A proposed genus boundary for the prokaroytes based on genomic insights

Supplemental Figures (Fig. S1 to Fig. S6) and Tables (Table S1, Table S2, Table S3) for Online Posting

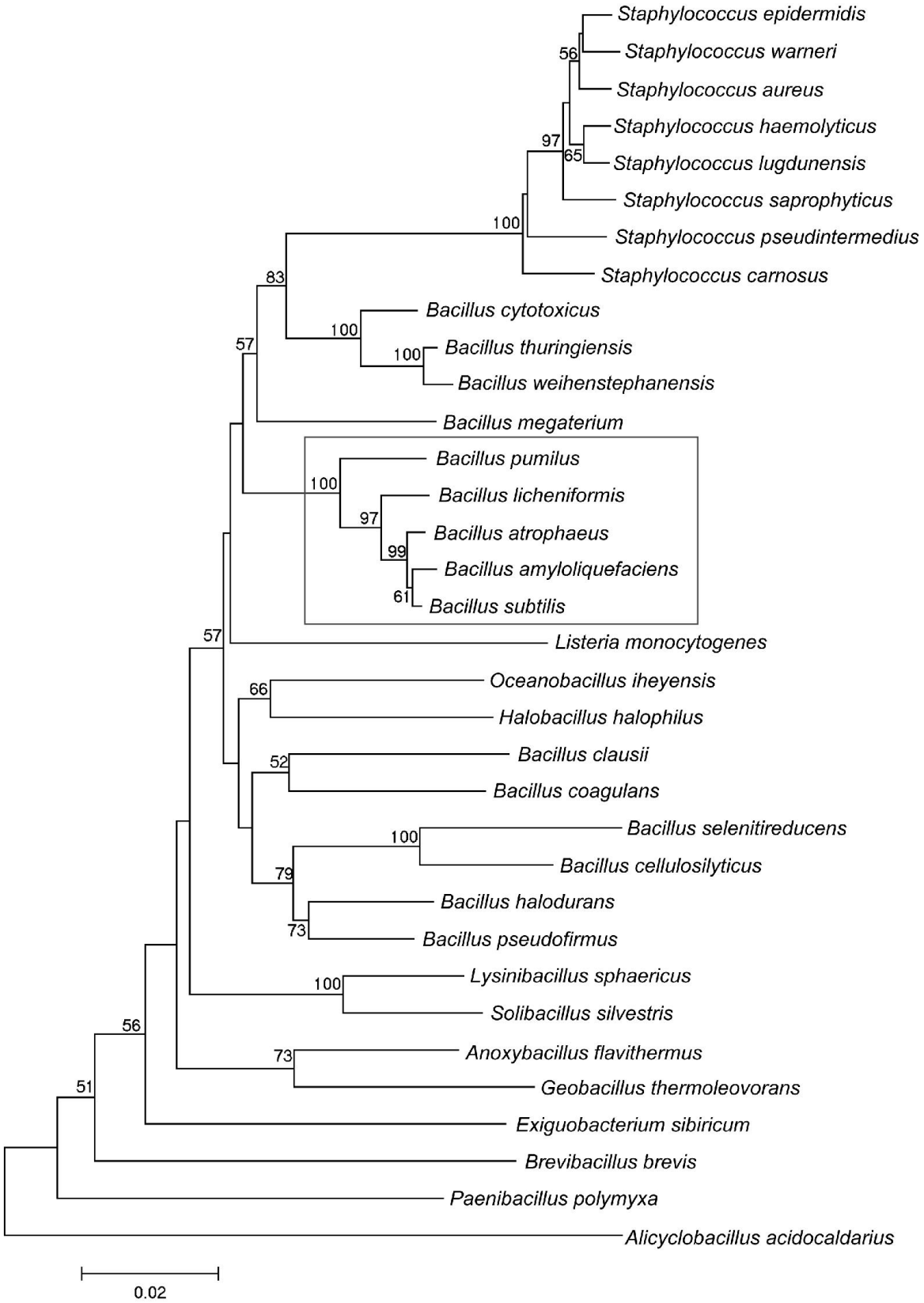

Fig. S1. Neighbor-joining phylogenetic tree of the analyzed species belonging to the order *Bacillales* based on 16S rRNA gene sequences. The species in rectangle are retained to represent the genus *Bacillus*.

