

Supplementary Appendix

Bacterial culture and characterisation

Samples were cultured at the Leeds Teaching Hospitals NHS Trust microbiology laboratory using selective VCNT agar (E&O), incubated in 5-10% CO₂ at 35-37°C. *N. gonorrhoeae* colonies were identified using morphology, Gram stain, oxidase positivity and MALDI-TOF identification. Antimicrobial susceptibility testing was performed by inoculating a 0.5 McFarland suspension in brain-heart infusion broth (E&O) onto GC agar base with 1% Vitox (Oxoid), and applying diffusion strips (E-test, BioMérieux) to determine the isolate's minimum inhibitory concentration (MIC). MIC for ceftriaxone and azithromycin were tested for all isolates; those non-susceptible to either antibiotic were tested for sensitivity to tetracycline, ciprofloxacin and spectinomycin. Isolates with azithromycin MIC = 0.5 mg/L were considered to have intermediate susceptibility to azithromycin. Isolates resistant to ceftriaxone (MIC > 0.125 mg/L) or azithromycin (MIC > 0.5 mg/L) were sent to the Public Health England reference laboratory for confirmation.

Clinical data collection

Clinical data was collected on all cases in WGS reports, including gender, sexual orientation, age, dates of presentation and symptom onset if any, anatomical sites tested, results of NAATs, cultures and antimicrobial susceptibility testing, treatment given, dates of treatment, and test of cure results when available. PN data collected included date of PN discussion, number of reported partners, partner types (casual or regular), partner genders, the availability of identifying information, whether partners had verified attendance at a sexual health clinic in Leeds or elsewhere, and results of tested partners when available.

24 Whole genome sequencing

25 DNA was extracted using the QIAamp 96 DNA QIAcube HT Kit (Qiagen) following
26 manufacturer's instructions. Sequencing libraries were prepared using Nextera XT (Illumina),
27 and WGS was performed on Illumina MiSeq instruments with 75-bp paired-end reads. Sequence
28 reads were assembled and analyzed using a previously described pipeline for bacterial
29 pathogens(1, 2). Sequences were compared using single nucleotide polymorphisms (SNPs)
30 obtained from recombination-adjusted maximum likelihood phylogenies. Reads were mapped
31 with Stampy(3) (version 1.0.20, without Burrows-Wheeler Aligner pre-mapping, using an
32 expected substitution rate of 0.01) to the *Neisseria gonorrhoeae* NCCP11945 reference genome
33 (NCBI Reference Sequence: NC_011035.1). Samples were compared using single nucleotide
34 polymorphisms (SNPs) identified with bcftools mpileup(4) (version 1.9) with the extended base-
35 alignment quality flag (bcftools mpileup -Q25 -q30 -E -o40 -e20 -h100 -m2 -F0.002). A
36 consensus of $\geq 75\%$ was required to support a SNP, and calls were required to be homozygous
37 under a diploid model. Only SNPs supported by ≥ 5 reads, including one in each direction were
38 accepted. SNPs were not called in repetitive regions of the genome identified by using
39 BLAST(5) to search for repeat regions >100 base pairs in length. Filtering rules were based on
40 sequencing of technical replicates of bacterial genomes using the same DNA pool, e.g. in(1),
41 including visual inspection of alignments and chosen to keep the false positive SNP rate to
42 around 1 per 100Mb of genome sequenced. An example workflow implementing the variant
43 calling can be found here - <https://github.com/davideyre/bug-flow>.

44

45 Sequences where $<70\%$ of the reference sequence was mapped to were excluded from the
46 analysis. To improve computational efficiency in identifying closely related sequences,

47 sequences within ≤ 500 SNPs of any other sequence were initially pooled into groups. All
48 variable sites within each group were then identified, sequences with $< 70\%$ of these variable
49 sites called (either as wild type or variant) were excluded from further analysis, as these
50 represent possible cross-contamination amongst closely related samples in the same sequencing
51 batch. Within each group, variable sites that were not called (either as wild type or variant) in
52 $\geq 70\%$ of samples were also excluded from further analysis, as such sites may represent either
53 regions of the genome that are difficult to identify with certainty from short-read sequencing or
54 regions that are variably present or absent.

55

56 Multi-locus sequence types (MLSTs) were determined using mlst
57 (<https://github.com/tseemann/mlst>; version 2.19.0) with mapped alignments.

58

59 Sequence comparisons

60 For each group of sequences within ≤ 500 pairwise observed SNPs, initial maximum likelihood
61 phylogenetic trees were constructed using PhyML (version 3.0)(6), using a generalised time
62 reversible substitution model, and the “BEST” tree topology search operation option. These trees
63 were then adjusted to remove recombining regions using ClonalFrameML (version 1.25)(7),
64 using default parameters. Each recombination adjusted phylogenetic tree was used to determine
65 the number of SNPs between all pairs of sequences (i.e. the patristic distance between them). An
66 example implementation of this approach can be found here -

67 <https://github.com/davideyre/runListCompare>.

