

SUPPLEMENTAL MATERIAL

Directional Evolution of *Chlamydia trachomatis* Towards Niche-specific Adaptation

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SUPPLEMENTAL TABLES

TABLE S1 List of strains studied.

Strain name	GenBank Accession number	Reference or source
<i>Chlamydia trachomatis</i> A/2497	NC_016798	5
<i>Chlamydia trachomatis</i> A/363	HE601796	5
<i>Chlamydia trachomatis</i> A/5291	HE601810	5
<i>Chlamydia trachomatis</i> A/7249	HE601797	5
<i>Chlamydia trachomatis</i> A/Har13	NC_007429	1
<i>Chlamydia trachomatis</i> B/TZ1A828/OT	NC_012687	10
<i>Chlamydia trachomatis</i> B/Jali20/OT	NC_012686	10
<i>Chlamydia trachomatis</i> Ba/Apache-2	--- ^a	This publication
<i>Chlamydia trachomatis</i> C/TW3	--- ^a	This publication
<i>Chlamydia trachomatis</i> D/SotonD1	HE601798	5
<i>Chlamydia trachomatis</i> D/SotonD5	HE601799	5
<i>Chlamydia trachomatis</i> D/SotonD6	HE601800	5
<i>Chlamydia trachomatis</i> D/UW3	NC_000117	12
<i>Chlamydia trachomatis</i> E/SW2	FN652779	14
<i>Chlamydia trachomatis</i> E/SW3	HE601801	5
<i>Chlamydia trachomatis</i> E/SotonE4	HE601802	5
<i>Chlamydia trachomatis</i> E/SotonE8	HE601803	5
<i>Chlamydia trachomatis</i> E/11023	NC_017431	6
<i>Chlamydia trachomatis</i> E/150	NC_017439	6
<i>Chlamydia trachomatis</i> E/Bour	HE601870	5
<i>Chlamydia trachomatis</i> F/SW4	HE601804	5
<i>Chlamydia trachomatis</i> F/SW5	HE601805	5
<i>Chlamydia trachomatis</i> F/SotonF3	HE601806	5
<i>Chlamydia trachomatis</i> F/IC-Cal3	--- ^a	This publication
<i>Chlamydia trachomatis</i> G/9301	NC_017432	6
<i>Chlamydia trachomatis</i> G/9768	NC_017429	6
<i>Chlamydia trachomatis</i> G/11074	NC_017440	6
<i>Chlamydia trachomatis</i> G/11222	NC_017430	6
<i>Chlamydia trachomatis</i> G/SotonG1	HE601807	5

<i>Chlamydia trachomatis</i> G/UW57	--- ^a	This publication
<i>Chlamydia trachomatis</i> H/UW43	--- ^a	This publication
<i>Chlamydia trachomatis</i> I/UW12	--- ^a	This publication
<i>Chlamydia trachomatis</i> Ia/SotonIa1	HE601808	5
<i>Chlamydia trachomatis</i> Ia/SotonIa3	HE601809	5
<i>Chlamydia trachomatis</i> J/6276	ABYD01000001	6
<i>Chlamydia trachomatis</i> J/UW36	--- ^a	This publication
<i>Chlamydia trachomatis</i> K/SotonK1	HE601794	5
<i>Chlamydia trachomatis</i> K/UW31	--- ^a	This publication
<i>Chlamydia trachomatis</i> L1/1322/p2	HE601951	5
<i>Chlamydia trachomatis</i> L1/115	HE601952	5
<i>Chlamydia trachomatis</i> L1/224	HE601953	5
<i>Chlamydia trachomatis</i> L1/440/LN	HE601950	5
<i>Chlamydia trachomatis</i> L2/25667R	HE601954	5
<i>Chlamydia trachomatis</i> L2/434/Bu	NC_010287	13
<i>Chlamydia trachomatis</i> L2b/UCH-1	NC_010280	13
<i>Chlamydia trachomatis</i> L2b/8200/07	HE601795	5
<i>Chlamydia trachomatis</i> L2b/UCH-2	HE601956	5
<i>Chlamydia trachomatis</i> L2b/Canada1	HE601963	5
<i>Chlamydia trachomatis</i> L2b/Canada2	HE601957	5
<i>Chlamydia trachomatis</i> L2b/LST	HE601958	5
<i>Chlamydia trachomatis</i> L2b/CV204	HE601960	5
<i>Chlamydia trachomatis</i> L2b/795	HE601949	5
<i>Chlamydia trachomatis</i> L2b/Ams1	HE601959	5
<i>Chlamydia trachomatis</i> L2b/Ams2	HE601961	5
<i>Chlamydia trachomatis</i> L2b/Ams3	HE601962.1	5
<i>Chlamydia trachomatis</i> L2b/Ams4	HE601964.1	5
<i>Chlamydia trachomatis</i> L2b/Ams5	HE601965.1	5
<i>Chlamydia trachomatis</i> L2c	NC_015744	11
<i>Chlamydia trachomatis</i> L3/404/LN	HE601955	5
<i>Chlamydia muridarum</i> Nigg	NC_002620	9