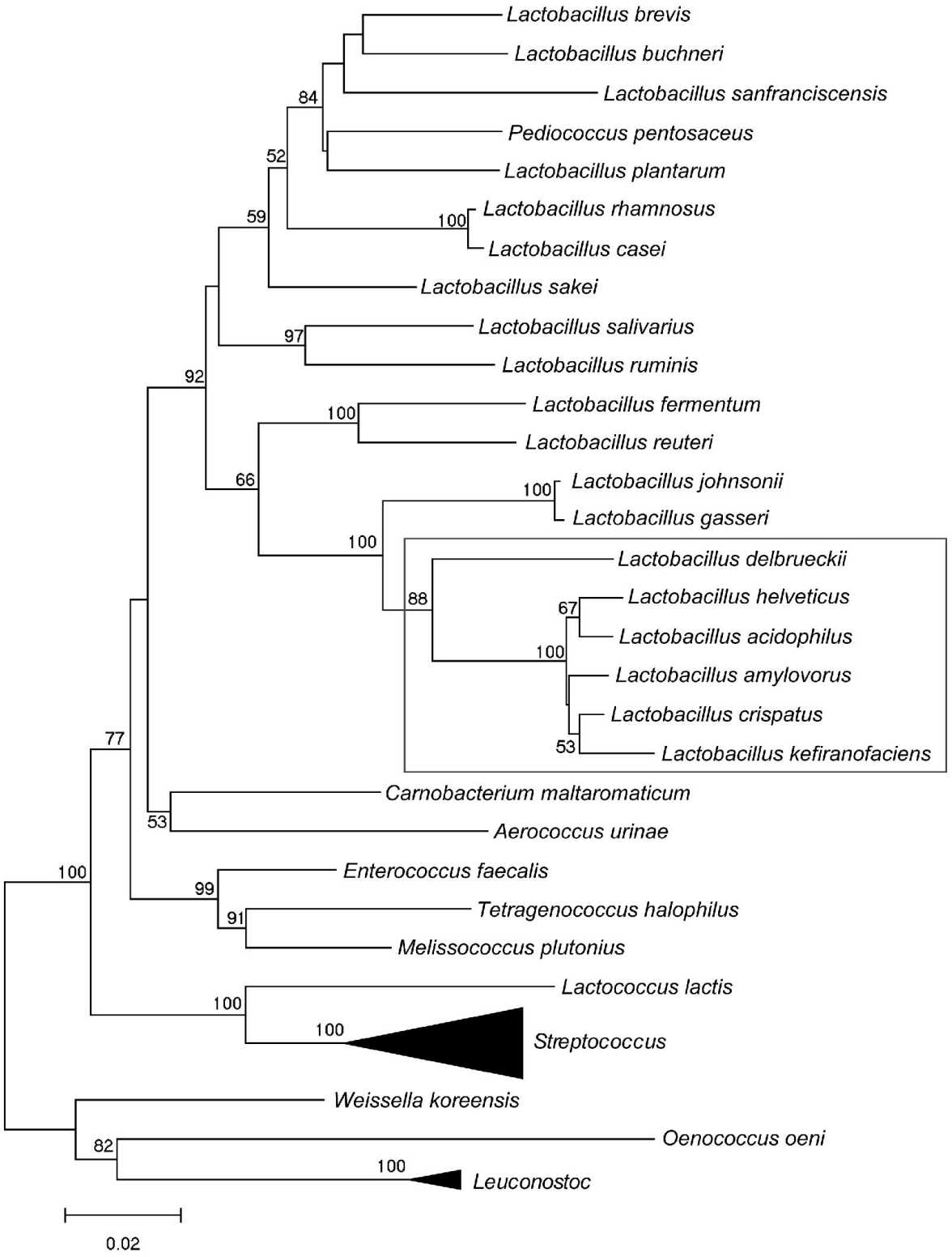

Fig. S2. Neighbor-joining phylogenetic tree of the analyzed species belonging to the order *Lactobacillales* based on 16S rRNA gene sequences. The species in rectangle are retained to represent the genus *Lactobacillus*.

