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Disulfide Bond Formation and Cysteine Exclusion in Gram-positive Bacteria^{*†‡}

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Most secretion pathways in bacteria and eukaryotic cells are challenged by the requirement for their substrate proteins to mature after they traverse a membrane barrier and enter a reactive oxidizing environment. For Gram-positive bacteria, the mechanisms that protect their exported proteins from misoxidation during their post-translocation maturation are poorly understood. To address this, we separated numerous bacterial species according to their tolerance for oxygen and divided their proteomes based on the predicted subcellular localization of their proteins. We then applied a previously established computational approach that utilizes cysteine incorporation patterns in proteins as an indicator of enzymatic systems that may exist in each species. The Sec-dependent exported proteins from aerobic Gram-positive Actinobacteria were found to encode cysteines in an even-biased pattern indicative of a functional disulfide bond formation system. In contrast, aerobic Gram-positive Firmicutes favor the exclusion of cysteines from both their cytoplasmic proteins and their substantially longer exported proteins. Supporting these findings, we show that Firmicutes, but not Actinobacteria, tolerate growth in reductant. We further demonstrate that the actinobacterium *Corynebacterium glutamicum* possesses disulfide-bonded proteins and two dimeric Dsb-like enzymes that can efficiently catalyze the formation of disulfide bonds. Our results suggest that cysteine exclusion is an important adaptive strategy against the challenges presented by oxidative environments.

Proper protein targeting and maturation are fundamental to the homeostasis of all organisms. Whereas many of the features that direct the membrane topology and localization of a protein

are known, the strategies employed by cells to promote the proper maturation of their proteins under different environmental conditions are not entirely understood.

Protein secretion is a dynamic and complex process due to the membrane separation of synthesis and folding (1–3). The majority of secreted proteins are translocated across a membrane into an oxidizing environment, where their folding and maturation occur. Bacteria have evolved several secretory pathways for translocating their proteins across the plasma membrane that can be categorized as either Sec-dependent or Sec-independent (4, 5). The Sec-dependent pathway, which is essential in all organisms, is the major route of protein secretion.

In bacteria, the majority of proteins that are targeted for Sec-dependent secretion contain a canonical N-terminal signal peptide that is either directly recognized by SecA or delivered to SecA with the aid of the secretion-dedicated chaperone SecB (6). SecA then drives the translocation of the protein through the SecYEG channel. Alternatively, most plasma membrane proteins and a small number of secretory proteins are recognized as ribosome-bound nascent chains by the signal recognition particle, which directs them to SecYEG, where they are co-translationally inserted into or translocated across the plasma membrane. Upon emergence from the translocon, nascent secretory proteins mature after removal of their signal peptide by either the type I signal peptidase or, in the case of lipoproteins, the type II signal peptidase (7).

Although the obstacles associated with protein folding are fundamentally similar across all organisms, the machinery involved in this process and the environment where it occurs can differ dramatically (8–10). In Gram-negative bacteria, secreted proteins mature in the oxidative periplasm with the aid of molecular chaperones and disulfide bond-forming machinery (11–13). Gram-positive bacteria lack this protected compartment due to the absence of an outer membrane. Thus, their secreted proteins have to mature in the unregulated environment of the thick, negatively charged cell wall composed of multilayered networks of peptidoglycan and teichoic acids (supplemental Fig. S1).

Cysteine residues perform a variety of essential functions in proteins, from metal binding and enzyme catalysis to structural stability through the formation of disulfide bonds (14). However, the reactive thiol side chain that is amenable to these roles can also negatively impact the function of a protein by the formation of incorrect disulfide bonds or other deleterious modi-

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fications. The high reactivity of cysteine thiols and their susceptibility to attack by reactive oxygen species likely create an oxidative selection pressure on cysteine-containing proteins that relates directly to their environment and localization (15). Consequently, bacteria have evolved systems that keep their cytoplasm in a reducing state and maintain the proper oxidation state of the cysteines in their extracellular proteins (16–19).

The periplasm-localized disulfide oxidoreductases of the Dsb family have been best characterized in *Escherichia coli*. *E. coli* DsbA is a soluble monomeric enzyme with a thioredoxin-like fold and a Cys-X-X-Cys active site that aids in the maturation of secreted proteins by catalyzing their disulfide bond formation immediately after their translocation into the periplasm (supplemental Fig. S1) (20, 21). DsbA is maintained in its oxidized state by the membrane-bound protein DsbB (22, 23). DsbC and DsbG, homodimers that function as disulfide isomerases or reductases, and DsbE, a thioredoxin-like protein involved in cytochrome *c* maturation, are all kept in their reducing state by the membrane-bound protein DsbD (19, 24–26). Together, the Dsb proteins in Gram-negative bacteria aid in the maturation of exported proteins and protect them from improper oxidation.

