Outbreak of Sexually Transmitted Nongroupable *Neisseria meningitidis*–Associated Urethritis, Vietnam

Hao Trong Nguyen,[1] Thanh V. Phan,[1] Hau Phuc Tran, Thao Thi Phuong Vu, Nhi Thi Uyen Pham, Tho Thi Thanh Nguyen, Ha Manh Bui, Bao Hac Duong, Thu Nguyen Anh Luu, Nguyen Nhat Pham, Phuc Duy Nguyen, Tu Ngoc Le, Thu Quang Le, Dai Thi Trang Vo, Lan Trong Phan, Nghia Van Khuu, Quang Duy Pham, Thuong Vu Nguyen

We report on an outbreak of nongroupable *Neisseria meningitidis*–associated urethritis, primarily among men who have sex with men in southern Vietnam. Nearly 50% of *N. meningitidis* isolates were resistant to ciprofloxacin. This emerging pathogen should be considered in the differential diagnosis and management of urethritis.

Urogenital and anorectal infections caused by *Neisseria meningitidis* have been reported in several countries and found to be more prevalent among men who have sex with men (MSM) than among heterosexual men or women (*1*–*3*). During 2013–2016, rising numbers of a novel clade of nongroupable *N. meningitidis* (NmNG) urethritis were reported in multiple US cities and have been termed US NmNG urethritis clade (*4*). Two cases of US NmNG urethritis were also documented among MSM in the United Kingdom in 2019 (*5*). We report an outbreak of urethritis associated with US NmNG urethritis clade among men in southern Vietnam.

The Study

We conducted a matched case–control study to investigate *N. meningitidis* urethritis and risk factors in men seeking treatment for urinary discharge at Ho Chi Minh City Hospital of Dermato-Venereology (HHDV; Ho Chi Minh City, Vietnam). Cases of *N. meningitidis* urethritis were confirmed by either real-time PCR or culture of urethral discharge. Controls were matched to case-patients by age range and sexual orientation (Appendix, https://wwwnc.cdc.gov/EID/article/29/10/22-1596-App1.pdf). During September 2019–December 2020, we recruited 19 case-patients and 76 controls from HHDV (Appendix Figure 1). We collected information on sociodemographic factors, sexual behaviors, and medical history by face-to-face interviews and from medical records (Appendix). The HHDV institutional review board approved the study.

We identified *N. meningitidis* by using bacterial culture and real-time PCR targeting the *sodC* gene (*6*) and determined serogroups by using latex agglutination and real-time PCR. We performed antimicrobial susceptibility testing according to Clinical and Laboratory Standards Institute guidelines (*7*). We conducted whole-genome sequencing, then analyzed multilocus sequence types (MLST) in PubMLST (https://pubmlst.org/organisms/neisseria-spp). We used BEAST (http://beast.community) to estimate the time of bacterial arrival in Vietnam and to conduct antimicrobial-resistance typing (Appendix). We performed conditional logistic regression to assess risk factors for US NmNG urethritis clade by using Stata 14 (StataCorp LLC, https://www.stata.com). We used a log likelihood-ratio test to select the best-fitting model (Appendix).

The mean age of case-patients was lower than that of controls (26.9 vs. 27.8 years). Condom use was low in both case-patients and controls before pyuria developed (5.3% of case-patients, 2.7% of

Author affiliations: Ho Chi Minh City Hospital of Dermato-Venereology, Ho Chi Minh City, Vietnam (H.T. Nguyen, T.T.P. Vu, N.T.U. Pham, T.T.T. Nguyen, H.M. Bui, B.H. Duong, T.N.A. Luu, N.N. Pham); Pasteur Institute of Ho Chi Minh City, Ho Chi Minh City (T.V. Phan, H.P. Tran, P.D. Nguyen, T.N. Le, T.Q. Le, D.T.T. Vo, L.T. Phan, N.V. Khuu, Q.D. Pham, T.V. Nguyen)

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[1]These first authors contributed equally to this article.

Table 1. Correlates of several selected factors among male patients with *Neisseria meningitidis* US NmNG urethritis and controls, Vietnam*

Variables	Cases, n = 19	Controls, n = 76	Univariable analysis† OR (95% CI)	p value	Multivariable analysis† aOR (95% CI)	p value
Mean age, y	26.9	27.8	0.90 (0.75–1.07)	0.232	NA	
Mean years of education	10.8	11.0	0.96 (0.75–1.23)	0.760		
Currently living in Ho Chi Minh City	14 (73.4)	56 (73.7)	1.00 (0.30–3.28)	>0.999		
Living arrangements						
Live with a female partner	1 (5.3)	22 (30.1)	Referent		Referent	
Live with a male partner	8 (42.1)	10 (13.7)	17.03 (1.83–158.26)	0.013	14.41 (1.01–204.62)	0.049
Other, e.g., live alone, or with friends or family	3 (15.8)	12 (16.4)	5.44 (0.65–45.54)	0.118	7.0 (0.61–80.24)	0.118
Ever had sex with						
Male sexual partners	14 (73.7)	49 (64.5)	Referent			
Female sexual partners	4 (21.1)	16 (21.1)	0.41 (0.02–7.54)	0.551		
Both male and female partners	1 (5.3)	11 (14.5)	0.17 (0.01–3.17)	0.238		
Ever had oral sex	19 (100.0)	74 (97.4)	NA			
Ever participated in group sex	1 (5.3)	2 (2.6)	2 (0.18–22.06)	0.571		
During past 12 mo						
Oral or vaginal sex with female partner	4 (21.1)	44 (57.9)	0.10 (0.02–0.47)	0.004	0.13 (0.02–0.87)	0.035
Oral or anal sex with male partner	15 (78.9)	39 (51.3)	10.57 (1.28–87.34)	0.029	NA	
Any casual partners	5 (26.3)	34 (44.7)	0.39 (0.12–1.32)	0.131	NA	
Commercial sex worker partner	3 (15.8)	28 (36.8)	0.30 (0.08–1.15)	0.078		
Drunkenness during sex	3 (15.8)	23 (30.3)	0.33 (0.07–1.65)	0.177		
Sex with a foreign-born partner in the past month	3 (15.8)	1 (1.3)	12.0 (1.25–115.36)	0.031	26.78 (1.03–697.82)	0.048
Condom use during sex before symptom onset‡	1 (5.3)	2 (2.7)	1.81 (0.16–20.08)	0.628		
Used social media sites to find sexual partners	13 (68.4)	32 (42.1)	4.07 (1.17–14.13)	0.027	NA	
Ever used ATS§	1 (5.3)	4 (5.3)	1.00 (0.10–10.07)	>0.999		