68

69 The initial screening threshold of ≤ 500 observed SNPs was chosen to ensure tree construction
70 was computationally efficient for real-time reporting, but still placed together genomes related
71 by up to 20 SNPs after correction for recombination and any missing data. At the end of the
72 study, we also repeated the phylogenetic analysis combining all genomes into a single tree. Of
73 the 1285 pairs of genomes in the study within 20 SNPs in the tree with all genomes considered,
74 only 4 pairs (all involving the same genome) do not fall within the same 500 SNP cluster, i.e.
75 99.7% are correctly placed in a cluster using our approximation.

76

77 WGS data usage and analysis


78 Data from WGS reports were compiled weekly by a trained research assistant to form a database
79 of growing clusters of patients with isolates within 20 SNPs. These were examined weekly
80 alongside PN data and discussed with the clinical team. General characteristics about clusters
81 were noted by the clinical team, such as number of cases within clusters and characteristics of
82 these cases (e.g. bridging between MSM and heterosexuals, mixing of patients with discordant
83 HIV serostatuses). Links between individuals with PN and WGS concordance were also easily
84 observed with clusters. WGS links sometimes led to previously unidentified PN links as
85 described in the manuscript, which prompted discussion on the appropriateness of potential
86 further questions for the patients. Formal analysis, including application of the transmission
87 nomogram categories, occurred at the end of the study period when all data were available
88 (Supplementary Figure 2).

89


90 Data sharing

91 Sequences generated during this study can be found on the NCBI short read archive under
92 BioProject PRJNA689969. Epidemiological data on links between cases are available to
93 accredited researchers from the University of Leeds (via the corresponding author) subject to an
94 application with a specific research question and setting up a data sharing agreement.

95 **Supplementary Figure 1. Example of WGS report.** Identifiers have been redacted.



UNIVERSITY OF OXFORD



UNIVERSITY OF LEEDS

Neisseria gonorrhoeae real-time sequencing Report

A National Institute for Health Research Health Protection Research Unit Project

Report version: 1.7
Reference genome: R00000419

Sample details

Local LIMS specimen identifier:		Collection Date:	
Sequence GUUID:		Sequencing date:	
Sequence plate name:		Report date:	

Sequencing quality

Total reads (millions):	9.8	[Expected value 1.0-20.0]	Pass
Reads mapped (%):	93.8	[Expected value 85.0-99.0%]	Pass
Reference genome coverage (%):	89.9	[Expected value 85.0-95.0%]	Pass

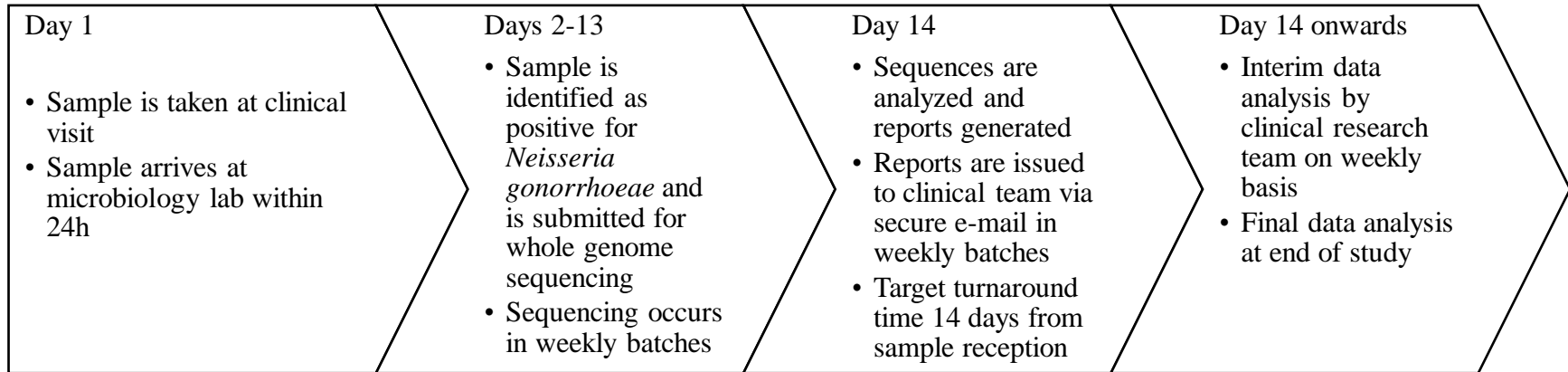
Samples should meet expected values to ensure high precision comparison of samples.