Several studies have searched for homologs of the known disulfide bond-forming machinery to predict whether certain bacteria are capable of oxidizing their secreted proteins (16, 27, 28). Mallick *et al.* (29) were the first to demonstrate that hyperthermophilic Archaea, which possess disulfide bonds in their cytoplasmic proteins, have a bias for encoding proteins with an even number of cysteine residues. Recently, Beckwith and co-workers (27) applied a similar reasoning to examine whether the predicted exported proteins from a large number of bacterial species have an even-number cysteine bias. These results were then correlated to whether DsbA and DsbB homologs were present in the corresponding genomes.

We used a combined computational and experimental approach to investigate the different strategies Gram-positive bacteria have evolved to compensate for changes in the oxidative environment that accompanies the localization of their proteins and their tolerance for oxygen. In agreement with recent work, our predictions indicate that Gram-positive Actinobacteria likely form disulfide bonds in their exported proteins (as do Gram-negative Proteobacteria) and that aerobic Gram-positive Firmicutes tend to generally exclude cysteines from their exported proteins (27). Additionally, we also found that aerobic Firmicutes have substantially longer exported proteins and favor the exclusion of cysteines from their cytoplasmic proteins. Supporting these predictions, we show that Firmicutes, but not Actinobacteria, can tolerate high levels of reductant, indicating that Firmicutes do not rely on disulfide-bonded proteins for growth. We observed several disulfide-bonded proteins in Actinobacteria, but not in Firmicutes, and show that two different dimeric Dsb-like proteins from the actinobacterial species *Corynebacterium glutamicum* are capable of productive disulfide bond formation. These results provide further insight into how organisms combat oxidative challenges and how their proteins have coevolved in the absence or presence of various enzymatic redox systems to overcome these obstacles.

EXPERIMENTAL PROCEDURES

Acquisition, Classification, and Homology Reduction of Bacterial Genomes—Proteomes from the 340 analyzed bacterial species were downloaded from the European Bioinformatics Institute (EMBL-EBI Database). When several sequenced strains from a given species were available, one proteome from an ATCC strain was chosen to represent that species. The species were manually classified into their respective phyla (Proteobacteria, Firmicutes, and Actinobacteria) and for their ability to grow in the presence of oxygen (strict aerobes and facultative anaerobes *versus* strict anaerobes). Supplemental Table 1 shows the species distribution in each category. The individual bacterial proteomes were restricted to those proteins that were at least 80 amino acid residues in length. To prevent bias due to redundant protein families in the individual species, each bacterial proteome sequence set was internally homology-reduced using CD-HIT with a sequence identity threshold of 40% (30). These two initial screening steps retained 945,399 amino acid sequences for analysis and discarded 121,981 proteins based on homology reduction and 64,920 sequences as peptides.

Protein Topology Prediction and Cysteine Analysis—Protein sequences were classified according to predictions for the presence of transmembrane (TM)⁴ regions using SCAMPI and signal peptidase (SPase) type I and II signal peptides using SignalP 3.0 and LipoP 1.0, respectively (supplemental Fig. S2) (31–33). The proteome sets for each of the four classifications were compiled and analyzed for their cysteine content: (i) lipoproteins, containing a SPase II signal peptide and no TM regions (21,826 or 2.3% of the sequences); (ii) secreted proteins, containing a SPase I signal peptide, no SPase II signal peptide, and no TM regions (77,419 or 8.2% of the sequences); (iii) TM proteins, containing at least one TM region, no SPase II signal peptide, but possibly a SPase I signal peptide (213,419 or 22.6% of the sequences); and (iv) cytoplasmic proteins, containing no signal peptide or TM regions (631,735 or 66.8% of the sequences). Sequences predicted to contain a SPase I signal peptide by only one of the neural network and hidden Markov model predictors and those with both a SPase II signal peptide and a TM region were discarded (1,000 or 0.1% of the sequences). In all sequence sets, the predicted signal peptides were removed and analyzed independently for cysteine occurrence. For lipoproteins, the N-terminal cysteine was excluded from the calculations.