^a As these historical prototype strains were not fully-sequenced so far, we amplified and sequenced the 75 genes under evaluation. The obtained nucleotide sequences have the GenBank accession numbers: JQ066324 - JQ066722.

TABLE S2 Identification of genes and corresponding functional category.

ORF (gene) ^a	Functional category ^b
CT048 (<i>yraL</i>)	HK
CT049	other
CT050	other
CT051	other
CT058	SEC
CT059 (<i>fer</i>)	HK

CT105	other
CT115 (<i>incD</i>)	SEC
CT116 (<i>incE</i>)	SEC
CT117 (<i>incF</i>)	SEC
CT118 (<i>incG</i>)	SEC
CT119 (<i>incA</i>)	SEC
CT143	other
CT144	other
CT147	SEC
CT192	SEC
CT195	SEC
CT214	SEC
CT222	SEC
CT223	SEC
CT228	SEC
CT229	SEC
CT232 (<i>incB</i>)	SEC
CT233 (<i>incC</i>)	SEC
CT249	SEC
CT288	SEC
CT293 (<i>accD</i>)	HK
CT365	SEC
CT383	SEC
CT412 (<i>pmpA</i>)	CEP
CT413 (<i>pmpB</i>)	CEP
CT414 (<i>pmpC</i>)	CEP
CT442 (<i>crpA</i>)	SEC
CT443 (<i>omcB</i>)	CEP
CT456 (<i>tarp</i>)	SEC
CT529	SEC
CT618	SEC
CT622	SEC
CT623	other
CT624 (<i>mviN</i>)	CEP
CT653 (<i>yhbG</i>)	HK
CT674 (<i>yscC</i>)	HK
CT675 (<i>karG</i>)	HK
CT676	other
CT677 (<i>frr</i>)	HK
CT678 (<i>pyrH</i>)	HK
CT679 (<i>tsf</i>)	HK
CT680 (<i>rs2</i> or <i>rpsB</i>)	HK
CT681 (<i>ompA</i>)	CEP
CT682 (<i>pbpB</i>)	CEP
CT683 (<i>TPR motif-protein</i>)	other

CT684	HK
CT685	HK
CT686 (<i>sufD</i>)	HK
CT687 (<i>yfhO_1</i>)	HK
CT688 (<i>parB</i>)	HK
CT689 (<i>dppF</i>)	HK
CT694	SEC
CT760 (<i>ftsW</i>)	HK
CT783	other
CT812 (<i>pmpD</i>)	CEP
CT813	SEC
CT818 (<i>tyrP_2</i>)	HK
CT852 (<i>yhgN</i>)	CEP
CT859 (<i>ispH</i>)	HK
CT860	SEC
CT861	SEC
CT862 (<i>lcrH_2</i>)	HK
CT867	SEC
CT868	SEC
CT869 (<i>pmpE</i>)	CEP
CT870 (<i>pmpF</i>)	CEP
CT871 (<i>pmpG</i>)	CEP
CT872 (<i>pmpH</i>)	CEP
CT874 (<i>pmpI</i>)	CEP

^a Open reading frame (ORF) numbers are based on the D/UW3 strain genome annotation (GenBank accession number NC_000117).

