

1 **Supplementary Tables describing isolates and oligonucleotides used in the study.**

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3 **Table S1.** Characteristics of *T. vaginalis* isolates used in the study. “+” indicates the presence of

4 the indicated SNP. “ND” indicates not determined. “Mixed” indicates a mixture of ≥ 0.25

5 ancestry proportion from both Population #1 and #2.

Isolate	Origin	Metronidazole MLC ($\mu\text{g/ml}$)	Population	<i>tntr4</i> C213G	<i>tntr6</i> A238T
MSA 1238	Charleston, SC	0.8	ND		
NY 12071	New York	0.8	ND		
NY 12260	New York	0.8	ND		
NY 12497	New York	0.8	ND		
SF-09081101	San Francisco, CA	0.8	ND		
WA05110911	Washington	0.8	1		
WA07130906	Washington	0.8	1		
PRA-98	ATCC	1 ^a	1		
CO-111763	Colorado	1.6	1		
CO-112068	Colorado	1.6	2	+	
CO-112362	Colorado	1.6	Mixed	+	
CO-112435	Colorado	1.6	1		
CO-112673	Colorado	1.6	2	+	
CO-113031	Colorado	1.6	1		
CO-113353	Colorado	1.6	1		
CO-114139	Colorado	1.6	1		
NY 11900	New York	1.6	ND		
NY 12102	New York	1.6	ND		
NY 12107	New York	1.6	ND		

NY 12487	New York	1.6	ND		
NY 12539	New York	1.6	ND	+	
SF-09042801	San Francisco, CA	1.6	1		
SF-09050402	San Francisco, CA	1.6	1		
SF-09051101	San Francisco, CA	1.6	1		
SF-09080302	San Francisco, CA	1.6	1		
SF-09081002	San Francisco, CA	1.6	1		
SF-09081103	San Francisco, CA	1.6	1		
WA05110918	Washington	1.6	1		
WA06020908	Washington	1.6	1		
WA06020910	Washington	1.6	1		
WA07130902	Washington	1.6	2	+	
CO-111629	Colorado	3.1	1		
CO-111682	Colorado	3.1	1		
CO-113632	Colorado	3.1	1		
CO-113739	Colorado	3.1	2	+	
LSU 4334	Baton Rouge, LA	3.1	ND		
MSA 1148	Richmond, VA	3.1	ND	+	
MSA 1248	Forrestville, MD	3.1	ND		
NY 10060906	New York	3.1	ND		
NY 10070903	New York	3.1	ND		
NY 10080904	New York	3.1	ND		
NY 10140901	New York	3.1	ND		
NY 11763	New York	3.1	ND		
NY 11789	New York	3.1	ND	+	
NY 11790	New York	3.1	ND	+	
NY 12068	New York	3.1	ND		

NY 12144	New York	3.1	ND	+	
NY 12186	New York	3.1	ND	+	+
NY 12233	New York	3.1	ND		
SF-09062401	San Francisco, CA	3.1	1		
SF-09063001	San Francisco, CA	3.1	1		
SF-09070801	San Francisco, CA	3.1	2	+	+
SF-09081102	San Francisco, CA	3.1	ND		
WA06150905	Washington	3.1	Mixed		
CO-112117	Colorado	6.3	2		
CO-113328	Colorado	6.3	2		
MSA 1175	Boston, MA	6.3	ND	+	
MSA 1239	Richardson, TX	6.3	ND		
NY 10220905	New York	6.3	ND	+	
NY 12145	New York	6.3	ND	+	
NY 12174	New York	6.3	ND	+	
SF-09042701	San Francisco, CA	6.3	2	+	
SF-09060901	San Francisco, CA	6.3	1		
SF-09061501	San Francisco, CA	6.3	1		
WA05110913	Washington	6.3	1		
30001	ATCC	8 ^a	1		
CO-112062	Colorado	12.5	2	+	
CO-113124	Colorado	12.5	1		
CO-113289	Colorado	12.5	2	+	
CO-113747	Colorado	12.5	Mixed	+	
MSA 1150	Indianapolis, IN	12.5	ND		
MSA 1171	Charlotte, NC	12.5	ND		
MSA 1218	New Haven, CT	12.5	ND		