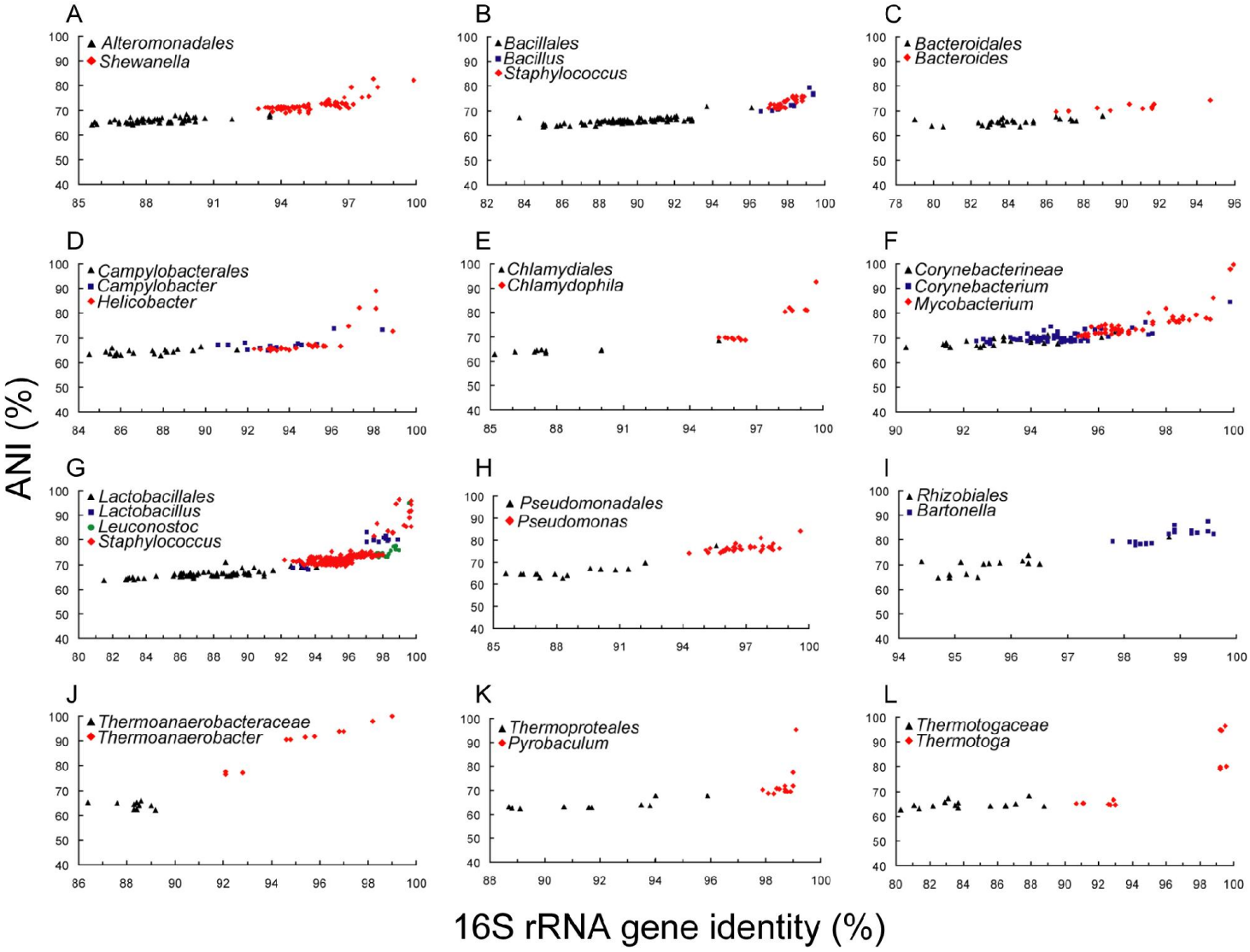

Fig. S3. Relationships between ANI and 16S rRNA gene identity for pairs of genomes. Genomes are from the orders *Alteromonadales* (A), *Bacillales* (B), *Bacteroidales* (C), *Campylobacterales* (D), *Chlamydiales* (E), the suborder *Corynebacterineae* (F), the orders *Lactobacillales* (G), *Pseudomonadales* (H), *Rhizobiales* (I), the family *Thermoanaerobacteraceae* (J), the order *Thermoproteales* (K) and the family *Thermotogaceae* (L). Black triangles represent the inter-genera comparisons in every order/family. Red diamonds and/or blue squares, green circles represent the inter-species comparisons.

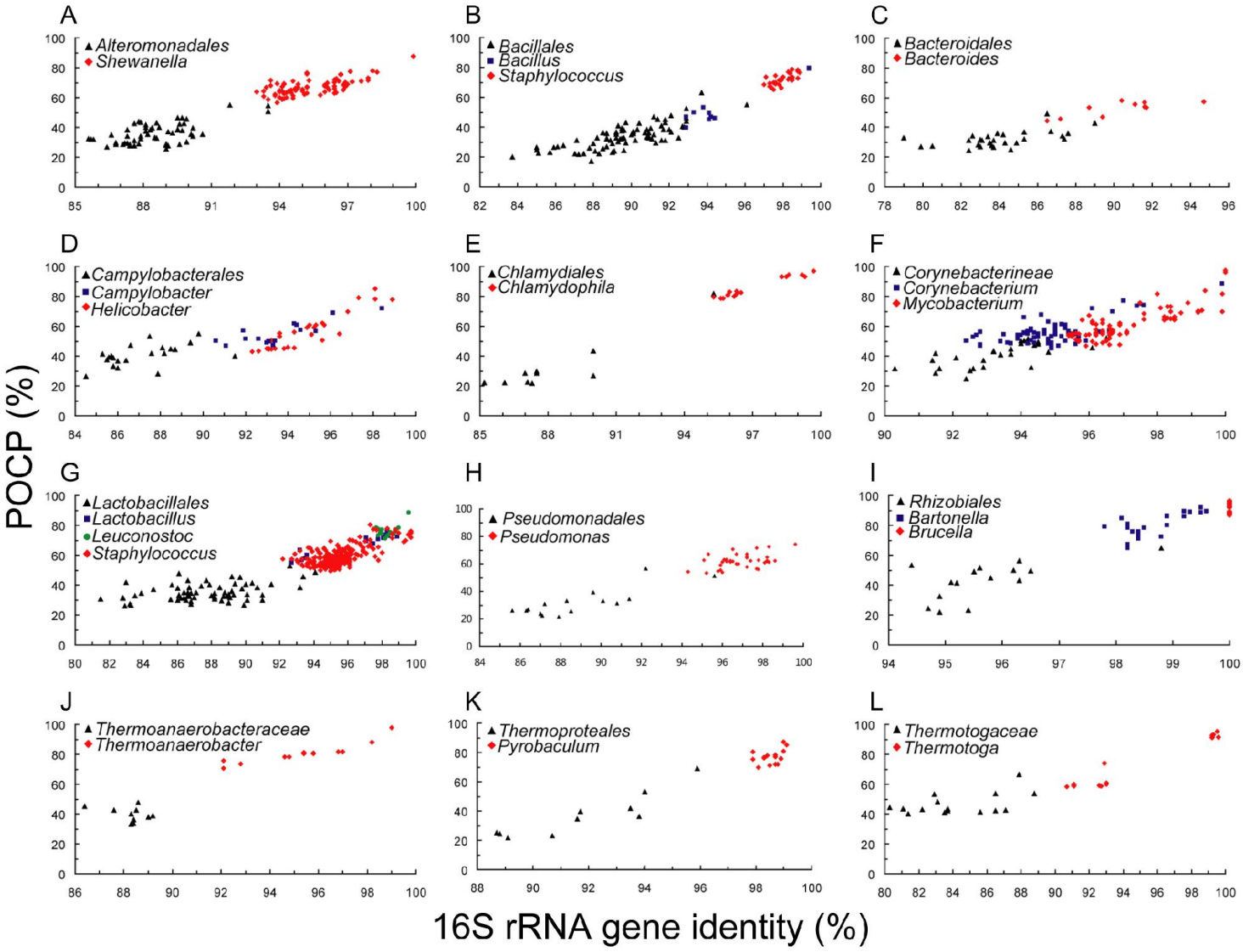

Fig. S4. Relationships between POCP and 16S rRNA gene identity for pairs of genomes. The designations of A-L are same to Fig. S3. Black triangles represent the inter-genera comparisons in every order/family. Red diamonds and/or blue squares, green circles represent the inter-species comparisons.