^b The 75 genes under evaluation were categorized according to their functional role: housekeeping genes (HKs), genes encoding well-known cell envelope proteins (CEPs), genes coding for proteins secreted into the cytosol of the host cells or to the inclusion membrane (SEC), and genes encoding proteins with unknown function or with non-consensual biological role (classified as “other”).

TABLE S3 Oligonucleotide primers used for PCR and sequencing.

ORF (gene) ^a	Primer ^a	Primer sequence (5' to 3') ^a	Primer location ^a	Amplicon size (bp) ^a
CT048 (<i>yraL</i>)	CT048-1 ^b	GAGCCGGCTCTTTTAAATGGTTT	53104 - 53126	1059
	CT048-2 ^b	GTCGACGGAACAGACGAAGAAA	54141 - 54162	
CT049 ^c	---	---	---	---
CT050/CT051	CT050/51-1A ^{d, e}	TGGGCGCTGGTTATTA ACTATTTG	377806 - 377829	3704
	CT050/51-2A ^{b, e}	GACCCCATCCCCTTTGGAGT	381490 - 381509	
	CT050/51-1D ^b	ACAAAGCGCTTTCAGA ACATACAT	55452 - 55475	3589
	CT050/51-2D ^b	AGGGCGTCTTTTTCATGATTCTAT	59017 - 59040	
	CT050/51-1 ^f	CTAAGAGTTATGTAGCTATC	55542 - 55561	
	CT050/51-1S ^f	AGTTAAGGGAGAGAATCTC	55593 - 55611	
	CT050/51-3 ^f	TTGTAGTGTGCAAGATTGTC	55857 - 55876	
	CT050/51-4 ^f	TTGTGCCACTACAATACCTT	58726 - 58745	
	CT050/51-5 ^f	AACCTTTCCAATATCACCGT	56566 - 56585	
	CT050/51-6 ^f	GCACAGATCGCCAATATCAA	58076 - 58095	