Related samples

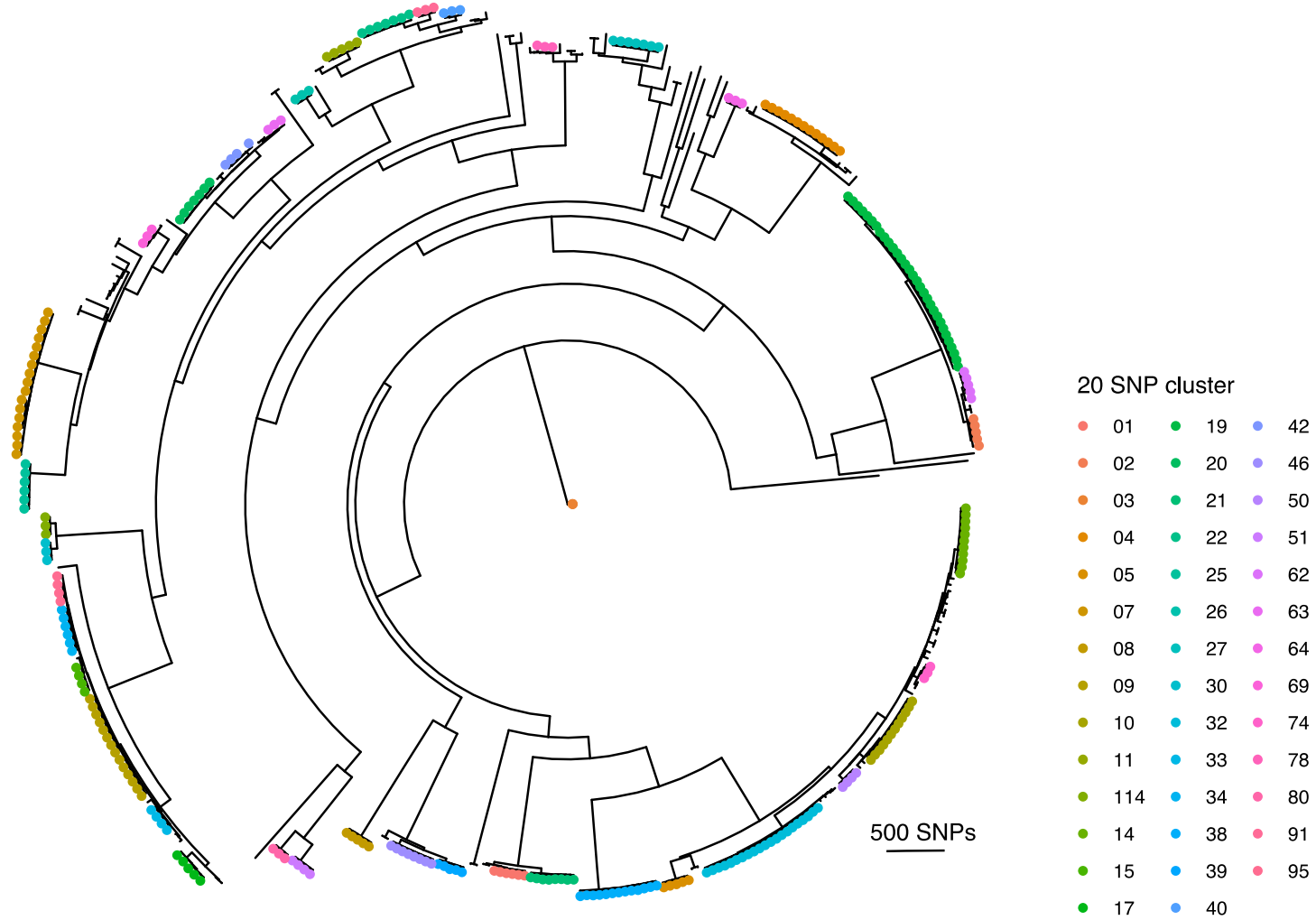
Samples have been compared with all previously sequenced samples. Samples related within 20 single nucleotide variants (SNPs) are shown below, these samples are compatible with being a potential source for the sequenced case in the last three months. More closely related samples are genetically more likely to be the source of the sequenced case.

Local specimen identifier	Collection date	SNPs	Sample location	Sequencing plate
	28-Jun-2018	0	Leeds	LD2426_Ngon_L
	29-May-2018	0	Leeds	LD2422_Ngon_L
	15-Jun-2018	0	Leeds	LD2421_Ngon_L

Supplementary Figure 2. Target timeline of sampling, sequencing, reporting and data analysis.



Supplementary Figure 3. Recombination corrected maximum likelihood phylogeny of 380 *N. gonorrhoeae* genomes sequenced during the study. Each cluster of sequences within 20 SNPs and containing ≥ 3 cases is shown as a different colour. Note all SNP clusters form a monophyletic clade.



Supplementary Table 1. Time, in days, from sample collection to time points in the sequencing pipeline.

	Sample reception in microbiology laboratory	Isolate submission for sequencing	DNA extraction	Successful sequencing	Report generation and issuance to clinical team
Mean	0	5	9	13	15
Range	0-2	1-16	3-20	4-176	6-181
SD	0	2	3	14	15
Median	0	4	8	11	13
IQR	0	3	5	6	6

SD, standard deviation

IQR, interquartile range

Supplementary Table 2. Sample list and patient, sample characteristics. Multi-locus sequence types (MLSTs) for each genome are provided. MLSTs for genomes with a novel or indeterminate sequence type are shown as a dash.