*Values are no. (%) except as indicated. ATS, amphetamine-type stimulants; aOR, adjusted odds ratio; NA, not applicable; OR, odds ratio.
†Conditional logistic regression.
‡Symptoms were pyuria, dysuria, or both.
§Ecstasy, crystal meth (methamphetamine), or other substances used to increase excitement when having sex (also called chemsex).

Table 2. Correlates of demographic characteristics, STI symptoms and pathogens among male patients with *N. meningitidis* US NmNG urethritis and controls, Vietnam*

Variables	Cases, n = 19	Controls, n = 76	Univariable analysis† OR (95% CI)	p value	Multivariable analysis† aOR (95% CI)	p value
Mean age, y	26.9	27.8	0.90 (0.75–1.07)	0.232	NA	
Mean years of education	10.8	11.0	0.96 (0.75–1.23)	0.760	NA	
Medical examination ≥3 d after symptom onset	13 (68.4)	11 (14.5)	18.41 (4.04–83.86)	<0.001	16.00 (2.00–127.54)	0.009
Symptoms						
Pyuria	2 (10.5)	10 (13.2)	0.79 (0.16–3.79)	0.764	NA	
Dysuria	13 (68.4)	65 (85.5)	0.39 (0.12–1.19)	0.098	NA	
Burning sensation during urination	2 (10.5)	57 (75.0)	0.05 (0.01–0.22)	<0.001	0.08 (0.01–0.46)	0.005
Discharge	4 (21.1)	16 (21.1)	1.00 (0.26–3.89)	>0.999		
Time between symptom onset and medical consultation, d						
Mean	4.7	3.0	1.92 (1.18–3.13)	0.009	NA	
Median (range)	5 (3–12)	3 (1–14)				
History of meningococcal vaccine						
Ever	0	1 (1.3)	NA			
Never	12 (63.2)	51 (67.1)	NA			
Do not know, do not remember	7 (36.8)	24 (31.6)	NA			
Positive tests						
HIV	2 (10.5)	6 (7.9)	1.36 (0.26–7.20)	0.716	NA	
Syphilis	2 (10.5)	3 (3.9)	2.67 (0.45–15.96)	0.283	NA	
Gonorrhea	0 (0.0)	75 (98.7)	NA		NA	
Chlamydia	4 (21.1)	7 (9.2)	2.57 (0.67–9.83)	0.168	3.83 (0.37–39.43)	0.258
Ureaplasma	1 (5.3)	8 (10.5)	0.48 (0.06–4.01)	0.502	NA	
Mycoplasma	1 (5.3)	10 (13.2)	0.39 (0.04–3.10)	0.372	NA	

*Values are no. (%) except as indicated. aOR, adjusted odds ratio; NA, not applicable; OR, odds ratio.
†Conditional logistic regression.