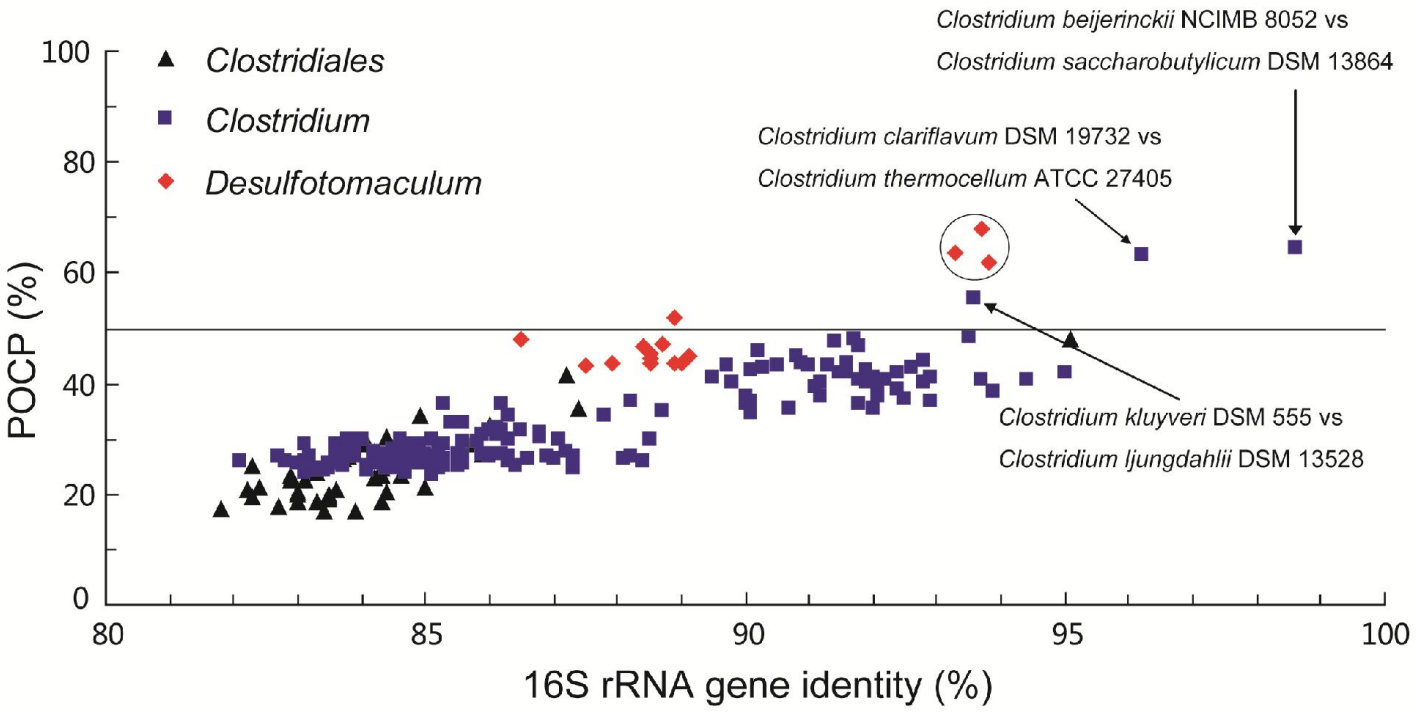

Fig. S5. Relationships between POCP and 16S rRNA gene identity for pairs of genomes from the order *Clostridiales*. Black triangles represent the inter-genera comparisons in the order *Clostridiales*. Red diamonds and blue squares represent the inter-species comparisons. The three diamonds in circle represent the comparisons of *D. carboxydivorans*, *D. reducens* and *D. ruminis* of the genus *Desulfotomaculum*.