	CT050/51-7 ^f	AGTCACTCCAGACAATTCTA	57699 - 57718	
	CT050/51-8 ^f	TTAGTGAGACAGGCATTGA	57160 - 57178	
CT058/CT059 (<i>fer</i>)	CT058/9-1 ^b	AGTACCGGCCGAATCTCTTTCTCC	67315 - 67338	1634
	CT058/9-2 ^b	GTCGGGGGTTCTGAATCCCTCTA	68927 - 68948	
	CT058/9-3 ^f	TCTCATTACTTCTCTTGCGT	68432 - 68451	
CT115-118 ^g	---	---	---	---
CT119 (<i>incA</i>)	CT119-1 ^b	GGGGCATTTCGATGATATAAATAAG	136695 - 136718	1273
	CT119-2 ^b	TGCACTGCGCAGACAAGAA	137949 - 137967	
CT143	CT143-1 ^b	AAGGGCGGAGACTATGTGGACA	159770 - 159791	1325
	CT143-2 ^b	AGCGCTGAACGCATACTTATTTTA	161071 - 161094	
CT144 ^g	---	---	---	---
CT147 ^h	CT147-1 ^b	GGGGAAAGTGAGCTCTTCGGTATC	165430 - 165453	2819
	CT147-2 ^b	CGCCGCTACAACAGCTTTAGTGA	168226 - 168248	
	CT147-3 ^b	ATTGCGTCCCAAGATATACGACAG	167928 - 167951	2294
	CT147-4 ^b	CACGCCAACCCAGAATCCTT	170202 - 170221	
	CT147-5 ^f	TAATCATCCACTAGAAGCG	166229 - 166247	
	CT147-6 ^f	TGTTCTAGCTGCTCTTGAAT	167649 - 167668	
	CT147-7 ^f	TGGATGGTGTGTCAGAATTA	168511 - 168530	
	CT147-8 ^f	TACCTCTAGATGTTTTGCGT	169786 - 169805	
CT192	CT192-1 ^b	ATATGCGCAAGCACACCTTCC	215717 - 215737	1016
	CT192-2 ^b	CTGGGCGTCCATTACACAACA	216713 - 216732	
	CT192-3 ^f	CGTATCGATTCCCTTCTTCTA	215998 - 216017	
	CT192-4 ^f	CTCCTCTTATTGAAGAAGCT	2156196 - 216215	
CT195	CT195-1 ^b	CCTCCGCCTAATCCTCGACTACAT	219655 - 216678	1320
	CT195-2 ^b	CCAGCGGTTGATATTTCTTGATTA	220951 - 220974	
CT214	CT214-1 ^b	AGGGCTTCTATTCCTCAAACAGTA	241463 - 241486	1797
	CT214-2 ^b	TTCCCCGTTCTAAAGATCAGTTAT	243236 - 243259	
	CT214-3 ^f	AACAGCCTGGATCTATATCA	242027 - 242046	
CT223	CT223-1 ^b	GCAACGCATATCGCTCCTCA	251102 - 251121	1308
	CT223-2 ^b	GTGCGCCCCTTCTCGTAAAG	252390 - 252409	
CT228/CT229	CT228/9-1 ^b	CGGTCCC GGATTATCAAACAAGT	254667 - 254690	1816
	CT228/9-2 ^b	ATGCGGCCATCCCAGAAGC	256464 - 256482	
	CT228/9-3 ^f	AGATTACGCAAACGTTGCTC	255033 - 255052	
	CT228/9-4 ^f	GTTGTGATTGCAGCAGTAG	255972 - 255990	
CT232/CT233 (<i>incB/incC</i>)	CT232/3-1 ^b	GATTAGGCGGAGGGGTTCTCTT	259223 - 259244	1262
	CT232/3-2 ^b	CTCTCCGCGACGCAAATAAG	260464 - 260484	
CT249	CT249-1 ^b	ACCACCCTTTAGCCATCCATTCC	279170 - 279192	704
	CT249-2 ^b	AATTGCGCCGCCTCCTTGTA	279854 - 279873	
CT288	CT288-1 ^b	TTTTACGCACAATGAACCCAGAAA	321582 - 321605	2063
	CT288-2 ^b	CGGGCTCCTCGGGAACAG	323627 - 323644	
	CT288-3 ^f	TTACCTGACCTCAGACACC	322164 - 322182	
	CT288-4 ^f	GTCAGCTCGTCGTTTATTG	323121 - 323139	
CT293 (<i>accD</i>)	CT293-1 ^b	TGCGCCAGAAGCTCCAGAAGTAGC	326322 - 326345	1350
	CT293-2 ^b	AGGATCTGGCTGGGGATGGTTAGC	327648 - 327671	
CT365	CT365-1 ^b	AAATTCGCAAACCTTGCTCTTTTC	416102 - 416125	2213
	CT365-2 ^b	GATCGGGATTCCCCTGGATA	418295 - 418314	
	CT365-3 ^f	CTAACTTCCAAGTTTCCTCT	416572 - 416591	
	CT365-4 ^f	CTCATTGCAGGTATTGTTGT	417720 - 417739	
CT412 (<i>pmpA</i>) ⁱ	---	---	---	---
CT413 (<i>pmpB</i>) ⁱ	---	---	---	---
CT414 (<i>pmpC</i>) ^j	---	---	---	---
CT442 (<i>crpA</i>)	CT442-1 ^b	CTCCTCCCTTTCCATACATCATCT	511256 - 511279	783
	CT442-2 ^b	AAGCGATTCTTCTTCCGATACAT	512015 - 512038	