	Sample ID	Age	Gender	Anatomic site of isolation	Ceftriaxone	Azithromycin	MLST
1	8aff0324	23	F	Cervix	S	S	8130
2	e2bcb2ba	61	M	Urethra	S	R	9363
3	8e705c68	48	M	Rectum	S	I	11982
4	15e12648	29	M	Rectum	S	S	13292
5	ea0049c9	25	T	Rectum	S	S	8122
6	27e5df50	44	M	Rectum	S	S	1599
7	9c63ec34	55	M	Rectum	S	I	13292
8	6d41839b	28	M	Urethra	S	S	-
9	54fe3f60	35	M	Rectum	S	S	7359
10	28101e46	25	F	Cervix	S	S	1580
11	f0a17eb3	30	M	Urethra	S	S	13292
12	af3847e9	27	M	Urethra	S	S	7359
13	ceb6f99b	25	M	Urethra	S	S	7359
14	bf46fd5a	28	M	Rectum	S	S	1893
15	13ada6ed	23	F	Cervix	S	S	1594
16	f55a2392	29	F	Cervix	S	R	9363
17	f7a0608d	27	F	Cervix	S	S	1594
18	c73914c7	22	F	Cervix	S	S	13266
19	e38bbeac	24	M	Urethra	S	S	1594
20	22125dab	42	M	Rectum	S	S	7359
21	0c97a9f9	22	M	Urethra	S	S	8156
22	611b6cb5	23	M	Urethra	S	S	1594
23	5169f1c0	25	M	Urethra	S	S	8156
24	3374a73a	33	M	Rectum	S	S	8156
25	628d466b	22	M	Pharynx	S	I	10314
26	01a7dcdd	31	M	Urethra	S	S	8143
27	5c8790bc	30	M	Urethra	S	R	9363
28	0cf2f19d	26	M	Rectum	S	S	-
29	6d305ad9	27	F	Cervix	S	S	1594
30	56aab0b9	20	F	Cervix	S	S	1587
31	b29edf1a	22	F	Cervix	S	S	1599
32	9388a68b	24	F	Cervix	S	I	7363
33	e82f1d00	22	M	Urethra	S	S	1599

34	53e5f7ba	29	M	Rectum	S	S	8156
35	b597bbd0	24	M	Urethra	S	R	9363
36	b22f1d56	62	M	Rectum	S	S	1599
37	fe42e4de	24	F	Cervix	S	S	1594
38	7d1ed152	21	F	Cervix	S	S	-
39	f2cfe5e9	18	F	Cervix	S	S	-
40	cf668e23	30	M	Urethra	S	S	11233
41	c05efa52	23	M	Rectum	S	S	1599
42	03035246	23	M	Urethra	S	S	8156
43	92a0e245	38	M	Urethra	S	S	1893
44	734df9e7	38	M	Urethra	S	S	1893
45	c9a392b1	29	M	Rectum	S	S	7359
46	71353f93	53	M	Urethra	S	S	8130
47	7f8e2b8a	33	F	Cervix	S	S	11990
48	1b5be167	43	M	Rectum	S	S	1583
49	5ee2393a	59	M	Urethra	S	I	1579
50	be26d39f	44	F	Cervix	S	S	8122
51	989b1b6f	34	M	Urethra	S	S	7359
52	6fa1e39a	34	M	Urethra	S	S	7822
53	a283da7c	66	M	Rectum	S	S	1599
54	ba32bd74	37	M	Urethra	S	S	1599
55	df35ae34	47	M	Urethra	S	R	8156
56	b6e1934d	36	M	Urethra	S	S	1599
57	f0f84660	45	M	Urethra	S	S	11990
58	17ea9767	28	M	Rectum	S	S	1599
59	f38ceb7d	35	M	Rectum	S	S	8156
60	e5b8a6f4	69	M	Rectum	S	S	1599
61	9cc59ce5	31	M	Rectum	S	S	1599
62	cc92dcf1	54	F	Cervix	S	S	-
63	86b7993a	26	M	Urethra	S	S	11990
64	d39bc322	51	M	Rectum	S	S	1599
65	93b05c47	28	M	Urethra	S	S	13266
66	cadde8b7	29	M	Urethra	S	S	8135
67	53fd8960	31	M	Pharynx	S	S	1599
68	4f3bae6c	36	M	Rectum	S	S	10314
69	6fb94345	62	M	Rectum	S	S	1599
70	0c2ec44a	40	M	Pharynx	S	S	1599
71	b2a152d2	39	M	Rectum	S	S	8156
72	e5275b8b	28	M	Rectum	S	S	1599