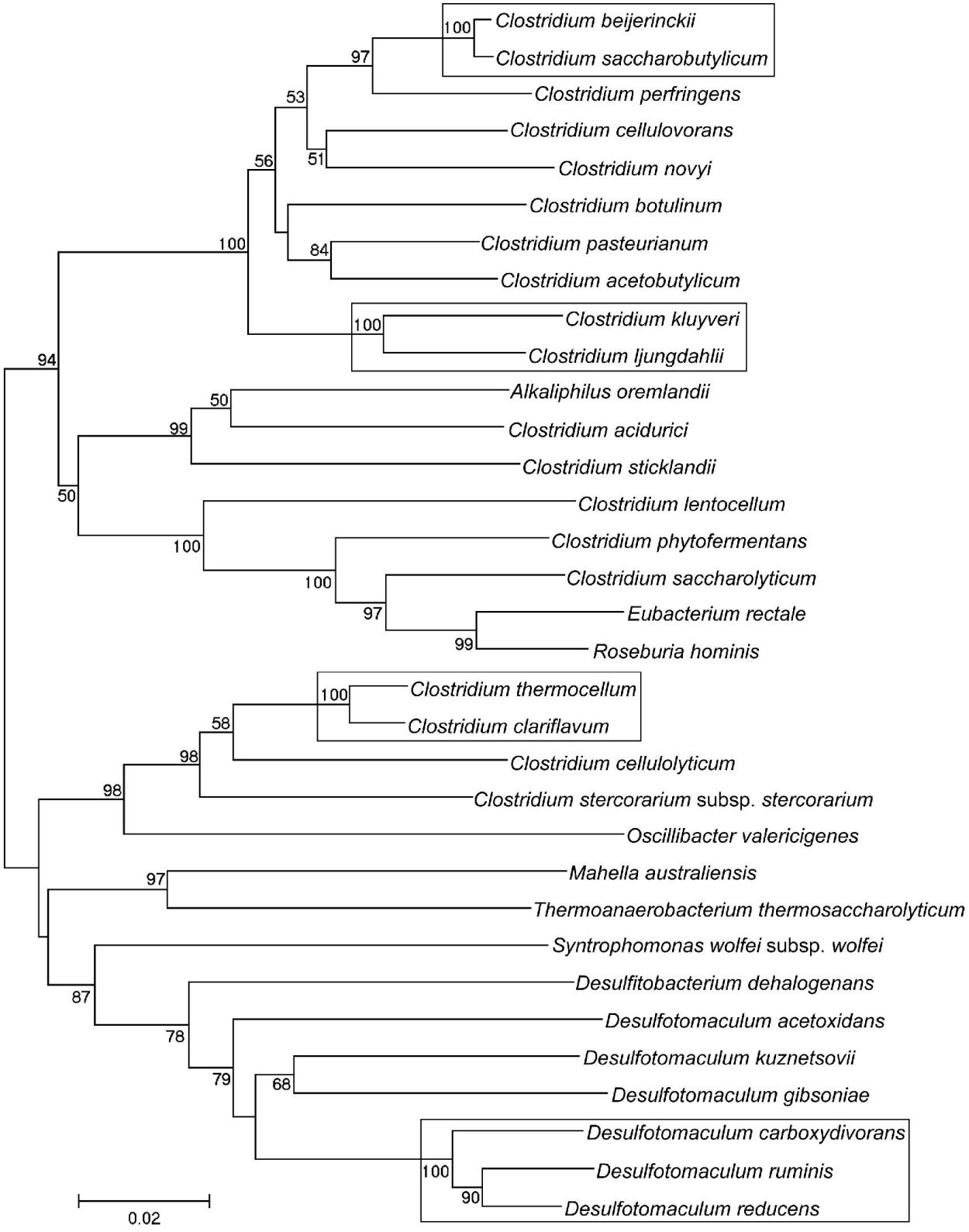

Fig. S6. Neighbor-joining phylogenetic tree of the analyzed species belonging to the order *Clostridiales* based on 16S rRNA gene sequences. The species in rectangle are within a same genus according to the POCP analyses.

Table S1 List of species in this study. For inter-species comparisons, whether the representative stain corresponds to the type strain is indicated (Y: yes; N: no); for inter-genera comparisons, whether the representative species corresponds to the type species is indicated (Y: yes; N: no).

***Shewanella* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Alteromonadales***	**Genome size (Mb)**	**Type species**
Shewanella amazonensis SB2B	4.31	Y	*Alteromonas macleodii* ATCC 27126	4.65	Y
Shewanella baltica OS678	5.29	N	*Colwellia psychrerythraea* 34H	5.37	Y
Shewanella denitrificans OS217	4.55	Y	*Marinobacter hydrocarbonoclasticus* ATCC 49840	3.99	Y
Shewanella frigidimarina NCIMB 400	4.85	N	*Ferrimonas balearica* DSM 9799	4.28	Y
Shewanella halifaxensis HAW-EB4	5.23	Y	*Glaciecola nitratireducens* FR1064	4.13	N
Shewanella loihica PV-4	4.60	Y	*Idiomarina loihiensis* L2TR	2.84	N
Shewanella oneidensis MR-1	4.97	Y	*Pseudoalteromonas haloplanktis* TAC125	3.85	N
Shewanella pealeana ATCC 700345	5.17	Y	*Psychromonas ingrahamii* 37	4.56	N
Shewanella piezotolerans WP3	5.40	Y	*Saccharophagus degradans* 2-40	5.06	Y
Shewanella putrefaciens CN-32	4.66	N	*Shewanella putrefaciens* CN-32	4.66	Y
Shewanella sediminis HAW-EB3	5.52	Y	*Teredinibacter turnerae* T7901	5.19	Y
Shewanella violacea DSS12	4.96	Y			
Shewanella woodyi ATCC 51908	5.94	Y			
***Bacillus* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Bacillales***	**Genome size (Mb)**	**Type species**
Bacillus amyloliquefaciens DSM 7	4.02	Y	*Alicyclobacillus acidocaldarius* subsp. *acidocaldarius* DSM 446	3.02	Y

Bacillus atrophaeus 1942	4.17	N	*Anoxybacillus flavithermus* WK1	2.85	N
Bacillus licheniformis ATCC 14580	4.22	Y	*Brevibacillus brevis* NBRC 100599	6.30	Y
Bacillus pumilus SAFR-032	3.70	N	*Exiguobacterium sibiricum* 255-15	3.04	N
Bacillus subtilis subsp. *subtilis* str. 168	4.22	N	*Geobacillus thermoleovorans* CCB_US3_UF5	3.60	N
***Staphylococcus* Species**			*Halobacillus halophilus* DSM 2266	4.17	Y
Staphylococcus aureus subsp. aureus MRSA252	2.90	N	*Listeria monocytogenes* 08-5923	3.00	Y
Staphylococcus carnosus subsp. carnosus TM300	2.57	N	*Lysinibacillus sphaericus* C3-41	4.82	N
Staphylococcus epidermidis RP62A	2.64	N	*Oceanobacillus iheyensis* HTE831	3.63	Y
Staphylococcus haemolyticus JCSC1435	2.69	N	*Paenibacillus polymyxa* M1	6.23	Y
Staphylococcus lugdunensis HKU09-01	2.66	N	*Solibacillus silvestris* StLB046	3.98	Y
Staphylococcus pseudintermedius HKU10-03	2.62	N	*Staphylococcus aureus* subsp. *aureus* 11819-97	2.85	
Staphylococcus saprophyticus subsp. *saprophyticus* ATCC 15305	2.52	Y	*Bacillus subtilis* subsp. *subtilis* str. 168	4.22	Y
Staphylococcus warneri SG1	2.49	N			
***Bacteroides* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Bacteroidales***	**Genome size (Mb)**	**Type species**
Bacteroides fragilis NCTC 9343	5.24	Y	*Alistipes finegoldii* DSM 17242	3.73	N
Bacteroides helcogenes P 36-108	4.00	Y	*Bacteroides fragilis* NCTC 9343	5.24	Y
Bacteroides salanitronis DSM 18170	4.24	Y	*Odoribacter splanchnicus* DSM 20712	4.39	Y
Bacteroides thetaiotaomicron VPI-5482	6.26	Y	*Paludibacter propionicigenes* WB4	3.69	Y
Bacteroides vulgatus ATCC 8482	5.16	Y	*Parabacteroides distasonis* ATCC 8503	4.81	Y
			Porphyromonas asaccharolytica DSM 20707	2.19	Y
			Tannerella forsythia	3.41	Y