CT443 (<i>omcB</i>) ^k	---	---	---	---
CT456 (<i>tarp</i>)	CT456-1 ^b	ACAAACGTTACCCGGTATGCTGTT	530723 - 530746	3362
	CT456-2 ^b	TTGCGCCTTGTGCGATTGTGAT	534064 - 534084	
	CT456-3 ^f	TACCTCATCAAGCGATCATA	531252 - 531271	
	CT456-4 ^f	CCACCAGTTGTTATTATGTC	533470 - 533489	
	CT456-5 ^f	AGACATGTCTCTTCCTTCAT	531867 - 531886	
	CT456-6 ^f	TACATCAGAGATTACGTCTC	532889 - 532908	
	CT456-7 ^f	GAGTTTCATTGGAGAAGGAA	532413 - 532432	
	CT456-8 ^f	CGTTACCCGGTATGCTGTT	530728 - 530746	
	CT456-9 ^f	TACAAACACTACTGCCTTCA	533363 - 533382	
	CT456-10 ^f	TTGTTACTACCTACGTCATC	531328 - 531347	
	CT456-11 ^f	CTAATTAATCGGCTGTTG	530869 - 530887	
CT529	CT529-1 ^b	ACGCGGCTCCTTAAAGCAAACAA	596464 - 596486	1659
	CT529-2 ^b	CGCGCATATCCGGGGAGTCT	598103 - 598122	
	CT529-3 ^f	TCTCGCAAGCATTTCCTCT	596984 - 597003	
CT618	CT618-1 ^b	TCCCGATATGCCTCCTTTTGAGTC	698080 - 698103	1360
	CT618-2 ^b	ATGCGCACGCAAGCCAATC	699421 - 699439	
CT622	CT622-1 ^b	GGCTCCCCCTCAATTCACAAACTT	707046 - 707069	2319
	CT622-2 ^b	GGTCGCGGAAACCAAATGAAATA	709342 - 709364	
	CT622-3 ^f	TGATTGCTTGTATTTCCGGCT	707784 - 707803	
	CT622-4 ^f	TTCAGCATCGTCCTCTGTAA	708885 - 708904	
	CT622-5 ^f	AGAAGAGATTATGCAGAAGC	707298 - 707317	
CT623	CT623-1 ^b	TTTGCCATTAATAATTGGATTCA	708957 - 708980	1516
	CT623-2 ^b	CATGGGTCGGTTGTATGAGATGT	710450 - 710472	
CT624 (<i>mviN</i>)	CT624-1 ^b	ATCGAGCGCGGACATCTCC	710225 - 710243	2326
	CT624-2 ^b	GATATCGCGCCCTTCGTAAGA	712529 - 712550	
	CT624-3 ^f	CTAAAGGTTTGTGGGACT	710827 - 710846	
	CT624-4 ^f	ACGTGCGCAAATATTTCTC	712033 - 712052	
CT653 (<i>yhbG</i>)	CT653-1 ^b	AGCCGCGATAGCTAACGAAGTG	750194 - 750215	922
	CT653-2 ^b	GAAGGCGGAATGAAAGTCCTCTC	751093 - 751115	
CT674 (<i>yscC</i>)	CT674-1 ^b	TTTCAAGCGGAATCGCAAGGAAT	770234 - 770256	3229
	CT674-2 ^b	CCGGGATCGAACCGACGAC	773444 - 773462	
	CT674-3 ^f	AGAGCCATCAGATTTTCTCT	770846 - 770865	
	CT674-4 ^f	AGAGGAAGAGAAGTACTGAGTAA	772996 - 773015	
	CT674-5 ^f	ATGACTTGAAAAGTCGTTGAA	771470 - 771489	
	CT674-6 ^f	TCCGTACATCATACTACTGA	772505 - 772524	
	CT674-7 ^f	GATATCGGAGTCAATCTTGTT	772771 - 772791	
CT675 (<i>karG</i>)/ CT676	CT675/6-1 ^b	CCCGGCTTTGGGCATTCC	773458 - 773475	1933
	CT675/6-2 ^b	TCATTCCGGTAACAGGGGTTCCG	775369 - 775390	
	CT675/6-3 ^f	CGTGATCAGATTAATCAGCT	774669 - 774688	
CT677/CT678/ CT679	CT677/9-1 ^b	ATGGGGCCAGGACGGGTCTA	775420 - 775439	2393
	CT677/9-2 ^b	AAATTTTATCTCCGGTGCCTCCTG	777789 - 777812	
(<i>frr</i> , <i>pyrH</i> , <i>tsf</i>)	CT677/9-4 ^f	GAAGATATCACTGTACCAAC	776088 - 776107	
	CT677/9-5 ^f	TGGCTAAAGACATTGCTATG	777107 - 777126	
CT680 (<i>rs2</i> or <i>rpsB</i>) ^c	---	---	---	---
CT681 (<i>ompA</i>) ^j	---	---	---	---
CT682 (<i>pbpB</i>) ^h	pbpB-1 ^{b, c}	---	---	---
	pbpB-2 ^{b, c}	---	---	---
	CT682-1 ^b	GCATTGTGATCGCGCAGGAGTA	780841 - 780862	3149
	CT682-2 ^b	TTTCCGCCTCTTCCATAGTCGTT	783966 - 783989	
	CT682-3 ^f	TTGATAGGCAAGCGATCTAT	781484 - 781503	
	CT682-4 ^f	ATATTCTCCAGGAAGTCCTA	783318 - 783337	