73	ac83cd9f	37	M	Urethra	S	S	1594
74	1670bc9c	25	F	Cervix	S	S	8156
75	8d85a975	32	F	Cervix	S	S	11990
76	eaca36ba	23	F	Cervix	S	S	1594
77	b00879f5	32	F	Cervix	S	S	1594
78	0c012e24	48	F	Cervix	S	S	7363
79	6b3c2772	23	F	Cervix	S	S	7363
80	3b647d52	25	F	Cervix	S	S	11999
81	7b896ace	21	F	Cervix	S	S	1594
82	a40b4ef7	23	F	Cervix	S	S	8135
83	091b305d	33	F	Cervix	S	S	1594
84	05e85f3d	19	F	Cervix	S	S	1580
85	20a09ce2	21	F	Cervix	S	S	11999
86	cc12eda7	22	F	Cervix	S	S	1587
87	286bb162	20	F	Cervix	S	S	1594
88	0b073637	27	F	Cervix	S	S	1580
89	16bb251e	22	F	Cervix	S	S	1580
90	aebb33ef	19	F	Cervix	S	S	1594
91	ea6e4973	19	F	Cervix	S	S	8156
92	f519a46c	17	F	Cervix	S	S	1594
93	28119cbd	19	F	Cervix	S	S	1594
94	edbc9970	17	F	Cervix	S	S	7363
95	14b0f57d	18	F	Cervix	S	S	1594
96	6a5ba051	23	F	Cervix	S	I	8156
97	9fce541e	19	F	Cervix	S	S	1594
98	5c4e8895	31	F	Cervix	S	S	1594
99	5bc0d27c	20	F	Cervix	S	S	1594
100	725135a6	19	F	Cervix	S	S	8156
101	f1767d86	17	F	Cervix	S	S	8163
102	d5dda33b	20	F	Cervix	S	S	-
103	410808b8	23	F	Cervix	S	S	1594
104	d38f27e5	21	F	Cervix	S	S	1594
105	58453e6b	18	F	Cervix	S	S	7363
106	4bd2749e	21	F	Cervix	S	S	1594
107	a77bcd03	17	F	Cervix	S	S	1594
108	9fbc19df	19	F	Cervix	S	S	-
109	c4172773	19	F	Cervix	S	S	8163
110	e938f8be	19	F	Cervix	S	R	9363
111	d748e033	22	F	Cervix	S	S	1594

112	f1884728	19	F	Cervix	S	S	1594
113	a6ad47ab	20	F	Pharynx	S	S	7363
114	0940a144	18	F	Cervix	S	S	11990
115	24db4d10	30	F	Pharynx	S	S	1893
116	5c10624b	20	F	Cervix	S	S	1595
117	1c4b89af	18	F	Cervix	S	S	8156
118	4a8f374e	21	F	Cervix	S	S	1595
119	5bff891e	17	F	Cervix	S	S	1595
120	a58c4e63	17	F	Cervix	S	S	1594
121	2bd79b44	21	F	Cervix	S	S	13532
122	f1c6e0df	33	F	Cervix	S	S	1901
123	93e33740	16	F	Cervix	S	S	1594
124	0aa8fe4e	16	F	Cervix	S	S	11990
125	67fbda3a	20	F	Cervix	S	S	8163
126	23f4c853	15	F	Cervix	S	S	1594
127	eeff6987	19	F	Cervix	S	S	-
128	96d9ac30	21	F	Cervix	S	S	11990
129	8ffb5782	16	F	Cervix	S	S	1594
130	f3c6e333	17	F	Cervix	S	S	1594
131	e265e9d9	32	F	Cervix	S	S	8135
132	938b9cc6	16	F	Pharynx	S	S	11990
133	1a9ca4f8	26	F	Cervix	S	S	1594
134	f867f0dd	19	F	Cervix	S	S	11990
135	6c07eb8c	16	F	Cervix	S	S	11990
136	3da7a71e	19	F	Cervix	S	S	7363
137	94d88d9b	14	F	Cervix	S	S	11990
138	fd3b08c3	14	F	Cervix	S	S	11990
139	e2fa5159	18	F	Cervix	S	S	1594
140	eee2e74f	16	F	Cervix	S	S	8135
141	ac907ae6	39	F	Cervix	S	S	8135
142	d617feb1	20	F	Cervix	S	S	1580
143	762ff642	26	F	Cervix	S	S	8135
144	6043531f	25	F	Pharynx	S	I	11428
145	8533f6fc	21	F	Cervix	S	S	7363
146	94df0281	18	F	Cervix	S	S	8126
147	f28345ea	33	F	Cervix	S	S	-
148	955fcde3	17	F	Cervix	S	S	1594
149	347b4bae	21	F	Cervix	S	S	1599
150	20e2a3a3	18	F	Cervix	S	S	1594