MSA 1223	Chapel Hill, NC	12.5	ND		
MSA 1240	Baltimore, MD	12.5	ND	+	
MSA 1245	Quincy, FL	12.5	ND	+	
MSA 1246	Cookeville, TN	12.5	ND	+	
MSA 1249	Carrolton, GA	12.5	ND	+	
MSA 1265	Frankfort, KY	12.5	ND	+	
NY 10060907	New York	12.5	ND	+	
NY 10130902	New York	12.5	ND	ND	
NY 12143	New York	12.5	ND	+	
NY 12182	New York	12.5	ND	+	
NY 12188	New York	12.5	ND	+	
NY 12209	New York	12.5	ND	+	
NY 12344	New York	12.5	ND	+	
SF-09060101	San Francisco, CA	12.5	2	+	
SF-09061601	San Francisco, CA	12.5	2	+	+
SF-09073001	San Francisco, CA	12.5	2	+	
SF-09081003	San Francisco, CA	12.5	ND	+	
CO-113208	Colorado	25	2		
CO-113795	Colorado	25	Mixed	+	
CO-114154	Colorado	25	2	+	
MSA 1159	Hartford, CT	25	ND	+	
MSA 1176	Indianapolis, IN	25	ND		
MSA 1229	Columbus, OH	25	ND	+	
MSA 2055	Baton Rouge, LA	25	ND	+	+
NY 11788	New York	25	ND	+	
SF-09081001	San Francisco, CA	25	2	+	
MSA 1157	Lake Havaso, AZ	50	ND	+	

MSA 1160	Rochester, MN	50	ND		
MSA 1164	Houston, TX	50	ND	+	
MSA 1177	Minneapolis, MN	50	ND	+	
MSA 1234	Tallahassee, FL	50	ND		
MSA 1244	Beaverton, OR	50	ND		
MSA 1254	Dade City, FL	50	ND	+	
NY 12498	New York	50	ND	+	
NY 3241004	New York	50	ND	+	+
1113	East Hartford, CT	100	2	+	+
1133	Petersburg, VA	100	2	+	+
1181	Liberty, MO	100	2	+	+
1219	Houston, TX	100	2	+	
MSA 1041	Brooklyn, NY	100	ND		
MSA 1156	Los Angeles, CA	100	ND	+	
MSA 1158	Allentown, PA	100	ND	+	
MSA 1172	Ashtabula, OH	100	ND	+	
MSA 1221	Philadelphia, PA	100	ND	+	
MSA 1237	Great Falls, MT	100	ND	+	
MSA 1247	Charlotte, NC	100	ND	+	
MSA 1250	St. Louis, MO	100	ND	+	+
MSA 1268	Chicago, IL	100	ND	+	
50142	New York, NY	125 ^a	1		
50143	Columbus, OH	150 ^a	2	+	+
1184	Ft. Pierce, FL	100-200	2	+	+
1110	Waco, TX	200	1		
1115	Eau Claire, WI	200	2	+	+
1116	Fulton, MO	200	1		

1117	Atlanta, GA	200	2		
1118	Boston, MA	200	Mixed	+	
1119	Galveston, TX	200	2	+	+
1124	Rock Hill, SC	200	Mixed	+	
1178	Atlanta, Ga	200	1		
1179	Albany, OR	200	2	+	
1208	Palos Heights, IL	200	2		
1216	New Orleans, LA	200	2	+	
MSA 1149	Greensboro, NC	200	ND	+	+
MSA 1152	Auckley, IA	200	ND	+	+
MSA 1155	Dallas, TX	200	ND	+	
MSA 1174	Durham, NC	200	ND	+	
MSA 1226	Cincinnati, OH	200	ND		
MSA 1227	Fall River MA	200	ND	+	+
MSA 1236	New Orleans, LA	200	ND	+	+
MSA 1251	Plano, TX	200	ND	+	
MSA 1259	Lancaster, CA	200	ND		
MSA 1262	Vestavia, AL	200	ND		
MSA 1263	Augusta, GA	200	ND		
MSA 1266	Portland, OR	200	ND	+	+
50141	CT	200 ^a	1/2	+	+
1205	New York, NY	200-400	2	+	
1222	Indianapolis, IN	200-400	2	+	
1224	Seattle, WA	200-400	2	+	+
1112	Dedham, MA	400	1		
1122	Exton, PA	400	2	+	+
1130	Tucson, AZ	400	2	+	+