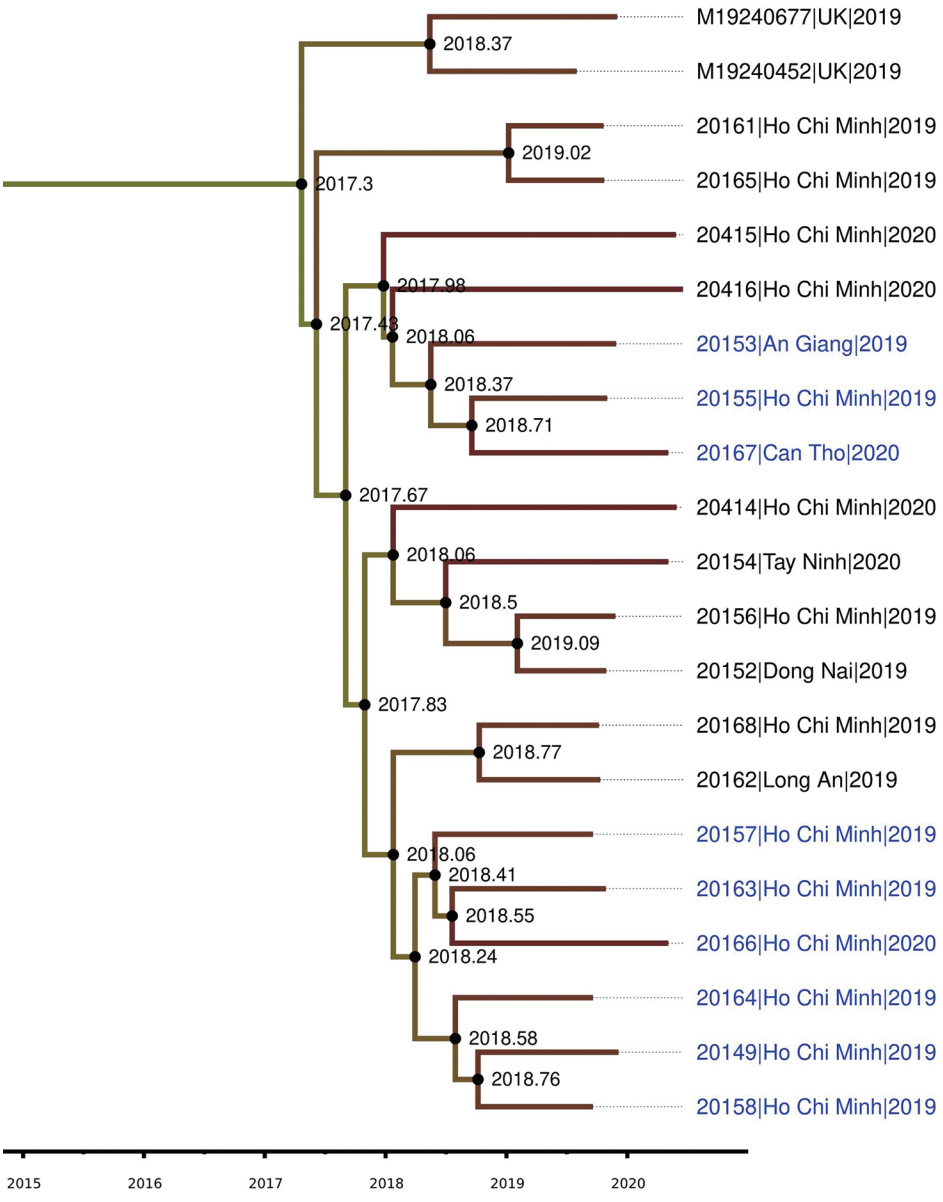

Figure 1. Phylogenetic tree of isolates from an outbreak of sexually transmitted nongroupable *Neisseria meningitidis*–associated urethritis, Vietnam. Phylogenetic tree was constructed using Baysian Skygrid model, performing with BEAST/BEAGLE version 1.10.4 (https://beast.community/beagle), and displaying with FigTree version 1.4.4 (http://tree.bio.ed.ac.uk/software/Figtree). Blue text indicates ciprofloxacin-resistant strains. Scale bar indicates the time of evolutionary history.

controls). More case-patients than controls lived with male partners (42.1% vs. 13.7%) and had sex with foreign-born persons (15.8% vs. 1.3%). Multivariate analysis results showed that persons living with male partners (adjusted odds ratio [aOR] 14.41, 95% CI 1.01–204.62) and having sex with foreign-born persons (aOR 26.78, 95% CI 1.03–697.82) were more likely to contract US NmNG urethritis (Table 1). Moreover, most (79%) case-patients reported sex with male partners. Persons having oral or vaginal sex with female partners in the past 12 months were less likely to have US NmNG urethritis (aOR 0.13, 95% CI 0.02–0.87). Those findings suggest that the US NmNG outbreak was concentrated within the MSM population.

Among 19 case-patients, 7 were co-infected with ≥1 other pathogen: 2 (11%) cases of syphilis, 4 (21%) cases of chlamydia, and 1 (0.1%) case involving both ureaplasma and mycoplasma. Ten (83%) case-patients without co-infections and 6 (86%) with co-infections experienced ≥1 symptom. Pyuria was reported in 2 (29%) co-infected case-patients, and dysuria was reported in 10 (83%) case-patients without co-infections and 3 (43%) with co-infections (Appendix Table 1). Most participants had not received meningococcal

vaccines, nor recalled being vaccinated against *N. meningitidis* (Table 2). The prevalence of HIV, syphilis, and chlamydia infections was higher among case-patients compared to controls but not statistically significant, whereas gonorrhea was only found in controls (98.7%) (Table 2). Multivariate analysis showed that those who had US NmNG urethritis were less likely to report burning sensations during urination (odds ratio [OR] 0.08, 95% CI 0.01–0.46) and more likely to delay seeking treatment (OR 16.0, 95% CI 2.0–127.54) (Table 2).

In this study, uncomplicated gonorrhea was treated with a single 500-mg intramuscular dose of ceftriaxone, followed by either a 7-day course of doxycycline (100 mg 2×/day) or a single 1,000-mg dose of azithromycin. In Vietnam, gonococci isolated in 2011 and during 2015–2016 increasingly resisted antimicrobial drugs except for ceftriaxone, spectinomycin, and azithromycin (*8*). Moreover, 98.3% of *N. gonorrhoeae* isolates were ciprofloxacin-resistant (*9*). In a national survey conducted in Vietnam, 30% of persons reported purchasing antibiotics primarily for addressing symptoms, including genitourinary manifestations, and 81.7% did so without a prescription; ciprofloxacin was among the top 5 antimicrobial drugs acquired (*10*). In another study among MSM in Vietnam, 64% reported ever taking antibiotics without a prescription (*11*).

The US NmNG urethritis clade in our study displayed intermediate susceptibility to penicillin, with MIC values ranging from 0.125–0.38 mg/L. Nine of the 19 isolates demonstrated resistance to ciprofloxacin (MIC 0.19–3.0 μg/mL). MLST analysis revealed that the isolates belonged to the sequence type 11 complex (Appendix Table 2). A phylogenetic tree displayed the isolates from Vietnam and the United Kingdom forming a monophyletic clade with those from Ohio, USA, one of the 2 US NmNG urethritis clades (*12*) (Appendix Figure 2). BEAST analysis estimated that the time of most recent common ancestor of Vietnam and UK isolates appeared between 2016 and 2018 (median 2017.3; 95% high posterior density interval 2016.4–2018.1), with a Bayesian posterior probability of 1.0 (Figure 1; Appendix Figure 3).