151	c7d43fc6	20	F	Cervix	S	I	13292
152	86a8a29d	28	F	Cervix	S	S	1594
153	942e6ece	22	F	Cervix	S	S	11990
154	8919bf14	41	F	Cervix	S	S	7363
155	3b719f1c	46	F	Cervix	S	I	8156
156	089f47b9	20	F	Cervix	S	S	1599
157	01418afc	19	F	Cervix	S	S	1580
158	576f2f18	19	F	Cervix	S	S	1594
159	f39e0926	21	F	Pharynx	S	S	1580
160	c1f4ad17	23	F	Cervix	S	S	8135
161	ca7b5d2d	19	F	Cervix	S	S	13333
162	b9c439ef	20	F	Cervix	S	S	8122
163	28256dc1	20	F	Cervix	S	S	8122
164	82787783	17	F	Cervix	S	S	1922
165	6bb906c4	20	F	Pharynx	S	S	1580
166	0e79eb59	18	F	Cervix	S	S	1580
167	4636552b	17	F	Pharynx	S	R	8163
168	d5fab5c6	31	F	Cervix	S	S	1587
169	55e85bdd	17	F	Cervix	S	S	1594
170	29257526	21	F	Cervix	S	S	7359
171	925a4b41	19	F	Cervix	S	R	1901
172	20ca16bf	19	F	Cervix	S	R	1901
173	f656394d	16	F	Cervix	S	S	1594
174	591e55d3	19	F	Cervix	S	S	8163
175	e243a5ba	16	F	Cervix	S	S	11990
176	6778f7aa	32	F	Cervix	S	S	14422
177	a3b0978b	16	F	Rectum	S	S	11990
178	ec69f221	19	F	Cervix	S	S	1580
179	83b40cf4	19	F	Cervix	S	S	1580
180	724ff8fe	26	M	Urethra	S	S	1594
181	fb49e193	26	M	Urethra	S	S	7827
182	46e8c9f8	27	M	Urethra	S	S	1594
183	a4a77494	24	M	Urethra	S	S	7363
184	de22c472	27	M	Urethra	S	S	-
185	abc9e9de	25	M	Urethra	S	S	11990
186	de22d8ce	19	M	Urethra	S	S	7363
187	2b2bbcff	20	M	Urethra	S	S	1599
188	541bdeaf	19	M	Urethra	S	S	11990
189	70158e5c	21	M	Urethra	S	S	11990

190	1ace30e4	25	M	Rectum	S	S	13956
191	6f936f8e	21	M	Urethra	S	S	8156
192	6a0300ba	19	M	Urethra	S	S	11990
193	15b2b6da	25	M	Urethra	S	S	8156
194	31a6225d	25	M	Pharynx	S	S	7359
195	feb77314	17	M	Urethra	S	S	7827
196	796dbeba	19	M	Urethra	S	S	11990
197	9b6eb57d	21	M	Urethra	S	S	8163
198	1a85307f	26	M	Rectum	S	S	1599
199	679e9e63	21	M	Urethra	S	S	-
200	f8ab47e0	21	M	Urethra	S	S	8163
201	4012d55d	23	M	Urethra	S	S	10314
202	359775e1	41	M	Rectum	S	S	1599
203	a888feb7	27	M	Rectum	S	S	8156
204	1cf4a7e0	40	M	Rectum	S	S	1583
205	169a53a7	40	M	Rectum	S	R	9363
206	e93d2234	26	M	Urethra	S	S	1599
207	1c2f960f	28	M	Urethra	S	S	13292
208	14437ace	22	M	Urethra	S	S	-
209	0c81c6cc	35	M	Rectum	S	S	10314
210	fd8f208c	25	M	Rectum	S	R	9363
211	8ba875fc	34	M	Urethra	S	S	8111
212	dff2e907	51	M	Urethra	S	I	7822
213	f2206083	23	M	Urethra	S	S	1599
214	64050aac	21	M	Urethra	S	R	1580
215	4e8d6f90	30	M	Urethra	S	S	1599
216	ccaf079e	30	M	Urethra	S	S	7365
217	03b53fd9	22	M	Pharynx	S	I	11982
218	02586e6e	21	M	Urethra	S	S	1594
219	71452d1b	23	M	Urethra	S	S	1583
220	7bbdbbd4	47	M	Rectum	S	S	8156
221	5f6a8611	27	M	Pharynx	S	S	1599
222	6463bcbf	31	M	Rectum	S	I	11982
223	5fd08588	43	M	Rectum	S	S	10314
224	611f2dcd	39	M	Urethra	S	S	13292
225	b06ec95c	77	M	Rectum	S	S	13292
226	aee61044	51	M	Urethra	S	S	8143
227	40ba057a	27	M	Urethra	S	S	7359
228	a81ffd17	23	M	Urethra	S	I	8156