			ATCC 43037		
			Prevotella melaninogenica ATCC 25845	3.17	Y
***Campylobacter* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Campylobacterales***	**Genome size (Mb)**	**Type species**
Campylobacter concisus 13826	2.05	N	*Arcobacter nitrofigilis* DSM 7299	3.19	Y
Campylobacter curvus 525.92	1.97	N	*Wolinella succinogenes* DSM 1740	2.11	Y
Campylobacter fetus subsp. *fetus* 82-40	1.77	N	*Campylobacter fetus* subsp. *fetus* 82-40	1.77	Y
Campylobacter hominis ATCC BAA-381	1.71	N	*Helicobacter pylori* P12	1.67	Y
Campylobacter jejuni subsp. *doylei* 269.97	1.85	N	*Sulfurospirillum deleyianum* DSM 6946	2.31	Y
Campylobacter lari RM2100	1.53	N	*Sulfuricurvum kujiense* DSM 16994	2.82	Y
***Helicobacter* Species**			*Sulfurimonas autotrophica* DSM 16294	2.15	Y
Helicobacter acinonychis str. *Sheeba*	1.55	N			
Helicobacter bizzozeronii CIII-1	1.76	N			
Helicobacter cetorum MIT 99-5656	1.83	Y			
Helicobacter felis ATCC 49179	1.67	Y			
Helicobacter hepaticus ATCC 51449	1.80	N			
Helicobacter mustelae 12198	1.58	Y			
Helicobacter pylori P12	1.67	N			
Helicobacter cinaedi PAGU611	2.08	N			
***Chlamydophila* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Chlamydiales***	**Genome size (Mb)**	**Type species**
Chlamydophila abortus S26/3	1.14	N	*Chlamydia trachomatis* A/HAR-13	1.04	Y
Chlamydophila caviae GPIC	1.17	Y	*Chlamydophila psittaci* 6BC	1.17	Y
Chlamydophila felis Fe/C-56	1.17	N	*Parachlamydia acanthamoebae* UV-7	3.07	Y

Chlamydophila pecorum E58	1.11	Y	*Simkania negevensis* Z	2.63	Y
Chlamydophila pneumoniae TW-183	1.23	Y	*Waddlia chondrophila* WSU 86-1044	2.12	Y
Chlamydophila psittaci 6BC	1.17	Y			
***Lactobacillus* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Lactobacillales***	**Genome size (Mb)**	**Type species**
Lactobacillus acidophilus NCFM	1.99	N	*Aerococcus urinae* ACS-120-V-Col10a	2.08	N
Lactobacillus amylovorus GRL 1112	2.12	N	*Carnobacterium maltaromaticum* LMA28	3.65	N
Lactobacillus crispatus ST1	2.04	N	*Tetragenococcus halophilus* NBRC 12172	2.56	Y
Lactobacillus delbrueckii subsp. *bulgaricus* ATCC 11842	1.86	Y	*Enterococcus faecalis* OG1RF	2.74	Y
Lactobacillus helveticus DPC 4571	2.08	N	*Melissococcus plutonius* ATCC 35311	2.07	Y
Lactobacillus kefiranofaciens ZW3	2.35	N	*Weissella koreensis* KACC 15510	1.44	N
***Leuconostoc* Species**			*Pediococcus pentosaceus* ATCC 25745	1.83	N
Leuconostoc carnosum JB16	1.73	N	*Oenococcus oeni* PSU-1	1.78	Y
Leuconostoc citreum KM20	1.84	N	*Lactococcus lactis* subsp. *lactis* IO-1	2.42	Y
Leuconostoc gasicomitatum LMG 18811	1.95	Y	*Lactobacillus delbrueckii* subsp. *bulgaricus* ATCC 11842	1.86	Y
Leuconostoc gelidum JB7	1.89	N	*Streptococcus pyogenes* MGAS10750	1.94	Y
Leuconostoc kimchii IMSNU 11154	2.11	N	*Leuconostoc mesenteroides* subsp. *mesenteroides* ATCC 8293	2.07	Y
Leuconostoc mesenteroides subsp. *mesenteroides* ATCC 8293	2.07	Y			
***Streptococcus* Species**					
Streptococcus agalactiae NEM316	2.21	N			
Streptococcus dysgalactiae subsp. *equisimilis* GGS_124	2.11	N			
Streptococcus equi subsp. *equi* 4047	2.25	N			
Streptococcus gallolyticus	2.36	N			

subsp. *gallolyticus* ATCC BAA-2069					
Streptococcus gordonii str. *Challis substr*. CH1	2.20	N			
Streptococcus infantarius subsp. *infantarius* CJ18	1.99	N			
Streptococcus intermedius JTH08	1.93	N			
Streptococcus macedonicus ACA-DC 198	2.13	N			
Streptococcus mitis B6	2.15	N			
Streptococcus mutans UA159	2.03	N			
Streptococcus oralis Uo5	1.96	N			
Streptococcus parasanguinis ATCC 15912	2.15	Y			
Streptococcus parauberis KCTC 11537	2.14	N			
Streptococcus pasteurianus ATCC 43144	2.10	N			
Streptococcus pneumoniae Hungary19A-6	2.25	N			
Streptococcus pseudopneumoniae IS7493	2.19	N			
Streptococcus pyogenes MGAS10750	1.94	N			
Streptococcus salivarius CCHSS3	2.22	N			
Streptococcus sanguinis SK36	2.39	N			
Streptococcus suis S735	1.98	Y			
Streptococcus thermophilus LMD-9	1.87	N			
Streptococcus uberis 0140J	1.85	N			
***Pseudomonas* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Pseudomonadales***	**Genome size (Mb)**	**Type species**
Pseudomonas aeruginosa LESB58	6.60	N	*Acinetobacter calcoaceticus* PHEA-2	3.86	Y
Pseudomonas brassicacearum subsp. *brassicacearum* NFM421	6.84	N	*Azotobacter vinelandii* DJ	5.37	N
Pseudomonas entomophila L48	5.89	Y	*Cellvibrio japonicus* Ueda107	4.58	N