All isolates carried the penA_316 allele and point mutations that reduced their susceptibility to penicillin (*13*). Nine ciprofloxacin-resistant isolates

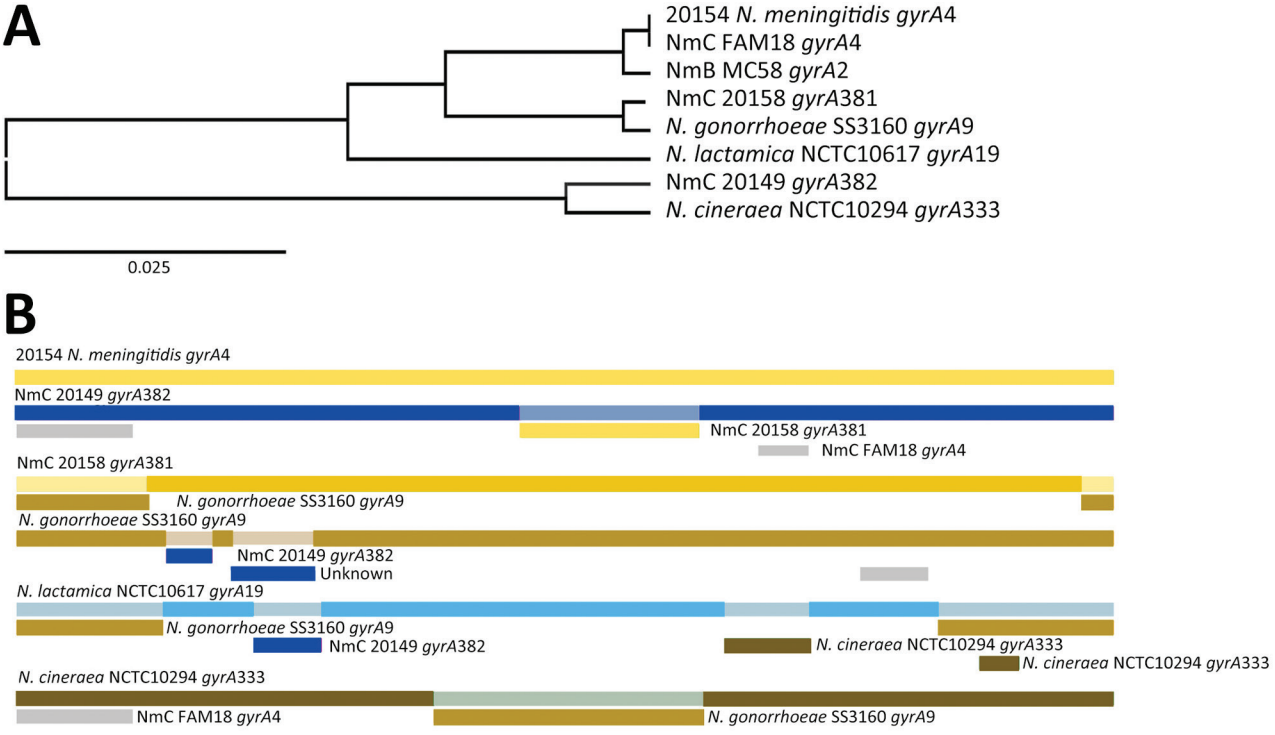

Figure 2. Whole-genome analysis of isolates from an outbreak of sexually transmitted nongroupable *Neisseria meningitidis*–associated urethritis, Vietnam. Comparison generated in RDP4 (https://rdp4.software.informer.com) for full length of 2,751-bp. A) Tree shows the genetic relationship between isolate 20158_*gyrA381* and gonococci_*gyrA9* using unweighted pair group method with arithmetic mean of the region derived from their parents, beginning at 1 to ending breakpoint at 337 bp. B) Bars show potential recombination breakage points identified with at least 1 of the 7 methods contained in RDP4. NmB, *N. meningitidis* B; NmC, *N. meningitidis* C.

exhibited 2 new alleles in *gyrA*, assigned as *gyrA*_381 (n = 8), which had dual mutations at T91F and D95A, and *gyrA*_382, which had monomutation at T91I. We used RDP4 (https://rdp4.software.informer.com) to analyze the full 2,751-bp length and found that *gyrA*_381 received a fragment containing a mutation from gonococci (Figure 2). Our study revealed that isolates containing mutations at both T91F and D95A in the *gyrA* gene displayed a high level of resistance to ciprofloxacin, similar to that found in *N. gonorrhoeae* (*14*). Moreover, isolate 20158 had a mutation at S87R of *parC* and the *gyrA*_381 allele had an elevated MIC of 3 μg/mL.

The emergence of ciprofloxacin-resistant *N. meningitidis* US NmNG urethritis clade in Vietnam is a major concern, especially considering ciprofloxacin resistance is rare in the United Kingdom and United States (*4*,*5*). A previous study observed that strains with MICs ≥0.064 mg/L were correlated with alterations in *gyrA* (*15*). Hence, when specimens cannot be cultured, *gyrA*-sequencing can be particularly useful in predicting susceptibility of ciproloxacin.

Conclusions

We report an outbreak of US NmNG urethritis among men in Vietnam, predominantly MSM. Having sex with foreign-born persons and living with male partners were factors strongly associated with the disease. Isolates in this outbreak might have originated from the Ohio (USA) clade and were mainly resistant to ciprofloxacin, which is commonly used for prophylaxis against invasive meningococcal diseases in Vietnam.

Symptoms among patients with US NmNG urethritis were milder than those in controls with gonococcal urethritis. Because US NmNG urethritis is less likely than gonococcal urethritis to manifest symptoms, clinicians should consider *N. meningitidis* when managing patients with urethral discharge. Bacterial culture should be routinely performed on urethritis specimens that test *N. gonorrhoeae*–negative by nucleic acid amplification tests to determine whether the infection is caused by *N. meningitidis*. More studies with larger sample sizes should be conducted to provide a more comprehensive picture of the burden and clinical symptoms of US NmNG urethritis.