229	c66eddb0	20	M	Rectum	S	S	9362
230	55c69c0f	25	M	Pharynx	S	S	1583
231	02b01f98	24	M	Urethra	S	S	7363
232	12f4ced1	20	M	Rectum	S	S	13292
233	fac4e516	16	M	Urethra	S	S	11990
234	8f8558aa	39	M	Pharynx	S	S	10314
235	9238a499	20	M	Urethra	S	S	8143
236	c6e951f9	20	M	Pharynx	S	S	-
237	0af02bf6	17	M	Urethra	S	S	11999
238	e317e4bd	38	M	Urethra	S	S	8156
239	75ee7267	38	M	Rectum	S	S	8156
240	261714ef	24	M	Rectum	S	S	8156
241	c7623c0d	21	M	Urethra	S	S	1599
242	c2193bd6	27	M	Rectum	S	S	1583
243	7c4cff00	27	M	Rectum	S	I	8156
244	45d190ce	27	M	Rectum	S	S	8156
245	7109bcc3	21	M	Urethra	S	S	7359
246	e2ff2857	27	M	Urethra	S	S	7822
247	cf624ecd	30	M	Pharynx	S	S	1583
248	d9d2cf50	23	M	Rectum	S	S	1893
249	fd699fe3	50	M	Urethra	S	S	1583
250	b31cfba1	21	M	Rectum	S	S	7822
251	8e160144	58	M	Urethra	S	S	13292
252	87be7d1b	29	M	Urethra	S	S	10314
253	621730e5	29	M	Urethra	S	S	10314
254	2405ef7b	29	M	Urethra	S	S	10314
255	caccb96c	19	M	Rectum	S	S	10314
256	5b8c23c8	23	M	Urethra	S	S	8143
257	d36288f9	20	M	Urethra	S	I	13292
258	686f1089	20	M	Pharynx	S	S	7363
259	d28db95f	25	M	Urethra	S	I	1579
260	312b8543	32	M	Pharynx	S	I	10314
261	662caa72	32	M	Rectum	S	I	8156
262	bc997cda	31	M	Urethra	S	S	1599
263	72101c22	17	M	Urethra	S	S	8156
264	d706f28a	21	M	Rectum	S	S	7822
265	c9b7d839	21	M	Rectum	S	S	7822
266	56f4065d	20	M	Urethra	S	S	1580
267	29801fed	26	M	Urethra	S	S	1599

268	65ea76fe	25	M	Urethra	S	S	1599
269	8f0d686d	45	M	Pharynx	S	S	8156
270	65fbb40f	21	M	Urethra	S	S	8156
271	456f2b45	30	M	Urethra	S	S	11990
272	07181853	19	M	Rectum	S	S	8143
273	5fd48ce6	19	M	Rectum	S	S	8156
274	cf6050ac	19	M	Pharynx	S	S	7359
275	bf0d96be	26	M	Urethra	S	I	11982
276	67ae6afc	28	M	Urethra	S	S	8156
277	a9b7eabe	19	M	Urethra	S	S	7363
278	6fcbfb23	29	M	Rectum	S	S	7359
279	be513ef5	21	M	Rectum	S	S	7359
280	a1fe16c4	20	M	Rectum	S	I	8156
281	85267c51	30	M	Rectum	S	S	11233
282	e23a62c5	26	M	Urethra	S	S	8143
283	30498b86	53	M	Urethra	S	S	8135
284	a7fac6ad	22	M	Urethra	S	S	8156
285	c4aee009	20	M	Rectum	S	S	8143
286	e5e37173	19	M	Pharynx	S	S	8156
287	64df8314	53	M	Urethra	S	S	8156
288	23ba1809	20	M	Urethra	S	S	8126
289	f2ddd5ca	17	M	Urethra	S	S	11255
290	3d13dce4	21	M	Urethra	S	S	1599
291	062261af	38	M	Urethra	S	S	8135
292	6c6cfe2d	19	M	Urethra	S	I	1580
293	fce817da	22	M	Urethra	S	S	1587
294	ddc526c7	29	M	Urethra	S	S	8156
295	cd480f2f	31	M	Pharynx	S	S	7359
296	84f02cf9	59	M	Rectum	S	S	1584
297	1304ff3e	29	M	Urethra	S	S	1599
298	3f4e9e40	21	M	Urethra	S	S	11999
299	bd030f9b	27	M	Urethra	S	S	13292
300	432f816d	27	M	Urethra	S	S	8156
301	aa2519e3	20	M	Urethra	S	S	7363
302	162c469e	17	M	Urethra	S	S	1599
303	36ca994c	24	M	Urethra	S	I	13292
304	93b5d069	24	M	Pharynx	S	I	13292
305	24e2e1c2	34	M	Urethra	S	S	1599
306	34b8b14b	22	M	Urethra	S	S	7363