Experimentally Investigated Bacterial Strains—*Arthrobacter luteus* (ATCC 21606), *Bacillus cereus* (ATCC 14579), *Bacillus subtilis* (ATCC 6051), *Corynebacterium glutamicum* (ATCC 13032), *E. coli* (UPEC (UT189)) (34), *Enterococcus faecalis* (ATCC 29212), *Haemophilus influenzae* (SMI 120 strain 1 type B), *Micrococcus luteus* (ATCC 11880), *Pseudomonas aeruginosa* (ATCC 33357), *Rhodococcus equi* (ATCC 6939), *Salmonella typhimurium* (ATCC 14028), *Staphylococcus aureus* (ATCC 25923), and *Streptococcus pyogenes* (ATCC 11434) were used in this study.

⁴ The abbreviations used are: TM, transmembrane; SPase, signal peptidase; DTT, dithiothreitol; BisTris, 2-[bis(2-hydroxyethyl)amino]-2-(hydroxymethyl)propane-1,3-diol.

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Two-dimensional SDS-PAGE—Bacterial cultures were grown in brain-heart infusion medium to mid-log phase and sedimented by centrifugation at $15,000 \times g$ for 10 min. Pellets were resuspended in 20 mM Tris-Cl, pH 7.5, 150 mM NaCl, 1% Nonidet P-40, 20 mM *N*-ethylmaleimide, and 200 μ g/ml mutanolysin (Sigma) and rotated for 30 min at 4 °C. Lysates were then adjusted to 1% SDS and 8 M urea and passed through a French press. Insoluble cell debris was removed by centrifugation at $15,000 \times g$ for 10 min. Samples were resolved in the first dimension via nonreducing SDS-PAGE. For the second dimension, the lanes were excised with a razor, placed in sample buffer containing 100 mM dithiothreitol (DTT), heated for 5 min at 95 °C, overlaid onto a 1.0-mm 2D-Well NuPAGE 10% BisTris gel, separated by electrophoresis, and resolved by Coomassie Blue staining (35).

Bacterial DTT Susceptibility Assay—Bacterial strains were grown at 37 °C to mid-log phase in brain-heart infusion medium supplemented with 10% glucose bouillon (10% horse serum, 1% meat extract, 1% peptone, 1% dextrose, and 0.5% NaCl, pH 7.5). Cultures were diluted to $OD_{600\text{ nm}} = 0.050$ in unconditioned growth medium and transferred to wells of a Honeycomb 2 plate (Isotron) containing DTT. The growth kinetics were monitored by changes in the $OD_{600\text{ nm}}$ at 37 °C using a Bioscreen C plate reader (LabSystems). For *H. influenzae*, the growth medium was supplemented with 0.2 mM NAD.

DNA Constructs—The coding sequences for CG0026, CG0354, and CG2799 from *C. glutamicum* (ATCC 13032) and DsbA (C4447) from *E. coli* (UT189) were PCR-amplified from chromosomal DNA with 5'-primers that excluded the N-terminal signal peptide and replaced it with a His₆ tag. The PCR products were subcloned into the pET21d expression vector (Novagen), and each construct was verified by DNA sequencing.

Recombinant Protein Expression, Purification, and Size Exclusion Chromatography—Bacterial cultures were grown to $A_{600\text{ nm}} = 0.6$; induced with 1 mM isopropyl β -D-thiogalactopyranoside for 3 h; sedimented; and resuspended in 20 mM Tris, pH 7.5, 500 mM NaCl, and 20 mM imidazole with 200 μ g/ml lysozyme and 200 μ g/ml DNase I. Following rotation for 30 min at 4 °C, the lysates were sonicated and clarified by centrifugation at $15,000 \times g$ for 10 min. Proteins were then isolated using nickel-Sepharose 6 Fast Flow (GE Healthcare); washed; eluted with 500 mM imidazole in 20 mM Tris, pH 7.5, and 500 mM NaCl; and dialyzed against 20 mM Tris, pH 7.5.

Protein concentrations were determined by the absorbance at 280 nm using their respective extinction coefficients and molecular masses. Size exclusion chromatography was performed on an ÄKTA FPLC system (GE Healthcare) equipped with a Superdex 75 column. The molecular mass standards were run according to the manufacturer's instructions; all protein samples were dialyzed into and separated with phosphate-buffered saline, pH 7.4; and the absorbance was monitored at 260 and 280 nm.