In conclusion, our findings emphasize the importance of ongoing monitoring of appropriate use of antibiotics and antimicrobial resistance to prevent the further spread of resistant US NmNG urethritis clade. Thus, clinicians should be aware of this emerging bacterium and include US NmNG in the differential diagnosis for urethitis.

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The findings and conclusions in this publication are those of the authors and do not necessarily represent the official position of the government of Vietnam, the Ministry of Health, the Pasteur Institute of Ho Chi Minh City, and the Ho Chi Minh City Hospital of Dermato-Venereology.

Authors contributions: H.T.N., T.V.P., Q.D.P., and T.V.N. conceived the work and designed the study. H.T.N. oversaw the data collection at the hospital, and T.V.P.was responsible for testing the specimens. T.V.N., H.T.N, T.V.P., and H.P.T.oversaw the study, interpreted the results and compiled the manuscript. T.T.PU, N.T.U.P., T.T.TN., H.M.B, B.H.D., T.N.A.L, and N.N.P. conducted data collection from patients and assisted in data management and analysis. P.D.N, N.V.K, T.N.L., Q.D.P., T.Q.L., H.P.T., D.V.T.T., and L.T.P contributed to data management and analysis. All authors approved the final manuscript.

About the Authors

Dr. H.T. Nguyen is a senior researcher in Ho Chi Minh City Hospital of Dermato-Venereology, Vietnam. His research interests include clinical dermatology and sexually transmitted diseases. Mr. Phan is a microbiologist at Pasteur Institute in Ho Chi Minh City, Vietnam. His research interests focus on the agents that cause bacterial meningitis, particularly *Neisseria meningitidis* and *Streptococcus pneumoniae*.

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Address for correspondence: Thuong Vu Nguyen, Department for Disease Control and Prevention, Pasteur Institute in Ho Chi Minh City, 167 Pasteur St, District 03, Ho Chi Minh City 700901, Vietnam; email: nguyenthuong@yahoo.com or thuongnv@pasteurhcm.edu.vn

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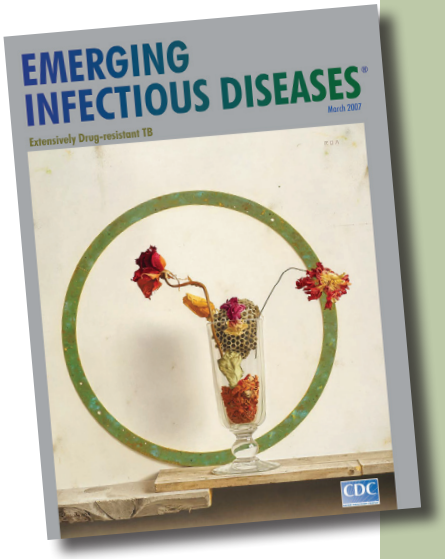

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Norovirus

[nor′-o-vi′rəs]

Genus of viruses that cause viral gastroenteritis. Noroviruses are named after the original strain, "Norwalk virus," which caused an outbreak of acute gastroenteritis among children at an elementary school in Norwalk, Ohio, in 1968. Numerous outbreaks of disease with similar symptoms have been reported since, and the etiologic agents were called "Norwalk-like viruses" or "small round-structured viruses." Noroviruses are transmitted primarily through the fecal-oral route and are highly contagious; as few as 10 viral particles may infect a person.

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https://wwwnc.cdc.gov/eid/article/13/3/e1-1303_article

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Outbreak of Sexually Transmitted Nongroupable *Neisseria meningitidis*–Associated Urethritis, Vietnam

Appendix

Methods

A matched case-control study was conducted among male patients with urinary discharge seeking treatment at Ho Chi Minh City Hospital of Dermato-Venereology (HHDV). A case was defined as a man who sought sexually transmitted infection (STI) treatment and care at HHDV and presented with urinary discharge and was confirmed with either real-time PCR or culture of urethral discharge specimen. A control was defined as a man who visited HHDV with the presence of urinary discharge with a diagnosis of a non–*Nessaria meningitidis* infection. For each case, four controls were recruited, matching the case-patient by age ranges (16–19, 20–29, 30–39, and ≥40 years) and sexual orientation (having sex with females only, having sex with males). In total, there were 19 cases and 76 controls recruited to the study during 9/2019 to 12/2020. From September 2019 to December 2020, 2,702 male patients were diagnosed with urethritis. Most male patients presented with symptoms of pyuria, dysuria, burning sensation during urination, and dripping. The study was approved by the HHDV institutional review board.

Data Collection

Demographic and other information of the cases, including age, gender, place of residence, clinical symptoms, diagnosis, and treatment, were retrieved from medical records and screening tests for HIV, chlamydia, syphilis, gonorrhea, ureaplasma, and mycoplasma that had been performed at the HHDV. Investigators conducted face-to-face interviews with the cases using a standardized questionnaire. Patients were asked about their social demographic

characteristics, medical and immunization history, access to social networks, and sexual and drug use behaviors. The controls were also interviewed using the same questionnaire.

Laboratory Investigation

Isolate Collection

At the HHDV, men presenting with urinary discharge were initially suspected to have gonococcal urethritis. However, 19 of the collected specimens tested negative for *Gonococci* using real-time PCR and subsequent bacterial culture yielded 19 isolates of Gram-negative diplococci, which could not be identified as gonococci. These isolates were then transferred to Pasteur Institute of Ho Chi Minh for further laboratory examination to confirm the actual etiology.

Identification and Characterization of *N. meningitidis*

To identify *N. meningitidis*, two methods were used: 1) a biochemical test kit of Analytical Profile Index Neisseria-Haemophilus (API-NH, bioMérieux); and 2) species-specific superoxide dismutase gene (¬sodC) real-time PCR (Quanta Bioscience). A slide agglutination serogrouping (SASG) kit of N. meningitidis Antiserum (Difco) and serogroup-specific (cps) gene rt-PCR were used to determine the serogroup of the samples, among A, B, C, W, X, Y or nongroupable (NG).