1131	Mobile, AL	400	2	+	
1185	St. Charles, MO	400	Mixed		
1189	Fall River, MA	400	1		
1191	Arlington, TX	400	2	+	
1200	Greenville, AL	400	1		
1202	Havre de Grace, MD	400	2	+	
1209	Houston, TX	400	2	+	+
1210	Jacksonville Beach, FL	400	2	+	
1212	Denver, CO	400	1		
1213	Boone, NC	400	2	+	
1214	Wichita, KS	400	2	+	
1215	Charlotte, NC	400	1		
1217	Baltimore MD	400	2	+	
1220	Lincoln, NE	400	2	+	+
MSA 1154	Richmond, CA	400	ND		
MSA 1162	Chicago, IL	400	ND	+	
MSA 1235	Indianapolis, IN	400	ND	+	
MSA 1252	San Diego, CA	400	ND	+	+
MSA 1253	Lincoln, Illinois	400	ND		
MSA 1255	Loveland, CO	400	ND	+	+
MSA 1258	Stone Mountain, GA	400	ND	+	+
MSA 1260	West Chester, OH	400	ND		
MSA 1261	Killen, TX	400	ND	+	
1107	Asheville, NC	>400	Mixed		+
1109	Mora, MN	>400	2	+	+
1121	Flowood, MS	>400	1		
1123	Richmond, VA	>400	2	+	+

1126	Brunswick, GA	>400	2	+	
1188	Charleston, SC	>400	1		
1190	Maitland, FL	>400	1		ND
1192	St. Louis, MO	>400	2	+	+
1201	Gallup, NM	>400	2	+	+
1203	Hampton, VA	>400	1		
1225	Evergreen Park, IL	>400	2	+	+
MSA 1165	Dearborn, MI	>400	ND	+	
MSA 1228	New Iberia, LA	>400	ND	+	+
MSA 1230	Springfield, MO	>400	ND		
MSA 12302	Atlanta, GA	>400	ND		
MSA 12303	Monroe, LA	>400	ND	+	+
MSA 1231	Dallas, TX	>400	ND		+
MSA 1241	Brunswick, ME	>400	ND	+	
MSA 1243	Manchester, CT	>400	ND	+	
MSA 1256	Cleveland, OH	>400	ND		
MSA 1257	West Chester, OH	>400	ND	ND	
MSA 1264	Cedar Park, TX	>400	ND	+	
MSA 1269	Surprise, AZ	>400	ND	+	+

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7 ^a Values found in reference (1).

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14 **Table S2.** Oligonucleotides used in the study. **Table S2.** Oligonucleotides used in the study.

