A Culture Collection of 50 Neisseria gonorrhoeae Isolates

H. Liu,a E. Vidyaprakash,a M. W. Schmerer,a D. C. Pham,a S. St Cyr,a E. N. Kersh a

*Division of STD Prevention, National Center for HIV/AIDS, Viral Hepatitis, STD, and TB Prevention, Centers for Disease Control and Prevention, Atlanta, Georgia, USA

ABSTRACT A culture collection of 50 Neisseria gonorrhoeae isolates is available from the CDC & FDA Antibiotic Resistance Isolate Bank. Associated data include antibiotic susceptibility information for azithromycin, cefixime, cefpodoxime, ceftriaxone, tetracycline, ciprofloxacin, penicillin, and spectinomycin and linked whole-genome sequences.

Antibiotic resistance is a major concern for clinical management of gonorrhea. The ability of Neisseria gonorrhoeae to shuffle and exchange its genes and to acquire antibiotic resistance is of particular global concern (1–3). At present in the United States, ceftriaxone in combination with azithromycin is the only first-line choice for treating uncomplicated N. gonorrhoeae infections (2, 4). Combating the spread of antibiotic resistance in N. gonorrhoeae requires an array of tools that include but are not limited to effective diagnostic tests and methods to improve treatment by detecting emerging resistance traits.

As part of the solution to combat antibiotic resistance, the CDC is providing a collection of N. gonorrhoeae strains (5–7). This panel consists of 50 isolates selected from the 2012 Gonococcal Isolate Surveillance Project (GISP) sentinel sites (8). N. gonorrhoeae strains in this panel have diverse antibiotic susceptibility profiles and were collected from various regions across the United States (Table 1). The antibiotics included in the profile are azithromycin, ceftriaxone, cefixime, ciprofloxacin, cefpodoxime, tetracycline, penicillin, and spectinomycin (7). Antimicrobial susceptibility testing (AST) was performed according to the Clinical and Laboratory Standards Institute (CLSI) M07 protocol (9, 10) and following Clinical Laboratory Improvement Amendments (CLIA) regulations.

To facilitate the development of genetic testing, the complete genome sequencing data for each isolate are also provided. DNA was extracted using the Promega genomic DNA purification kit (Promega, Madison, WI), and whole-genome sequencing was performed using a standard protocol; default parameters were used throughout (11). Specifically, libraries were prepared using the NEB genome library preparation kit (New England Biolabs, MA) and sequenced as 2 × 150-bp paired-end reads using the HiSeq 2500 platform (Illumina, CA). Raw sequence reads were analyzed in three stages. Preprocessing assessed the read quality with Trim Galore (v0.3.7), which contains FastQC and Cutadapt (12) to perform quality assessment, removal of duplicate reads, and trimming of reads. The genomes were assembled using SPAdes (v3.12.0) (13) and QUAST (v4.3) (14). Finally, annotation was completed using the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) (15, 16). Reads were mapped to the FA19 reference sequence (GenBank accession number CP012026). This N. gonorrhoeae isolate panel and associated AST data and genomic information are available on the website of the CDC & FDA Antibiotic Resistance Isolate Bank (https://www.cdc.gov/ARIsolateBank/Panel/PanelDetail?ID=11) (Table 1) (7).

The N. gonorrhoeae panel is provided as frozen isolates preserved in 300 to 500 μl of Trypticase soy broth (TSB) medium containing 20% glycerol and maintained at −70°C. To revive the frozen cultures, we recommend using a sterile cotton applicator.
| Antibiotic Resistance Isolate Bank no. | HHS region* | AST MIC (µg/ml) for?: | SRA accession no. (raw reads) | Genome assembly accession no. | No. of reads | N50 (bp) | GC content (%) | No. of contigs |
|---------------------------------------|-------------|----------------------|-------------------------------|-----------------------------|-------------|----------|---------------|---------------|
| 165                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 166                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 167                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 168                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 169                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 170                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 171                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 172                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 173                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 174                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 175                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 176                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 177                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 178                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 179                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 180                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 181                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 182                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 183                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 184                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 185                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 186                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 187                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 188                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 189                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 190                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 191                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 192                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 193                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 194                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 195                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 196                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 197                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 198                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 199                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 200                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 201                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 202                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 203                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 204                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 205                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 206                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 207                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 208                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 209                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 210                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 211                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 212                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 213                                   |             |                     | SRAV000000000                |                             |             |          |               |               |
| 214                                   |             |                     | SRAV000000000                |                             |             |          |               |               |

*Department of Health and Human Services (HHS) region [https://www.hhs.gov/about/agencies/eea/regional-offices/index.html].

b AZI, azithromycin; CFX, cefixime; CPD, cefpodoxime; CRO, ceftriaxone; TET, tetracycline; CIP, ciprofloxacin; PEN, penicillin; SPE, spectinomycin; S, susceptible.

or an inoculating loop to transfer a small amount (e.g., 50-µl equivalent) of the frozen culture onto an N. gonorrhoeae medium base supplemented with 1% IsoVitalex® or a chocolate II agar plate (BBL/Becton, Dickinson) following standard microbiological procedures. The plates should be incubated at 36°C ± 1°C for 20 to 24 h in a humidified chamber supplemented with 5% CO2.

Data availability. The GenBank and SRA accession numbers for the isolates are provided in Table 1. To acquire the panel, or for questions or suggestions, please...
contact the CDC & FDA Antibiotic Resistance Isolate Bank directly (https://wwwn.cdc.gov/ARIsolateBank) (7).

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