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# Urethritis Caused by Novel *Neisseria* meningitidis Serogroup W in Man Who Has Sex with Men, Japan

To the Editor: We report a case of urethritis caused by a novel multilocus sequence type (ST), 10651, of the ST11/electrophoretic type (ET)–37 complex *Neisseria meningitidis* serotype W. The patient was a man who has sex with men. We also report on the patient's male partner, who was colonized with the same bacteria.

In March 2013, a 33-year-old Japanese man sought medical care at Shirakaba Clinic (Tokyo) after experiencing a urethral discharge for 4 days. The man was HIV positive (CD4 count 649 cells/mL) but was not receiving antiretroviral therapy. Physical examination showed a mucous urethral discharge. Gram staining of a sample revealed many gram-negative diplococci phagocytosed by polymorphonuclear leukocytes. Eleven days before seeking care, the patient had oral and anal intercourse with his male partner. A diagnosis of suspected urethritis caused by Neisseria gonorrhoeae was made, and a sample of the urethral discharge was sent for culture and testing (Strand Displacement Amplification) for N. gonorrhoeae and Chlamydia trachomatis. The patient was intravenously administered a single dose of ceftriaxone (1 g) (intramuscular administration of ceftriaxone is not approved in Japan). He was also given a single dose of azithromycin (1g orally) for possible *C. trachomatis* urethritis (1).

Six days after receiving treatment, the patient showed improvement. Results of the Strand Displacement Amplification test were negative for *N. gonorrhoeae* and *C. trachomatis*. Eight days after the patient received treatment, the culture for the urethral discharge sample was shown

to be positive for *N. meningitidis*. Urine culture was negative 20 days after treatment.

The 33-year-old male partner of the case-patient was originally from the United States and had been living in Japan for 4 years. Because of his history of sexual contact with the casepatient, he was advised to undergo a screening test for HIV and N. meningitidis. The man underwent a physical examination at our clinic 40 days after the case-patient received treatment; findings were unremarkable, and the result for HIV testing done 2 days earlier was negative. Throat and urine samples were obtained for culture, and the man was intravenously administered ceftriaxone (1 g). The urine sample culture was negative, but the throat sample culture was positive for N. meningitidis. The throat culture result was negative 10 days after the patient's treatment.

We performed cultures and tests to identify N. meningitidis, and we conducted multilocus sequence typing (MLST), serotyping, PorA typing, and pulsed-field gel electrophoresis (PFGE) as described elsewhere (2). Isolates from both men were identified as serotype W and PorA type P1.5, 2. MLST showed that the strains were ST10651 (genes analyzed: aroE:3, adk:4, fumC:3, gdh:8, pdhC:4, pgm:6, and abcZ:662). Although abcZ:662 was a novel allele, ST10651 belongs to the ST11/ET37 complex (3). We used PFGE with restriction enzyme NheI to compare the *N. meningitidis* strains from the case-patient and his partner; the isolates had the same PFGE pattern (Figure). Both isolates were confirmed to be a novel multilocus ST, 10651, of the ST11/ET37 complex; however, novel MLST types frequently occur. By using the E-test (Sysmex bioMérieux, Tokyo, Japan), we determined that the 2 isolates required the same minimum inhibitory concentrations (MICs) for the following antimicrobial drugs: penicillin (MIC 0.125 mg/L), ceftriaxone (MIC 0.004 mg/L), ciprofloxacin (MIC 0.004 mg/L), and azithromycin (MIC 0.25 mg/L) (4).

Urethritis caused by N. meningitidis infection in men who have sex with men (MSM) has been reported, as has an association between urethritis and oral sex (5,6). Most previously reported urogenital isolates of N. meningitidis have belonged to serogroups B (5,6), Y (5,6), and C (5). Among 115 cases of N. meningitidis infection in Japan during the last 9 years, 22 (19.1%) were caused by serogroup B and 18 (15.7%) were caused by serogroup Y; only 3 (2.6%) cases were caused by serotype W (7).

*N. meningitidis* ST11/ET37 complex is a hyperinvasive lineage. During the 1990s, the serogroup C ST11/ET37 complex was prominent in Europe

Marker Case 1 Case 2 Marker

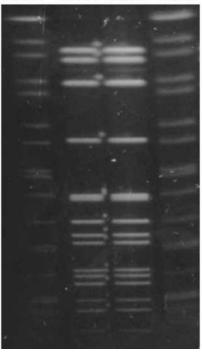


Figure. Pulsed-field gel electrophoresis (PFGE) patterns for *N. meningitidis* strains isolated from a man with urethritis (case 1) and his male sex partner (case 2), Japan. PFGE was performed with the restriction enzyme *Nhel*. Results showed the same PFGE pattern for both isolates. *Salmonella enterica* serovar Braenderup strain H9812 was used as the PFGE size marker strain; it was digested with *Xbal* and resolved by PFGE.

and North America. However, in 2000, an outbreak of N. meningitidis serotype W infections occurred among Hajj pilgrims ( $\delta$ ), and this serotype has now spread worldwide ( $3,\delta$ ).