	CT682-5 ^f	TGGAAATGTTTGAGTGTGAA	782079 - 782098	
CT683 (<i>TPR-motif protein</i>)	CT683-1 ^b	TCGCTGCGGTAGGATATGAAGATG	783756 - 783779	1702
	CT683-2 ^b	TCGCCGCGTAAATGAACCAAT	785437 - 785457	
	CT683-3 ^f	GACTGTCAAACAGCCCTTAA	785108 - 785127	
CT684 ^h	CT684-A1 ^b	TGGGCATTACAATCTTGGGTTATG	784615 - 784638	1471
	CT684-A2 ^b	AACAGCGGCATGCAGTTGATG	786065 - 786085	
	CT684-B1 ^b	ATTTCTGGGAGGCAATCCACAAT	785785 - 785806	1398
	CT684-B2 ^b	TCCCGGGAATCCATATACCTCTTC	787159 - 787182	
CT685/CT686	CT684-A3 ^f	TCATCAATAGGGATGCCTAA	785651 - 785670	
	CT684-A4 ^f	TCTGGATCTGCGTCTTCTAA	785609 - 785628	
	CT685/6-1 ^b	GCTCGGGAAGCAACCAAGTTATTA	786722 - 786745	2386
	CT685/6-2 ^b	AAGCGAGTCCCATGATACGAGAT	789084 - 789107	
CT687 (<i>yfhO_1</i>) ^g	CT685/6-3 ^f	TCTTGATGTCGATGCTTTGA	787308 - 787327	
	CT685/6-4 ^f	ATGCAGAAGCTCGATTACTT	788502 - 788521	
CT688 (<i>parB</i>) ^g	---	---	---	---
CT689 (<i>dppF</i>) ^g	---	---	---	---
CT694	CT694-1 ^b	TACAGGGGGAGGCGCTTCCTTA	796071 - 796092	1541
	CT694-2 ^b	CGCGCTCTTCTAGCTCTCCCTCTT	796528 - 797551	
	CT694-3 ^f	GATAACTCTTAACCCCATTTG	796267 - 796286	
CT760 (<i>ftsW</i>)	CT760-1 ^b	CTTGGGCCATTGCATTGAGTAAT	892695 - 892717	1417
	CT760-2 ^b	CCCCAGAGAACATCCGATTGAC	894090 - 894111	
	CT760-3 ^f	AGGATGTAGGTAAACTTGCA	893445 - 893464	
CT783	CT783-1 ^b	AGCGGGGATTTCAGCATTCT	919328 - 919347	1427
	CT783-2 ^b	TGCCCTCGCTCTTCATC	920676 - 920694	
	CT783-3 ^f	TGTCAATACCTCCCTAGTT	919913 - 919932	
CT812 (<i>pmpD</i>) ⁱ	---	---	---	---
CT813	CT813-1 ^b	CTGCGTGTGCTCTGGAAAATAAT	954988 - 955011	1460
	CT813-2 ^b	AGGCCGAGCCCTACTCAAAAAT	956365 - 956387	
CT818 (<i>tyrP_2</i>)	CT818-1 ^b	CCTGGCGGGAAAGGGACTCT	961600 - 961619	1531
	CT818-2 ^b	GCGCATAATCGCGATCATAACAATC	963107 - 963130	
	CT818-3 ^f	GAATAGCACGTTTAAACCTCA	962512 - 962531	
CT852 (<i>yhgN</i>)	CT852-1 ^b	CTGCCGCACCAGCAAGGAT	1001213 - 1001231	768
	CT852-2 ^b	TAGGCGCTCAACTTCTGGTATCTG	1001957 - 1001980	
CT859 (<i>ispH</i>)/ CT860	CT856/60-1 ^b	GAGGGGGCTTTGCGGATTTAT	1011689 - 1011709	2779
	CT856/60-2 ^b	CCGGAATGCTTGGCTTGACA	1014448 - 1014467	
	CT856/60-3 ^f	ACGACATTGAGTATGGATGA	1012253 - 1012272	
	CT856/60-4 ^f	ACCCGATATCTCATAAATC	1013829 - 1013848	
CT861/CT862 (<i>lcrH_2</i>)	CT856/60-5 ^f	GTATTTTCAGTTGCCTAAGGA	1012658 - 1012677	
	CT861/2-1 ^d	GAGGGCAGAGGCTTCTTCAACAAG	1014122 - 1014144	2400
	CT861/2-2 ^b	CTAGGCGTCCCAATTGGAGACTC	1016499 - 1016521	
	CT861/2-3 ^f	AATAGCTCTCCAACCATCAA	1014640 - 1014659	
	CT861/2-4 ^f	GACTATGAGGAAAGTTCTAC	1016071 - 1016090	
	CT861/2-5 ^f	TCTCCTGTTGCTATTGTTTG	1014159 - 1014178	
CT867/CT868	CT861/2-6 ^f	TTGACTTTCTCCTGATGCTT	1015102 - 1015121	
	CT867/8-1 ^d	TCCCGACTGCTGGGGCTTAGA	1022902 - 1022922	2889
	CT867/8-2 ^d	CATCGCGTCATGCCATGTCCTAT	1025768 - 1025790	
	CT867/8-3 ^f	TCCTTCAGCTACTCTGATT	1022935 - 1022953	
	CT867/8-4 ^f	TGGTAAGCGGATTACAGAT	1023641 - 1023659	
	CT867/8-5 ^f	TTCATGGCGTTCTACAGAAT	1024302 - 1024321	
	CT867/8-6 ^f	AATGTCAGAATCCCAAGCA	1024923 - 1024941	
CT867/8-7 ^f	AGAGGGGAAGTCTTAATTTCC	1025542 - 1025561		