Determination of Oxidase and Reductase Activities—Oxidase activity was assayed by analyzing the cleavage of cCMP by reduced RNase A refolded in the presence of each enzyme (36). RNase A (Sigma) was reduced by overnight incubation at 27 °C in 100 mM Tris acetate, pH 8.0, containing 6 M guanidine HCl

and 140 mM DTT. DTT and guanidine were removed by size exclusion chromatography with Sepharose G-10 beads. RNase A reduction was verified by the mobility shift created by the large 525-Da free thiol-modifying agent maleimide-polyethylene oxide-biotin and with 5,5'-dithiobis(2-nitrobenzoic acid) (supplemental Fig. S11A). Each enzyme was assayed in a 1-ml solution containing 100 mM Tris acetate, pH 8.0, 2 mM EDTA, 0.2 mM GSSG, 1 mM GSH, and 10 μ M reduced RNase A. Following a 2-min incubation, cCMP was added at a final concentration of 4.5 mM, and the absorbance at 296 nm was recorded. Reductase activity was determined at 27 °C by measuring the turbidity of insulin at $OD_{600\text{ nm}}$ that occurs upon precipitation of the reduced B chain (37). Insulin (Sigma) was resuspended at 5 mg/ml in 100 mM potassium acetate, pH 7.5, by dropping the pH to 3.0 with HCl and titrating back to pH 7.5 with KOH prior to adjusting to 2 mM EDTA and 10 μ M DTT. The enzymes were assayed in a 200- μ l solution containing 100 mM potassium acetate, pH 7.5, 2 mM EDTA, 320 μ M DTT, and 200 μ g of insulin.

RESULTS

Aerobic Gram-positive Actinobacteria Are Predicted to Stabilize Exported Proteins with Disulfide Bonds, Whereas Aerobic Firmicutes Exclude Cysteines—To gain an understanding of how Gram-positive bacteria have evolved to protect and stabilize their proteins in the different oxidative environments, we compared the cysteine incorporation patterns in different types of proteins from Gram-positive bacteria (Firmicutes and Actinobacteria) with those from Gram-negative bacteria (Proteobacteria). Initially, we downloaded the available proteomes from the 340 bacterial species of the major phyla of Gram-positive and Gram-negative bacteria (EMBL-EBI Database). The strict anaerobic species were then classified as "anaerobes," and all bacteria capable of aerobic respiration, including strict aerobes and facultative anaerobes, were grouped together as "aerobes" (supplemental Fig. S1 and Table 1).

Computational analysis was performed on the proteomes to separate them into the following subproteomes: lipoproteins, secreted proteins, TM proteins, and cytoplasmic proteins (Fig. 1A and supplemental Fig. S2). Because only cysteines with free thiols in mature secreted proteins are susceptible to oxidation, the signal peptides were removed and examined separately, and the lipoprotein lipid-linked N-terminal cysteines were excluded prior to analysis (supplemental Fig. S3) (38).

Bacteria with the capacity for disulfide bond formation have previously been shown to favor the incorporation of cysteines in pairs (*i.e.* an even-biased manner) in their substrate proteins (27, 29, 39). Thus, this characteristic can be used as a strong indication of disulfide bond formation. We first analyzed the average cysteine distribution in all of the predicted secreted proteins, lipoproteins, TM proteins, and signal peptides from aerobic and anaerobic Gram-positive and Gram-negative bacteria.

All but anaerobic Gram-positive bacteria favored an even number of cysteines in their secreted proteins and lipoproteins, and no obvious trends were observed in the signal peptides and TM protein loops (supplemental Fig. S3). However, when the exported protein (lipoprotein and secreted protein) subproteomes were analyzed separately, a large number of those from