Pseudomonas fluorescens Pf-5	7.07	N	*Moraxella catarrhalis* RH4	1.86	N
Pseudomonas fulva 12-X	4.92	N	*Psychrobacter arcticus* 273-4	2.65	N
Pseudomonas mendocina NK-01	5.43	N	*Pseudomonas aeruginosa* LESB58	6.60	Y
Pseudomonas putida KT2440	6.18	N			
Pseudomonas stutzeri A1501	4.57	N			
Pseudomonas syringae pv. tomato str. DC3000	6.54	N			
***Bartonella* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Rhizobiales***	**Genome size (Mb)**	**Type species**
Bartonella bacilliformis KC583	1.45	Y	*Bartonella bacilliformis* KC583	1.45	Y
Bartonella clarridgeiae 73	1.52	N	*Brucella melitensis* ATCC 23457	3.31	Y
Bartonella grahamii as4aup	2.35	N	*Mesorhizobium loti* MAFF303099	7.60	Y
Bartonella henselae str. Houston-1	1.93	Y	*Ochrobactrum anthropi* ATCC 49188	5.06	Y
Bartonella quintana str. Toulouse	1.58	N	*Rhizobium tropici* CIAT 899	6.69	N
Bartonella tribocorum CIP 105476	2.62	Y	*Sinorhizobium fredii* USDA 257	6.89	Y
Bartonella vinsonii subsp. *berkhoffii* str. Winnie	1.80	N			
***Thermoanaerobacter* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the family *Thermoanaerobacteraceae***	**Genome size (Mb)**	**Type species**
Thermoanaerobacter brockii subsp. *finnii* Ako-1	2.34	Y	*Ammonifex degensii* KC4	2.13	Y
Thermoanaerobacter italicus Ab9	2.45	Y	*Carboxydothermus hydrogenoformans* Z-2901	2.40	Y
Thermoanaerobacter mathranii subsp. *mathranii* str. A3	2.31	Y	*Thermacetogenium phaeum* DSM 12270	2,94	Y
Thermoanaerobacter pseudethanolicus ATCC 33223	2.36	Y	*Moorella thermoacetica* ATCC 39073	2.63	Y
Thermoanaerobacter tengcongensis MB4	2.69	Y	*Thermoanaerobacter italicus* Ab9	2.45	N
Thermoanaerobacter wiegelii	2.79	Y			

Rt8.B1					
***Pyrobaculum* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Thermoproteales***	**Genome size (Mb)**	**Type species**
Pyrobaculum aerophilum str. IM2	2.22	Y	*Caldivirga maquilingensis* IC-167	2.08	Y
Pyrobaculum arsenaticum DSM 13514	2.12	Y	*Pyrobaculum islandicum* DSM 4184	1.83	Y
Pyrobaculum calidifontis JCM 11548	2.01	Y	*Thermofilum pendens* Hrk 5	1.78	Y
Pyrobaculum islandicum DSM 4184	1.83	Y	*Thermoproteus tenax* Kra 1	1.84	Y
Pyrobaculum oguniense TE7	2.44	Y	*Vulcanisaeta distributa* DSM 14429	2.37	Y
Pyrobaculum neutrophilum V24Sta	1.77	Y			
***Thermotoga* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the family *Thermotogaceae***	**Genome size (Mb)**	**Type species**
Thermotoga lettingae TMO	2.14	Y	*Fervidobacterium nodosum* Rt17-B1	1.95	Y
Thermotoga maritima MSB8	1.86	Y	*Kosmotoga olearia* TBF 19.5.1	2.30	Y
Thermotoga naphthophila RKU-10	1.81	Y	*Marinitoga piezophila* KA3	2.23	N
Thermotoga neapolitana DSM 4359	1.88	Y	*Petrotoga mobilis* SJ95	2.17	Y
Thermotoga petrophila RKU-1	1.82	Y	*Thermosipho africanus* TCF52B	2.02	Y
Thermotoga thermarum DSM 5069	2.04	Y	*Thermotoga maritima* MSB8	1.86	Y
***Corynebacterium* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the suborder *Corynebacterineae***	**Genome size (Mb)**	**Type species**
Corynebacterium aurimucosum ATCC 700975	2.82	N	*Amycolicicoccus subflavus* DQS3-9A1	4.74	Y
Corynebacterium diphtheriae 31A	2.54	N	*Corynebacterium diphtheriae* 31A	2.54	Y
Corynebacterium efficiens YS-314	3.22	Y	*Gordonia bronchialis* DSM 43247	5.29	Y
Corynebacterium glutamicum ATCC 13032	3.36	Y	*Mycobacterium tuberculosis* H37Rv	4.41	Y
Corynebacterium jeikeium K411	2.48	N	*Nocardia cyriacigeorgica* GUH-2	6.19	N