CT869 (<i>pmpE</i>) ⁱ	---	---	---	---
CT870 (<i>pmpF</i>) ⁱ	---	---	---	---
CT871 (<i>pmpG</i>) ⁱ	---	---	---	---
CT872 (<i>pmpH</i>) ⁱ	---	---	---	---
CT874 (<i>pmpI</i>) ⁱ	---	---	---	---

^a Open reading frame (ORF) numbers, primer location (genome coordinates), primer sequences and amplicon size are based on the D/UW3 strain genome annotation (GenBank accession number NC_000117).

^b Amplification primers also used for automated sequencing.

^c Previously described (3).

^d Primers exclusively used for PCR amplification.

^e Primer sequences, location and amplicon size refers to the L2/434 strain genome annotation (GenBank accession number NC_010287) as these primers were designed for amplification and automated sequencing in LGV and ocular strains, and they have no homology in the D/UW3 genome sequence.

^f Primers exclusively used for automated sequencing.

^g Previously described (8).

^h Due to the large gene size (for CT147 and CT682) or for PCR optimization (for CT684), two PCR primer pairs were designed to generate two overlapping amplicons for the entire gene.

ⁱ Previously described (4).

^j Previously described (2).

^k Previously described (7).

REFERENCES

1. **Carlson JH, Porcella SF, McClarty G, Caldwell HD.** 2005. Comparative genomic analysis of *Chlamydia trachomatis* oculotropic and genitotropic strains. *Infect Immun*, **73**:6407–6418.
2. **Gomes JP, Bruno WJ, Borrego MJ, Dean D.** 2004. Recombination in the genome of *Chlamydia trachomatis* involving the polymorphic membrane protein C gene relative to *ompA* and evidence for horizontal gene transfer. *J. Bacteriol.* **186**:4295–4306.
3. **Gomes JP, Bruno WJ, Nunes A, Santos N, Florindo C, Borrego MJ, Dean D.** 2007. Evolution of *Chlamydia trachomatis* diversity occurs by widespread interstrain recombination involving hotspots. *Genome Res.* **17**:50–60.
4. **Gomes JP, Nunes A, Bruno WJ, Borrego MJ, Florindo C, Dean D.** 2006. Polymorphisms in the nine polymorphic membrane proteins of *Chlamydia trachomatis* across all serovars: evidence for serovar Da recombination and correlation with tissue tropism. *J. Bacteriol.* **188**:275–286.
5. **Harris SR, Clarke IN, Seth-Smith HM, Solomon AW, Cutcliffe LT, Marsh P, Skilton RJ, Holland MJ, Mabey D, Peeling RW, Lewis DA, Spratt BG, Unemo M, Persson K, Bjartling C, Brunham R, de Vries HJ, Morr e SA, Speksnijder A, B b ear CM, Clerc M, de Barbeyrac B,**