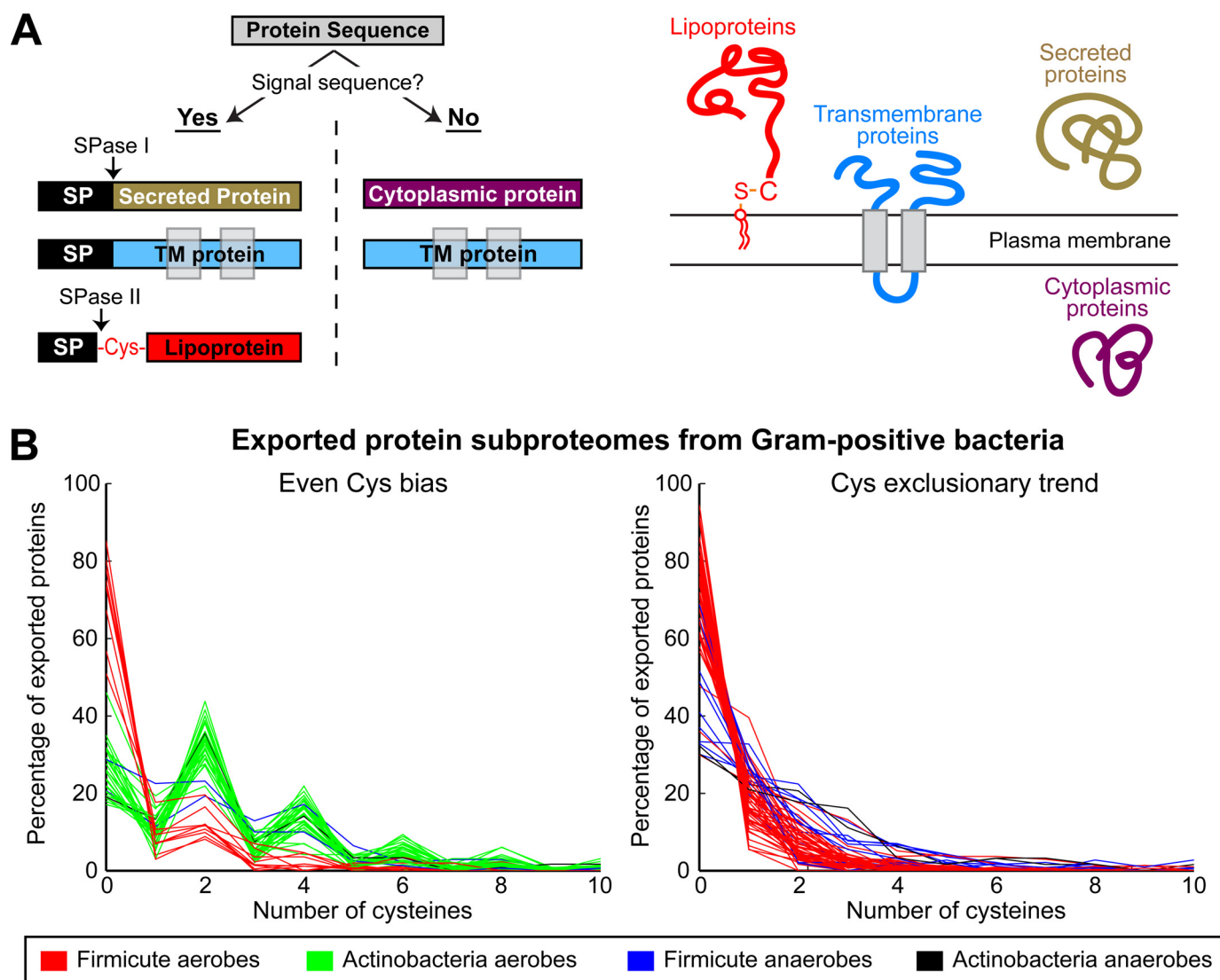


FIGURE 1. Exported proteins from aerobic Firmicutes largely exclude cysteines, whereas those from Actinobacteria favor paired cysteines. A, the schematics depict the protein characteristics used to classify the subproteomes. The SPase type I and II signal peptides (SP) and TM regions were used to identify and sort each proteome into its respective topology or subproteome. B, the exported proteins (secreted proteins and lipoproteins) from Actinobacteria show a profile suggestive of disulfide bond formation, whereas those from Firmicutes preferentially exclude cysteines. The subproteomes containing the exported proteins from Gram-positive bacteria were separated into two major cysteine incorporation patterns. All aerobic Gram-positive Actinobacteria possess an “even-numbered cysteine bias” (left panel) in their exported proteins, and the large majority of Gram-positive Firmicutes show a cysteine exclusion trend (right panel). Each line represents the cysteine distribution in the exported proteins from one bacterial proteome.

aerobic Gram-positive bacteria (~60%) were found to average less than one cysteine/protein and to lack the even-numbered cysteine bias (supplemental Fig. S4). From these observations, we were able to classify the vast majority of all of the exported protein subproteomes by one of two patterns.

The first is an “even cysteine-biased” pattern defined by an up-and-down trend in the percentage of exported proteins that correspond to an even (up) and odd (down) number of cysteines (0 Cys > 1 Cys and 2 Cys > 1 Cys), and the second is a “cysteine exclusion trend” (0 Cys > 1 Cys > 2 Cys) (Table 1). Only 5% of the exported protein subproteomes did not follow either pattern and were labeled as “remaining,” and those with fewer than 25 proteins were excluded from further analysis (supplemental Fig. S5 and Table 1). Following this separation, we could confirm a conclusion in a recent study that an obvious distinction

exists between the two major phyla of Gram-positive bacteria (27).