Antimicrobial Susceptibility Testing

Antimicrobial susceptibilities for the urethral isolates were determined through MIC with E-test, using six antibiotics: penicillin, cefotaxime, meropenem, azithromycin, ciprofloxacin, and rifampin (Liofilchem). The Clinical and Laboratory Standard Institute (CLSI) 2017 was used to determine breakpoints for susceptible, intermediate, or non-susceptible/resistant (*1*).

Whole-Genome Sequencing

Whole-genome sequencing of 19 urethral *N. meningitidis* isolates was performed at the PIHCM, Vietnam, from which DNA was extracted using the QIAmp DNA mini kit (QIAGEN). Libraries were then constructed using the Nextera XT DNA Library Prep Kit v2.0 (Illumina). Next, an Illumina-Miseq Platform generated 300-bp paired-end reads by running 300 cycles. Fastq reads were trimmed with Trimmomatic, and de novo assembled with SPAdes v3.13 (*2*,*3*). Multilocus sequence typing (MLST), fine-typing antigens, targeted outer-membrane proteins

(OMV), and antibiotic resistance genes were obtained through BIGSdb (*4*). All draft genomes were submitted to the National Center for Biotechnology Information (NCBI) under BioProject no. PRJNA672782 and were also uploaded to the PubMLST database.

Phylogenetic Analysis

Phylogenetic inferences based on differences in the gene-by-gene comparison of 1,605 defined loci of the core-genome MLST were made. The genomes of the isolates in Vietnam were compared against those in the United States (n = 209) and United Kingdom (n = 2), using an invasive isolate, M21273, as a reference (*5*,*6*). The phylogeny was inferred using the PubMLST-based Genome Comparator tool.

A subset of 81 genomes of the US NmNG UC isolates and 7 of invasive CC-11 as an outgroup was inferred with a time-measure phylogeny with BEAST v1.10 (*7*). A concatenated core alignment of 1,275,571 bps was generated through the genome-comparator and aligned with MAFFT. Subsequently, it was masked with Gubbins v2.2.1, leaving 1,081 bp of polymorphic sites (*8*). The Gubbins-masked alignment was generated using maximum-likelihood phylogeny with IQ-Tree, replicating 1,000 ultrafast-bootstrap (*9*).

To examine temporal signals, a root-to-tip linear regression was constructed with TempEst v1.5.3, estimating correlation coefficient was 0.91 and R^2 was 0.83 (*10*). A time-measured phylogeny was inferred by BEAST/BEAGLE, using the general time-reversible (GTR) substitution model and gamma heterogeneity site. The uncorrelated exponential relaxed clock (UCED), which allows each branch to evolve independently, was selected over the other molecular clocks, such as relaxed uncorrelated lognormal (UCLN), random local clock (RLC), and strict clock. The model was selected by using generalized stepping-stone sampling and estimation of the most recent common ancestor (TMRCA) of the subset. The marginal likelihood estimation was also used to evaluate the flexible nonparametric Skygrid model and other parametric models: constant, expansion, logistic, exponential growth, and GMRF Skyride. The Bayesian Skygrid was performed with 87 parameters corresponding to 88 sequences, and 17 grid points as the length of time from the newest to the root of the tree, running 250 million steps for two separate times.

Testing for Other Sexually Transmitted Infections (STIs)

Urethral swab specimens were collected following the hospital routine protocol to test for gonorrhea, *Chlamydia*, mycoplasma, and ureaplasma. From each participant 3 mL of venous blood was drawn to test for HIV and syphilis.

HIV was screened using a rapid test (Determine HIV-1/2, Alere Medical Co., Matsudo, Japan) at HHDV. Positive specimens were sent to Center for Disease Control of Ho Chi Minh City to confirm positivity with three different tests: Advia Centaur HIV Ag/Ab Combo (CHIV) Assay, Siemens Healthcare Diagnostics Inc., U.S.; SD. HIV 1/2 3.0, SD. Korea Co., Ltd, Korea; and Determine HIV, Alere Medical Co., Matsudo, Japan. Testubg followed the national HIV testing algorithm.

Gonorrhea, *Chlamydia*, mycoplasma, and ureaplasma were tested for using PCR (Panamax Viral DNA/RNA, PANAGENE Inc., Daejeon, Korea). Syphilis was screened for using rapid plasma reagin (RPR carbon kit, Lorne Laboratories Ltd, UK). Positive specimens were confirmed using the *Treponema pallidum* haemagglutination assay (TPHA Microtiter kit, Lorne Laboratories Ltd, UK) at HHDV.

Data Management and Analysis

All interview answer sheets were reviewed by the investigators for any missing information. These sheets were stored in locked cabinets in HHDV. Data were entered using Epi-Data version 3.1 (EpiData Association, Odense, Denmark), and all statistical analyses were conducted using Stata version 14.0 (StataCorp, Station, TX, USA). Continuous variables were described using mean, median and range. Categorical variables were presented as proportions.

Conditional univariate and multivariate logistic regressions were used to assess risk factors for US NmNG urethritis. Given the small sample size, we used forward selection to add in variables one by one: those variables in univariate analysis giving p values lower than the others were included in a multivariate model. A log likelihood-ratio test was used to select the better fit model between the former (without the add-in variable) and the current (with the add-in variable) models. If the test gave $p<0.1$, the newly added variable was retained in the model. If any two variables were thought to be highly correlated with each other, only one was included in multivariate analysis.