Chemoprophylaxis is indicated for persons who have close contact with someone with invasive meningococcal infection (9), but there is uncertainty regarding the treatment of asymptomatic persons who have contact with someone with *N. meningitidis* urethritis. To avoid a reinfection cycle between the men in this study, we treated the asymptomatic, *N. meningitidis*—colonized male partner.

Since the early 2000s, and especially since 2012, outbreaks of invasive serogroup C, ST11/ET37 complex meningococcal disease causing high rates of death have been reported among MSM in the United States and Europe (10). These outbreaks have raised policy questions concerning vaccination recommendations for HIV-infected persons and for the MSM population (10). In Japan, meningococcal vaccination has not been officially approved, and neither of the men in this study had been vaccinated against *N. meningitidis*.

A diagnosis of urethritis is often based on Gram staining or nucleic acid amplification tests (1). However, Gram staining cannot differentiate N. meningitidis from N. gonorrhoeae, and amplification tests only detect N. gonorrhoeae. This practice makes it difficult to diagnose and access the number of cases of N. meningitidis urethritis.

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## Highly Pathogenic Avian Influenza A(H5N8) Virus from Waterfowl, South Korea, 2014

To the Editor: To date, 18 hemagglutinin (HA) subtypes and 11 neuraminidase (NA) subtypes have been identified in influenza A viruses (I-4). Influenza A viruses containing HA subtypes 1-16 circulate in aquatic birds (I,2), whereas those harboring HA subtypes 17 and 18 are found in bats (3,4).

On January 18, 2014, the government of South Korea reported an outbreak of highly pathogenic avian influenza A(H5N8) virus in breeding ducks in the southern part of Jeollabuk-Do Province (5). More than 12 million poultry have since been culled, but the spread of the virus continues in duck and chicken farms. We report the genetic characterization of this virus.

On February 15, 2014, a total of 200 fecal samples were collected from waterfowl in the Pungse River in Chungnam Province, which is geographically close to Jeollabuk-Do Province. All samples were inoculated into hens' eggs, and influenza A viruses were confirmed by PCR by using influenza A–specific nucleoprotein (NP) primers. We obtained 1 isolate, A/ waterfowl/Korea/S005/2014 (H5N8), and sequenced the full regions of all 8 genes as described (6). These sequences were deposited into GenBank under accession nos. KJ511809–KJ511816.

We conducted a BLAST search (http://blast.ncbi.nlm.nih.gov/Blast.cgi, http://platform.gisaid.org/epi3/frontend#4ead5c) to identify the closest gene sequences to those of A/waterfowl/Korea/S005/2014 (H5N8) (Table). Sequences for polymerase basic (PB) 2 (99% homology), HA (97% homology), and NP (99% homology) genes were closely related to those of A/wild duck/Shandong/

628/2011 (H5N1). Sequences for PB1 (99% homology), polymerase acidic subunit (PA) (98% homology), matrix (M) (99% homology), and nonstructural (NS) (99% homology) genes were closely related to those of A/duck/Jiangsu/1-15/2011 (H4N2). Sequences for the NA (98% homology) gene were closely related to that of A/duck/Jiangsu/k1203/2010 (H5N8). Phylogenic analysis showed that all 8 genes of A/waterfowl/Korea/S005/2014 (H5N8) belonged to the Eurasian lineage, and that the HA gene clustered with clade 2.3.4 (online Technical Appendix Figure 1, http://wwwnc.cdc.gov/EID/ article/20/9/14-0390-Techapp1.pdf).

We further analyzed the amino acid sequences of the virus isolate (online Technical Appendix Table 1). Positions 138 and 160 of the HA protein (H3 numbering) contained an alanine (A) residue, which was previously found to be related to enhanced binding to the human influenza receptor (7). The connecting peptide of HA contained an insertion of 4 basic amino acids (arginine-arginine-arginine-lysine), which is the same as in the HA of A/duck/Korea/Buan2/2014 (H5N8), an isolate from a duck farm in South Korea (GenBank accession no. KJ413839.1-KJ413846.1). Aspartic acid was found in M1 at position 30 and alanine at position 215; this combination has been connected with increased virulence in mice (8). The NS1 sequence contained serine at position 42, which is related to the enhanced pathogenicity in mice, but a truncation of the amino acids at positions 218-230 that has been linked with reduced pathogenicity in mice (9) was not identified. Asparagine was identified at position 31 of M2, which is the same in M2 of A/ duck/Korea/Buan2/2014 (H5N8) and confers resistance to amantadine and rimantadine (10).

Because all 8 genes of A/water-fowl/Korea/S005/2014 (H5N8) are closely related to those of the A/duck/