All exported protein subproteomes from aerobic Actinobacteria fell into the even cysteine-biased pattern, whereas those from aerobic Firmicutes followed mainly the cysteine exclusion trend (Fig. 1B, Table 1, and supplemental Fig. S6). The exported proteins from aerobic Firmicutes have a mean of ~0.5 Cys, and an astonishing 70% of them are devoid of cysteine residues, compared with only 33% in anaerobic Firmicutes (supplemental Fig. S7). In contrast, the exported proteins from aerobic Actinobacteria have a mean of ~2.2 Cys, only 25% of them lack cysteine residues, and about 60% contain even numbers of cysteines, which is significantly higher than for aerobic Gram-negative Proteobacteria (supplemental Figs. S3A, S6, and S7). Thus, the maturation of the exported proteins from aerobic Acti-

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TABLE 1

Distribution of bacterial exported and cytoplasmic subproteomes to the indicated cysteine incorporation trends

The number of subproteomes that were classifiable into one of the two cysteine incorporation trends is displayed with respect to the total number of subproteomes that were analyzed from each class of bacteria. For the specific species distribution and those that were not classifiable by these trends, see the [supplemental data](#).

		Phyla	Exported proteins		Cytoplasmic proteins	
			Even bias	Exclusion	Semi-normal	Exclusion
Gram-positive bacteria						
Aerobic	Actinobacteria	31/31		27/31	4/31	
	Firmicutes	10/64	51/64	17/64	43/64	
Anaerobic	Actinobacteria	1/3	2/3	2/3	1/3	
	Firmicutes	2/20	11/20	16/20	2/20	
Gram-negative Proteobacteria						
Aerobic	Alpha	49/52		50/52	2/52	
	Beta	34/34		33/34	1/34	
Anaerobic	Delta	3/5	2/5	4/5		
	Epsilon	12/12		8/12	1/12	
	Gamma	80/86		83/86	1/86	
	Alpha	5/13	4/13	12/13	1/13	
	Beta	2/2		2/2		
	Delta	2/11	9/11	11/11		
	Epsilon	1/1		1/1		
	Gamma	5/6		5/6		

nobacteria likely involves disulfide bond formation, and aerobic Firmicutes tend to exclude cysteines from their exported proteins (27).

Exported Proteins from Firmicutes Are Significantly Longer—To understand how the exported proteins from Firmicutes have evolved in the absence of disulfide bond stabilization, we examined their length in comparison with that of their cytoplasmic proteins. In Firmicutes, Actinobacteria, and Gram-negative bacteria, the average length of their mature exported proteins is longer than that of their respective cytosolic proteins, which are ~325 amino acids in length (Fig. 2A). However, the mature exported proteins from aerobic Firmicutes have an average length that is ~70 amino acids longer than that of their cytoplasmic proteins. This difference is substantially greater than the 7- and 16-amino acid increases seen in aerobic Actinobacteria and Gram-negative Proteobacteria, respectively. Although different cell wall attachment motifs in the Firmicute exported proteins may contribute to the increase in length, it is also possible that these proteins require more extensive non-covalent structural interactions to stabilize their proteins in the absence of disulfide bonds.

Aerobic Firmicutes Exclude Cysteines from Their Cytoplasmic Proteins—Analysis of the cytoplasmic subproteomes revealed that aerobic Gram-positive bacteria encode cysteines with a low frequency, resulting in a unique distribution profile that peaks at 0 Cys compared with 2 Cys in Gram-negative Proteobacteria and Gram-positive anaerobes (Fig. 2B and [supplemental Fig. S8](#)). Upon investigating the population variations of the cytoplasmic subproteomes from Gram-positive bacteria, we were able to assign them to either a cysteine exclusion trend (0 Cys > 1 Cys > 2 Cys) or a “semi-normal cysteine trend” (1 Cys > 0 Cys). Using these parameters, the majority of aerobic Firmicutes (67%) were found to have a cysteine exclusion trend in their cytoplasmic proteins, whereas Actinobacteria and anaerobic Firmicutes had a strong inclination toward a semi-normal cysteine trend (Fig. 2C and Table 1). Thus, aerobic Firmicutes appear to possess a general tendency to exclude cysteines from their proteins in comparison with other bacteria.