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Appendix Table 1. Coinfection with other sexually transmitted infections among 19 cases of United States *Neisseria meningitidis* urethritis clade*

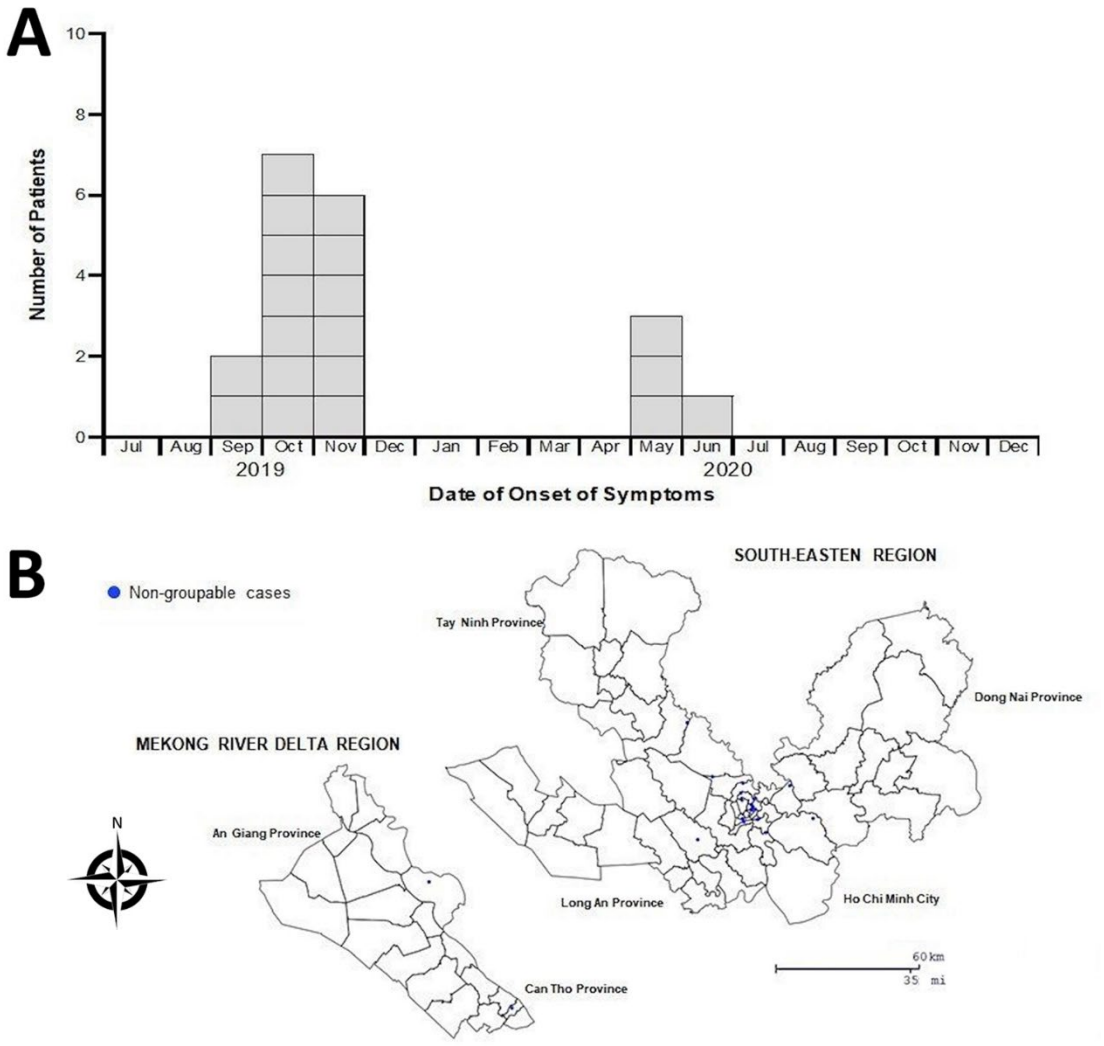
Symptoms	No STI coinfections, n = 12	STI coinfections, n = 7	p value†
Pyuria	0	2 (29)	0.12
Dysuria	10 (83)	3 (43)	0.13
Burning sensation during urination	1 (8)	1 (14)	1.0
Dripping	2 (17)	2 (29)	0.60
≥1 symptom	10 (83)	6 (86)	1.0