Corynebacterium kroppenstedtii DSM 44385	2.45	Y	*Rhodococcus opacus* B4	8.73	N
Corynebacterium pseudotuberculosis FRC41	2.34	N	*Segniliparus rotundus* DSM 44985	3.16	Y
Corynebacterium resistens DSM 45100	2.60	Y	*Tsukamurella paurometabola* DSM 20162	4.48	Y
Corynebacterium ulcerans BR-AD22	2.61	N			
Corynebacterium urealyticum DSM 7109	2.37	Y			
Corynebacterium variabile DSM 44702	3.43	N			
Corynebacterium halotolerans DSM 44683	3.14	Y			
***Mycobacterium* Species**					
Mycobacterium abscessus ATCC 19977	5.09	Y			
Mycobacterium africanum GM041182	4.39	N			
Mycobacterium avium 104	5.48	N			
Mycobacterium bovis BCG str. *Pasteur* 1173P2	4.37	N			
Mycobacterium chubuense NBB4	6.34	N			
Mycobacterium gilvum PYR-GCK	5.98	N			
Mycobacterium massiliense str. GO 06	5.07	N			
Mycobacterium intracellulare ATCC 13950	5.40	Y			
Mycobacterium marinum M	6.64	N			
Mycobacterium rhodesiae NBB3	6.42	N			
Mycobacterium smegmatis str. MC2 155	6.99	N			
Mycobacterium tuberculosis H37Rv	4.41	Y			
Mycobacterium ulcerans Agy99	5.81	N			
Mycobacterium vanbaalenii PYR-1	6.49	Y			

Table S2 Taxonomic positions of the 17 genera for inter-species analyses.
The genera grouped into the same family/order are in the same grid.

Domain	Phylum	Order	Family	Genus
Bacteria	*Actinobacteria*	*Corynebacterineae*	*Corynebacteriaceae*	*Corynebacterium*
Bacteria	*Actinobacteria*	*Corynebacterineae*	*Mycobacteriaceae*	*Mycobacterium*
Bacteria	*Bacteroidetes*	*Bacteroidales*	*Bacteroidaceae*	*Bacteroides*
Bacteria	*Chlamydiae*	*Chlamydiales*	*Chlamydiaceae*	*Chlamydia*
Bacteria	*Firmicutes*	*Bacillales*	*Bacillaceae*	*Bacillus*
Bacteria	*Firmicutes*	*Bacillales*	*Staphylococcaceae*	*Staphylococcus*
Bacteria	*Firmicutes*	*Lactobacillales*	*Lactobacillaceae*	*Lactobacillus*
Bacteria	*Firmicutes*	*Lactobacillales*	*Leuconostocaceae*	*Leuconostoc*
Bacteria	*Firmicutes*	*Lactobacillales*	*Streptococcaceae*	*Streptococcus*
Bacteria	*Firmicutes*	*Thermoanaerobacterales*	*Thermoanaerobacteraceae*	*Thermoanaerobacter*
Bacteria	*Proteobacteria*	*Alteromonadales*	*Shewanellaceae*	*Shewanella*
Bacteria	*Proteobacteria*	*Campylobacterales*	*Campylobacteraceae*	*Campylobacter*
Bacteria	*Proteobacteria*	*Campylobacterales*	*Helicobacteraceae*	*Helicobacter*
Bacteria	*Proteobacteria*	*Pseudomonadales*	*Pseudomonadaceae*	*Pseudomonas*
Bacteria	*Proteobacteria*	*Rhizobiales*	*Bartonellaceae*	*Bartonella*
Bacteria	*Thermotogae*	*Thermotogales*	*Thermotogaceae*	*Thermotoga*
Archaea	*Crenarchaeota*	*Thermoproteales*	*Thermoproteaceae*	*Pyrobaculum*

Table S3 List of the species analyzed in this study from the order *Clostridiales*.

***Clostridium* Species**	**Genome size (Mb)**	**Type strain**	**Species from different genera in the order *Clostridiales***	**Genome size (Mb)**	**Type species**
Clostridium acetobutylicum ATCC 824	4.13	Y	*Alkaliphilus oremlandii* OhILAs	3.12	N
Clostridium acidurici 9a	3.11	N	*Eubacterium rectale* ATCC 33656	3.45	N
Clostridium beijerinckii NCIMB 8052	6.00	N	*Thermoanaerobacterium thermosaccharolyticum* DSM 571	2.79	Y
Clostridium botulinum A2 str. Kyoto	4.16	N	*Mahella australiensis* 50-1 BON	3.14	Y
Clostridium cellulolyticum H10	4.07	Y	*Roseburia hominis* A2-183	3.59	N
Clostridium cellulovorans 743B	5.26	Y	*Desulfitobacterium dehalogenans* ATCC 51507	4.32	Y
Clostridium clariflavum DSM 19732	4.90	Y	*Oscillibacter valericigenes* Sjm18-20	4.41	Y
Clostridium kluyveri DSM 555	3.96	Y	*Syntrophomonas wolfei* subsp. *wolfei* str. Goettingen G311	2.94	Y
Clostridium lentocellum DSM 5427	4.71	Y	*Desulfotomaculum ruminis* DSM 2154	3.97	N
Clostridium ljungdahlii DSM 13528	4.63	Y	*Clostridium cellulovorans* 743B	5.26	N
Clostridium novyi NT	2.55	N			
Clostridium pasteurianum BC1	4.99	N			
Clostridium perfringens ATCC 13124	3.26	Y			
Clostridium phytofermentans ISDg	4.85	Y			
Clostridium saccharobutylicum DSM 13864	5.11	Y			
Clostridium saccharolyticum WM1	4.66	Y			
Clostridium stercorarium subsp. *stercorarium* DSM 8532	2.97	Y			
Clostridium sticklandii	2.72	Y			

DSM 519					
Clostridium thermocellum ATCC 27405	3.84	Y			
***Desulfotomaculum* Species**					
Desulfotomaculum acetoxidans DSM 771	4.55	Y			
Desulfotomaculum carboxydivorans CO-1-SRB	2.89	Y			
Desulfotomaculum gibsoniae DSM 7213	4.86	Y			
Desulfotomaculum kuznetsovii DSM 6115	3.60	Y			
Desulfotomaculum reducens MI-1	3.61	N			
Desulfotomaculum ruminis DSM 2154	3.97	Y			