Actinobacteria Possess Disulfide-bonded Proteins—To examine whether aerobic Actinobacteria possess disulfide-bonded

proteins, whole cell lysates from *C. glutamicum* were separated and resolved by two-dimensional nonreducing/reducing SDS-PAGE (35). In accordance with the predictive data, both intermolecular (Fig. 3, below the *diagonal*) and large intramolecular (above the *diagonal*) disulfide-bonded proteins were present in *C. glutamicum* and in the control lysate from *E. coli*. As expected, no obvious disulfide-bonded proteins were detected in the lysate from the Firmicute *S. aureus*. Because some exported Firmicute proteins do not bind to the cell envelope following secretion, concentrated conditioned LB media from *S. aureus* and other Firmicutes were examined, but only a few proteins were observed, and none of them showed mobility shifts attributed to disulfide bond formation (data not shown).

Firmicutes Are Resistant to Highly Reducing Conditions—Many proteins contain intramolecular disulfide bonds that do not cause a mobility shift when comparing their nonreduced and reduced states following separation by SDS-PAGE. Therefore, as a second test of our predictions, we investigated the ability of various bacteria to tolerate growth in rich media containing the disulfide bond-reducing agent DTT. We hypothesized that Firmicutes, whose exported proteins are predicted to lack disulfide bonds, would tolerate higher levels of DTT in comparison with the disulfide bond-forming Actinobacteria. Strikingly, 20 mM DTT had little effect on the liquid growth of various aerobic Firmicute species, and more impressively, they were all capable of growth in 100 mM DTT (Fig. 4 and [supplemental Fig. S9](#)). In contrast, the growth of numerous species from Actinobacteria and Gram-negative Proteobacteria was sensitive to DTT in a concentration-dependent manner, with 20 mM DTT almost entirely inhibiting growth.

Actinobacteria Encode Two Dimeric Oxidoreductases Capable of Productive Disulfide Bond Formation—In Gram-negative Proteobacteria, the enzymes that facilitate disulfide bond formation are oxidoreductases or DsbA homologs that reside in the periplasm and are recharged by a membrane-bound DsbB homolog (12). Several studies have identified the presence of DsbA and DsbB homologs across bacteria and used them as an indication of disulfide bond formation (16, 27, 28). To examine whether *C. glutamicum* possesses Dsb homologs capable of catalyzing disulfide bond formation, we identified three puta-

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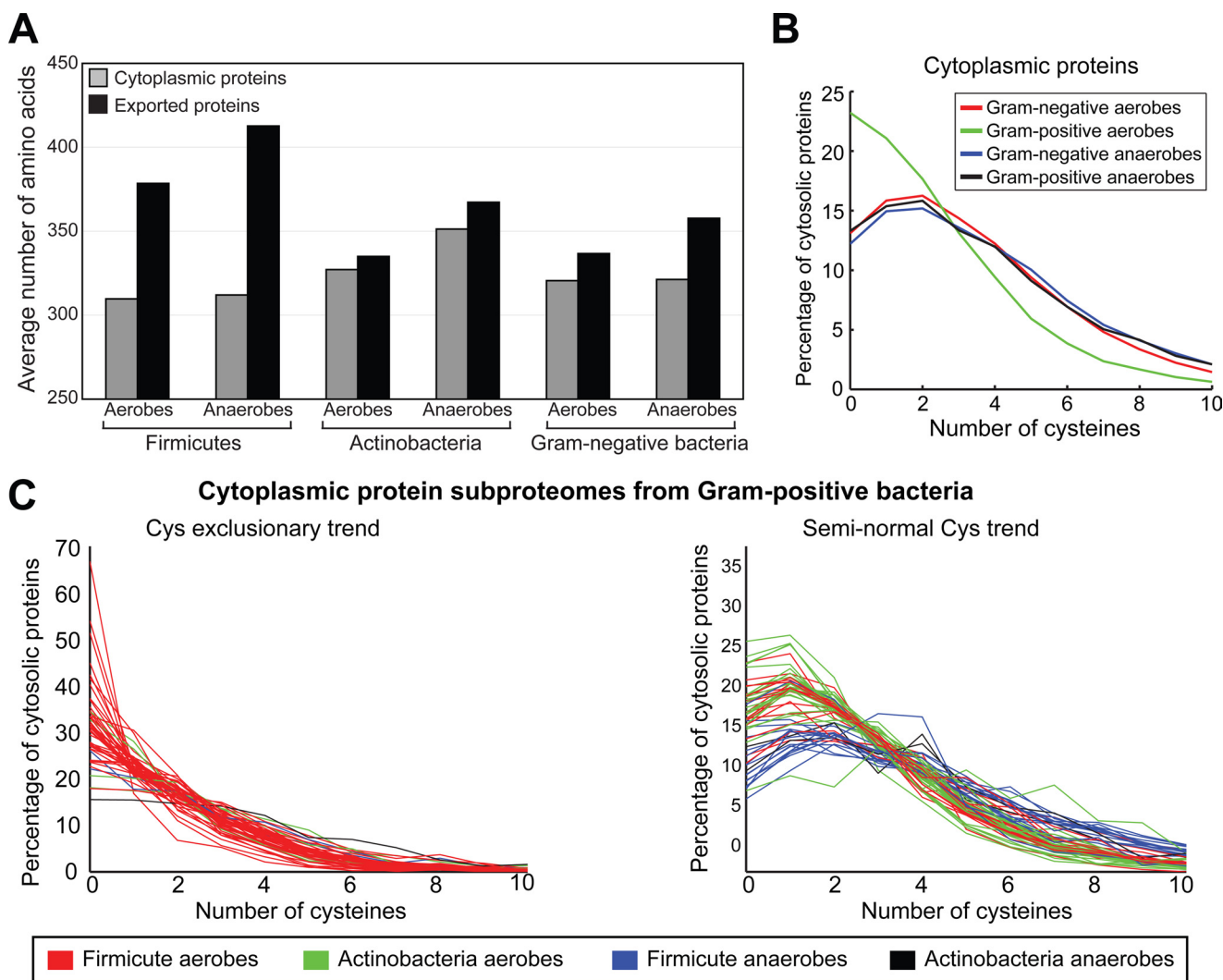