- Parkhill J, Thomson NR.** 2012. Whole-genome analysis of diverse *Chlamydia trachomatis* strains identifies phylogenetic relationships masked by current clinical typing. *Nat. Genet.* **44**:413–419.
6. **Jeffrey BM, Suchland RJ, Quinn KL, Davidson JR, Stamm WE, Rockey DD.** 2010. Genome sequencing of recent clinical *Chlamydia trachomatis* strains identifies loci associated with tissue tropism and regions of apparent recombination. *Infect. Immun.* **78**:2544–2553.
 7. **Millman KL, Tavaré S, Dean D.** 2001. Recombination in the *ompA* gene but not the *omcB* gene of *Chlamydia* contributes to serovar-specific differences in tissue tropism, immune surveillance, and persistence of the organism. *J. Bacteriol.* **183**:5997–6008.
 8. **Nunes A, Nogueira PJ, Borrego MJ, Gomes JP.** 2008. *Chlamydia trachomatis* diversity viewed as a tissue-specific coevolutionary arms race. *Genome Biol.* **9**:R153.
 9. **Read TD, Brunham RC, Shen C, Gill SR, Heidelberg JF, White O, Hickey EK, Peterson J, Utterback T, Berry K, Bass S, Linher K, Weidman J, Khouri H, Craven B, Bowman C, Dodson R, Gwinn M, Nelson W, DeBoy R, Kolonay J, McClarty G, Salzberg SL, Eisen J, Fraser CM.** 2000. Genome sequences of *Chlamydia trachomatis* MoPn and *Chlamydia pneumoniae* AR39. *Nucleic Acids Res.* **28**:1397–1406.
 10. **Seth-Smith HM, Harris SR, Persson K, Marsh P, Barron A, Bignell A, Bjartling C, Clark L, Cutcliffe LT, Lambden PR, Lennard N, Lockey SJ, Quail MA, Salim O, Skilton RJ, Wang Y, Holland MJ, Parkhill J, Thomson NR, Clarke IN.** 2009. Co-evolution of genomes and plasmids within *Chlamydia trachomatis* and the emergence in Sweden of a new variant strain. *BMC Genomics* **10**:239.
 11. **Somboonna N, Wan R, Ojcius DM, Pettengill MA, Joseph SJ, Chang A, Hsu R, Read TD, Dean D.** 2011. Hypervirulent *Chlamydia trachomatis* clinical strain is a recombinant between lymphogranuloma venereum (L(2)) and D lineages. *MBio* **2**:e00045-11.
 12. **Stephens RS, Kalman S, Lammel C, Fan J, Marathe R, Aravind L, Mitchell W, Olinger L, Tatusov RL, Zhao Q, Koonin EV, Davis RW.** 1998. Genome sequence of an obligate intracellular pathogen of humans: *Chlamydia trachomatis*. *Science* **282**:754–759.

13. Thomson NR, Holden MT, Carder C, Lennard N, Lockey SJ, Marsh P, Skipp P, O'Connor CD, Goodhead I, Norbertzcak H, Harris B, Ormond D, Rance R, Quail MA, Parkhill J, Stephens RS, Clarke IN. 2008. *Chlamydia trachomatis*: genome sequence analysis of lymphogranuloma venereum isolates. *Genome Res.* **18**:161–171.
14. Unemo M, Seth-Smith HM, Cutcliffe LT, Skilton RJ, Barlow D, Goulding D, Persson K, Harris SR, Kelly A, Bjartling C, Fredlund H, Olcén P, Thomson NR, Clarke IN. 2010. The Swedish new variant of *Chlamydia trachomatis*: genome sequence, morphology, cell tropism and phenotypic characterization. *Microbiology* **156**:139–1404.