doxyPEP reduced *N. gonorrhoeae* infections in men by 50%, but no difference was observed in cisgender women in Kenya taking doxyPEP compared with women in the standard care group (43). Many isolates tested at the GC Repository had the *tetM* gene, which is harbored in the easily spread pCONJ plasmid and can be transferred with *pbla* (44,45), which might counter the potential effectiveness of doxyPEP for gonorrhea prevention. Although doxycycline therapy is not commonly used for contemporary treatment of *N. gonorrhoeae*, continued surveillance is essential to understand the potential effects of doxyPEP on transmission and AMR.

Limitations of this study include low sample size and a study population that might be neither population-representative nor representative of the United States or partner nation militaries. As previously mentioned, most isolates originated from urethral samples taken from men, largely because of both local clinical standards of care at collection sites and ease

of sample collection and culture. Genital specimens from female patients, in contrast, are more difficult to culture, possibly because of the robust female urogenital microbiome. Extragenital isolates, which were infrequent in this study, are particularly relevant because of their proximity to commensal *Neisseria*, which may provide opportunities for horizontal gene transfer and acquisition of genetic determinants of AMR. In addition, the GC Repository has limited access to demographic and clinical data, such as sex or military status.

As of January 2024, two new antibiotic drugs for the treatment of gonorrhea infections, zoliflodacin and gepotidacin (46,47), have undergone Phase III clinical trials with promising results. Even with impending availability, however, the ease of AMR development in *N. gonorrhoeae* still portends a grim outlook for long-term treatment effectiveness. Without a vaccine, enhanced surveillance of *N. gonorrhoeae* AMR that combines culture, epidemiologic information, and molecular data must continue to identify genetic determinants of AMR and inform appropriate treatment recommendations.

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