307	02370a7c	23	M	Urethra	S	S	8156
308	af29d630	28	M	Urethra	S	S	1594
309	f999e919	32	M	Rectum	S	S	13292
310	91683878	20	M	Rectum	S	S	7359
311	09475152	37	M	Urethra	S	S	8135
312	de379bcf	22	M	Urethra	S	S	1580
313	66a60207	22	M	Urethra	S	S	1594
314	fe6f284a	18	M	Urethra	S	S	1594
315	f52c5435	21	M	Pharynx	S	S	1599
316	b90ca45f	26	M	Rectum	S	S	-
317	c93dd951	22	M	Urethra	S	S	11990
318	0d442e5a	21	M	Urethra	S	S	-
319	16702412	23	M	Urethra	S	S	11990
320	cbf10bad	59	M	Urethra	S	S	1599
321	e7e82af2	22	M	Urethra	S	S	14283
322	90e1f75d	35	M	Urethra	S	S	1595
323	54fe002e	28	M	Urethra	S	S	10314
324	b344bfac	53	M	Urethra	S	S	1599
325	c442ff8e	23	M	Urethra	S	S	11255
326	08b3c51a	22	M	Urethra	S	S	11990
327	196399a4	24	M	Urethra	S	S	7822
328	645a9b10	18	M	Urethra	S	S	11990
329	ae3c77c7	26	M	Urethra	S	S	1588
330	b106ffea	22	M	Urethra	S	S	1599
331	54958d7a	41	M	Urethra	S	S	1599
332	59e4a70f	30	M	Urethra	S	S	1603
333	7c323282	26	M	Urethra	S	S	9362
334	72eceff3	32	M	Urethra	S	S	13413
335	30d9fc89	37	M	Urethra	S	S	8135
336	1abeb22c	22	M	Urethra	S	S	14394
337	01dad278	18	M	Urethra	S	S	1594
338	ebe1cbbc	32	M	Urethra	S	S	1893
339	c7fdcc6	21	M	Urethra	S	S	8156
340	795939f1	20	M	Urethra	S	R	1580
341	4df1d64f	25	M	Urethra	S	S	13292
342	3ee88c2c	21	M	Urethra	S	S	11990
343	43699704	25	M	Urethra	S	S	1594
344	7631e69c	20	M	Urethra	S	I	11982
345	3dfe7446	19	M	Urethra	S	S	-

346	7ee619e6	21	M	Urethra	S	S	1599
347	46ab6321	18	M	Urethra	S	S	8156
348	ea3e8412	29	M	Pharynx	S	S	1599
349	4e91da43	27	M	Pharynx	S	S	10314
350	85d21740	22	M	Urethra	S	I	9363
351	0a103273	31	M	Urethra	S	S	8156
352	a5202c31	23	M	Urethra	S	S	8135
353	ecf3201c	31	M	Urethra	S	S	8156
354	7d86f311	19	M	Urethra	S	S	13333
355	4c3cedfa	20	M	Urethra	S	S	8122
356	026bc52a	30	M	Urethra	S	S	7363
357	906ebf6d	21	M	Urethra	S	S	8156
358	1cb8685e	21	M	Rectum	S	I	13292
359	789e2b89	22	M	Urethra	S	S	1580
360	55bcccd9	23	M	Urethra	S	S	1599
361	c8d423d0	26	M	Urethra	S	S	11990
362	2264ec13	21	M	Pharynx	S	I	11982
363	e0758881	44	M	Rectum	S	S	8143
364	d95ca8f6	18	M	Urethra	S	I	13292
365	ff8ed10e	18	M	Urethra	S	S	8135
366	b7bbbc4e	34	M	Urethra	S	S	1594
367	bf52ed50	24	M	Pharynx	S	S	7827
368	5d29ad9a	18	M	Urethra	S	I	1901
369	af564fa3	22	M	Pharynx	S	S	1599
370	ef838d46	26	M	Rectum	S	S	7822
371	599cf5ab	17	M	Urethra	S	S	1599
372	2005f839	18	M	Urethra	S	S	1594
373	ecacd654	33	M	Urethra	S	S	7359
374	5f6ff0af	19	M	Urethra	S	S	1594
375	5cb028b8	18	M	Urethra	S	S	1594
376	b886145a	24	M	Pharynx	S	S	7371
377	1a512b5e	19	M	Urethra	S	S	11990
378	ea2067df	20	M	Urethra	S	S	-
379	3214b5ff	63	M	Urethra	S	S	1579
380	a0f535c8	18	M	Urethra	S	R	8126

References

1. Eyre DW, Cule ML, Wilson DJ, et al. Diverse sources of *C. difficile* infection identified on whole-genome sequencing. *N Engl J Med*. 2013;369(13):1195-205.
2. De Silva D, Peters J, Cole K, et al. Whole-genome sequencing to determine transmission of *Neisseria gonorrhoeae*: an observational study. *Lancet Infect Dis*. 2016;16(11):1295-303.
3. Lunter G, Goodson M. Stampy: a statistical algorithm for sensitive and fast mapping of Illumina sequence reads. *Genome Res*. 2011;21(6):936-9.
4. Li H, Handsaker B, Wysoker A, et al. The Sequence Alignment/Map format and SAMtools. *Bioinformatics*. 2009;25(16):2078-9.
5. Altschul SF, Madden TL, Schaffer AA, et al. Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. *Nucleic Acids Res*. 1997;25(17):3389-402.
6. Guindon S, Gascuel O. A simple, fast, and accurate algorithm to estimate large phylogenies by maximum likelihood. *Syst Biol*. 2003;52(5):696-704.
7. Didelot X, Wilson DJ. ClonalFrameML: efficient inference of recombination in whole bacterial genomes. *PLoS Comput Biol*. 2015;11(2):e1004041.