*Values are no. (%). STI, sexually transmitted infection.
†Fisher exact test

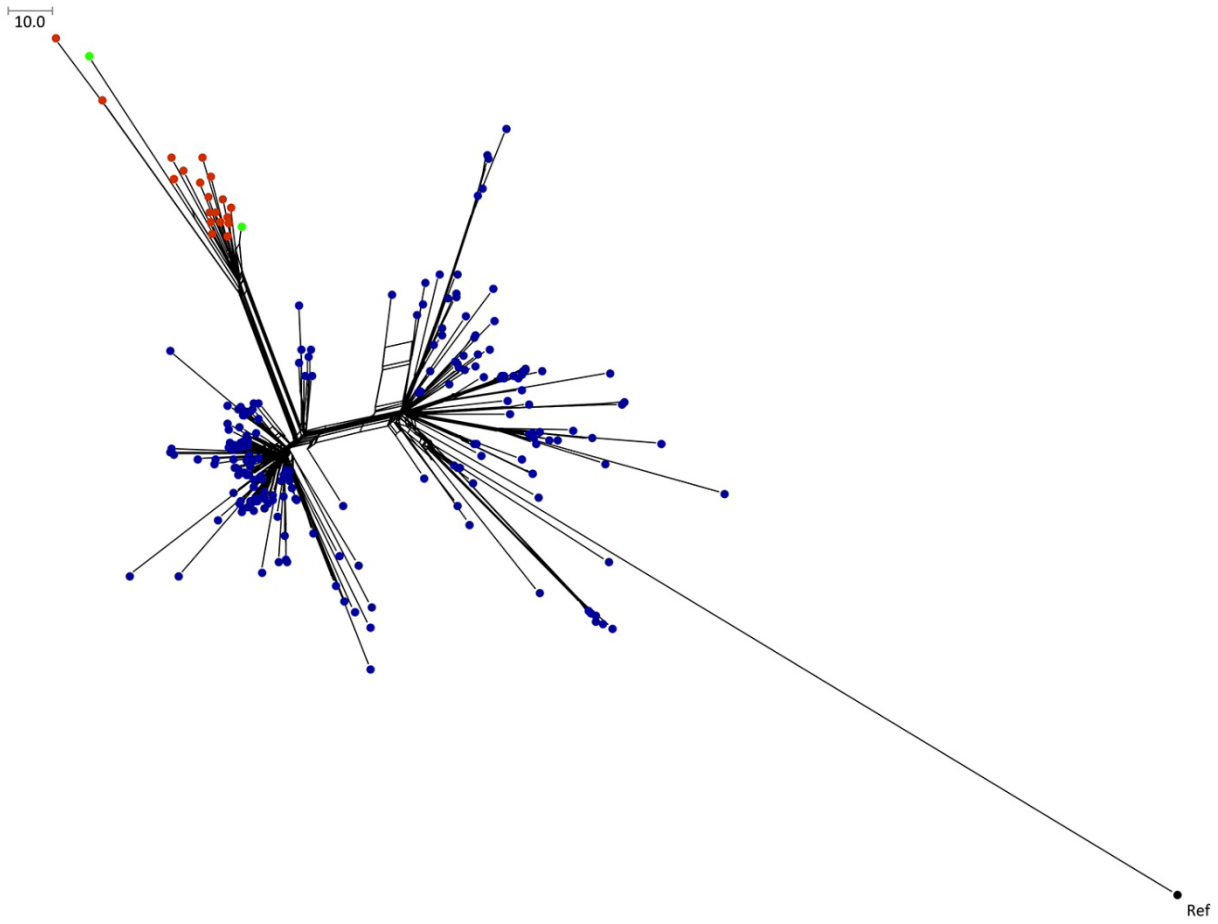
Appendix Table 2. Genome typing of isolates from 19 cases of United States *Neisseria meningitidis* urethritis clade, Vietnam*

Isolate no.	Region	Year	Serogroup / genegroup	ST	PorA type	FetA VR	PorB type	FHbp peptide	NadA peptide	NHbA peptide	*gyrA*	*penA*	*rpoB*
20149	Ho Chi Minh	2019	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	2	20	382†	316	8
20152	Dong Nai	2019	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	(–)‡	20	4	316	8
20153	An Giang	2019	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	(–)§	20	381†	316	8
20154	Tay Ninh	2020	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	218†	20	4	316	8
20155	Ho Chi Minh	2019	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	2	20	381†	316	8
20156	Ho Chi Minh	2019	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	(–)‡	20	4	316	8
20157	Ho Chi Minh	2019	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	2	20	381†	316	8
20158	Ho Chi Minh	2019	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	2	20	381†	316	8
20161	Ho Chi Minh	2019	NG/C	11	P1.5–1,10–8	F3–6	2–408†	896	2	20	4	316	8
20162	Long An	2019	NG/C	11	P1.5–1,10–8	F3–6	2–39	896	2	20	4	316	8
20163	Ho Chi Minh	2019	NG/C	11	P1.5–1,10–1	F3–6	2–2	896	2	20	381†	316	8
20164	Ho Chi Minh	2019	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	1	20	381†	316	8
20165	Ho Chi Minh	2019	NG/C	11	P1.5–1,10–8	F3–6	2–408†	896	2	20	4	316	8
20166	Ho Chi Minh	2020	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	2	20	381†	316	8
20167	Can Tho	2019	NG/C	11	P1.5–1,Δ	F3–6	2–2	896	2	20	381†	316	8
20168	Ho Chi Minh	2019	NG/C	12881	P1.5–1,10–8	F1–26	2–2	896	2	20	4	316	8
20414	Ho Chi Minh	2020	NG/C	11	P1.5–1,10–8	Inc.	2–2	896	2	20	4	316	8
20415	Ho Chi Minh	2020	NG/C	11	P1.5–1,10–8	F3–6	2–2	896	2	20	4	316	8
20416	Ho Chi Minh	2020	NG/C	11	P1.5–1,10–8	Inc.	2–2	896	2	20	4	316	8

*Inc., incomplete coding; NG, nongroupable; ST, sequence type.
†New allele.
‡Contains an inserted element
§Internal stop codon.



Appendix Figure 1. Number of cases per month and locations in an outbreak of sexually transmitted nongroupable *Neisseria meningitidis*–associated urethritis, Vietnam. A) Number of cases detected per month during 2019–2020. B) Locations of patient residences in Vietnam.



Appendix Figure 2. Phylogenic tree of isolates used to study an outbreak of sexually transmitted nongroupable *Neisseria meningitidis*–associated urethritis, Vietnam. The phylogenetic tree displayed the isolates in Vietnam (red dots) and the United Kingdom (green dots) forming a monophyletic clade with those from Ohio (blue dots), one of two clades of US NmNG urethritis. Scale bar indicates nucleotide substitutions per site.

Appendix Figure 3. Evolutionary plylogenetic tree of isolates from an outbreak of sexually transmitted nongroupable *Neisseria meningitidis*–associated urethritis, Vietnam. Phylogenetic tree was constructed using Baysian Skygrid model, performing with BEAST/BEAGLE v1.10.4 (https://beast.community/beagle), and displaying with FigTree v1.4.4 (http://tree.bio.ed.ac.uk/software/Figtree). Isolates from this outbreak are detailed in Figure 1. Scale bar indicates the time of evolutionary history.