Oligonucleotide	GenBank Accession #	Sequence	Orientation	Amplicon Size (bp)
ODH484	EAX85619 (<i>tntr1</i>) ^a ORF	ATGGCAATTTCTGCTATCGAAGCT	Forward	540
ODH485	EAX85619 (<i>tntr1</i>) ^a ORF	TTATTCATATAAAAAATCTTTCT	Reverse	
ODH570	EAX89456 (<i>tntr2</i>) ^a 200 bp 5' of ORF	CCAAAGGCACAAATAAACAAATTTT	Forward	594
ODH571	EAX89456 (<i>tntr2</i>) ^a 200 bp 3' of ORF	TCTATACAAATTATGAACCATCAT	Reverse	
ODH488	EAX89567 (<i>tntr3</i>) ^a ORF	ATGAGTGTTCACAGTGCATCCAA	Forward	549
ODH489	EAX89567 (<i>tntr3</i>) ^a ORF	TTAGTCAATATGAGTAATCTTTCC	Reverse	
ODH490	EAX94976 (<i>tntr4</i>) ^a ORF	ATGAGTGTCTTAAGTGCATCCAA	Forward	549
ODH491	EAX94976 (<i>tntr4</i>) ^a ORF	TTAGTCGGCATAAACTACCTTAGA	Reverse	
ODH572	EAX96000 (<i>tntr5</i>) ^a 200 bp 5' of ORF	AAGGAATAAGATGAAATAATAAAA	Forward	943
ODH573	EAX96000 (<i>tntr5</i>) ^a 200 bp 3' of ORF	TTGGATATAGATATCCGCAGAAAT	Reverse	
ODH528	EAX95789 (<i>tntr6</i>) ^a 200 bp 5' of ORF	CATTGAATTTATCGTICAAAATT	Forward	743
ODH495	EAX95789 (<i>tntr6</i>) ^a ORF	TTATTC AATGTA TGTAACCTTTCT	Reverse	
ODH496	EAY04224 (<i>tntr7</i>) ^a ORF	ATGTTTGACGC AATCAAAGAAAGA	Forward	516
ODH497	EAY04224 (<i>tntr7</i>) ^a ORF	TCAGTGAATCTTGGATGTGATTC	Reverse	
ODH498	EAY05392 (<i>tntr8</i>) ^a ORF	ATGGCTCTTGCGGCTCTCAAACAT	Forward	546
ODH499	EAY05392 (<i>tntr8</i>) ^a ORF	TTACTCTGGAACGAACTGAACCTT	Reverse	
ODH500	EAY15872 (<i>tntr9</i>) ^a ORF	ATGAACGTGCTTGATGCGATTICT	Forward	546
ODH501	EAY15872 (<i>tntr9</i>) ^a ORF	TCATTCATAAAATGATGCCTTGCA	Reverse	
ODH502	EAY16021 (<i>tntr10</i>) ^a ORF	ATGGCTCTTGAAGTCCTTAAGGCA	Forward	543
ODH503	EAY16021 (<i>tntr10</i>) ^a ORF	TTATTC AAGGAA TGTAAC TTTACG	Reverse	
ODH504	EAY14310 (<i>tntr11</i>) ^a ORF	ATGACAAGCGTGTGTAATGTATA	Forward	549
ODH505	EAY14310 (<i>tntr11</i>) ^a ORF	TTATTCATGTATGTGGCTTTGCA	Reverse	
ODH904	EAX94976 (<i>tntr4</i>)	GTCAGAGGCCAAGAAAAGCTTGCC	Forward	130
ODH905	EAX94976 (<i>tntr4</i>)	GAGCATCGCAGGTGATAACGTTC	Reverse	
ODH906 ^b	EAX94976 (<i>tntr4</i>) wild-type	CCTTCCAGAATACGCCACAAAGTAC	Forward	
ODH907 ^c	EAX94976 (<i>tntr4</i>) C213G	CCTTCCAGAATAGGCCACAAAGTAC	Forward	
ODH631	EAX95789 (<i>tntr6</i>)	CGTTGTTACAAAC AAGGAAAAACTCC	Forward	173
ODH632	EAX95789 (<i>tntr6</i>)	GGATGCACGCTCATICTTGA	Reverse	
ODH641 ^b	EAX95789 (<i>tntr6</i>) wild-type	GCTTCCCTTGATGAGAAGTCTCAAC	Forward	
ODH578 ^d	EAX95789 (<i>tntr6</i>) A238T	TGGAATGCAATAGCAGACACATGT	Forward	

ODH689 ^e	EAY02351	CGTCAACATCGGTGGCTTCA	Forward	489
ODH690 ^e	EAY02351	GCGACAGCGACGACATTCAT	Reverse	
ODH691 ^e	EAX82426	GTGCCATTAC AACAGCATCG	Forward	451
ODH692 ^e	EAX82426	CCAAGTATAGCTCCGCTGAC	Reverse	
ODH693 ^e	EAY10832	GAACAGGAGCACCAGCAGAA	Forward	412
ODH694 ^e	EAY10832	TCTCTAGCAACGCAGCCAAC	Reverse	
ODH695 ^e	EAY03282	TCGTCCAGGATGGTGCTTT	Forward	494
ODH696 ^e	EAY03282	ACGCCTTCCCTTCATCTT	Reverse	
ODH697 ^e	EAX88709	TCATCGGCCAATGGAACCAA	Forward	491
ODH698 ^e	EAX88709	TCCGTGCGGACAATCCAAG	Reverse	
ODH699 ^e	EAY10370	GCTGAGTGACGGTGGACATT	Forward	449
ODH700 ^e	EAY10370	GAAGATGAGGTCCTCCTTGA	Reverse	
ODH701 ^e	EAY13510	AGCCAGTTGGCTTCTGAGTT	Forward	459
ODH702 ^e	EAY13510	AACAATTCCGCAAGCTGGAG	Reverse	

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16 ^a Gene nomenclature (*tvntr*) is found in reference (2).

17 ^b TaqMan[®] probe containing a 5' Cy5 fluorophore and 3' Black Hole Quencher-2.

18 ^c TaqMan[®] probe containing a 5' FAM fluorophore and 3' Black Hole Quencher-1.

19 ^d TaqMan[®] probe containing a 5' FAM fluorophore and 3' Black Hole Quencher-2.

20 ^e Primer sequences originally found in reference (3).

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30 **References**

31

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