FIGURE 2. Cysteine exclusion from the exported proteins, but not from the cytoplasmic proteins, of aerobic Firmicutes correlates with a substantial increase in protein length. *A*, the average lengths of the predicted cytoplasmic and exported proteins from the indicated class of bacteria are displayed. The exported proteins from aerobic and anaerobic Firmicutes have an average length that is 22 and 32% longer than their respective cytoplasmic proteins, whereas those from aerobic and anaerobic Actinobacteria have average lengths that are only 2 and 5% longer, respectively. *B*, aerobic Gram-positive bacteria have a unique distribution in their cytoplasmic proteins with respect to cysteine content. Each line represents the percentage of cytoplasmic proteins from the different classes of bacteria that possess the indicated number of cysteine residues. *C*, the majority of aerobic Gram-positive Firmicutes show a cysteine incorporation pattern in their cytoplasmic proteins that follows the cysteine exclusion trend (*left panel*) versus the semi-normal cysteine distribution (*right panel*) favored by Actinobacteria and anaerobic Firmicutes.

tive secreted soluble Dsb-like proteins with Cys-*X-X*-Cys motifs (CG26, CG354, and CG2799) and analyzed their reductase and oxidase activities (Fig. 5).

In contrast to the DsbA homologs in Gram-negative bacteria, all three putative oxidoreductases from *C. glutamicum* exist as homodimers and not monomers (Fig. 5C). To test for reductase activity, equal molar amounts of each enzyme were examined for their ability to reduce insulin *in vitro* (37). CG26 reduced the highest amount of insulin, followed by DsbA from *E. coli* and CG2799, which showed weak activity. All of these enzymes functioned in a concentration-dependent manner, with the exception of CG354, which was inactive (Fig. 5D and supplemental Fig. S10).

Oxidation activity was determined by the ability of the enzymes to properly oxidize reduced RNase A (36). Both CG26 and CG2799, but not CG354, could productively oxidize reduced RNase A in a concentration-dependent manner (Fig.

5E and supplemental Fig. S11). Homology searches revealed that CG26 is a conserved secreted enzyme with its closest homologs in a diverse number of Actinobacteria (supplemental Fig. S12). In contrast, CG2799 has only two homologs, indicating that it likely performs a specific function for Corynebacteria (supplemental Fig. S13). Together, these data indicate that Actinobacteria also possess a functional DsbA-like homolog (A-DsbA) that potentially acts as a homodimer to oxidize their exported proteins during maturation.

DISCUSSION

Although several mechanisms are known for how Gram-negative bacteria and their proteins have adapted to the challenges posed by oxidative environments, very little is known about how Gram-positive bacteria have evolved to overcome these problems. To address this issue, we predicted the subcellular location of ~1,000,000 bacterial proteins by computa-

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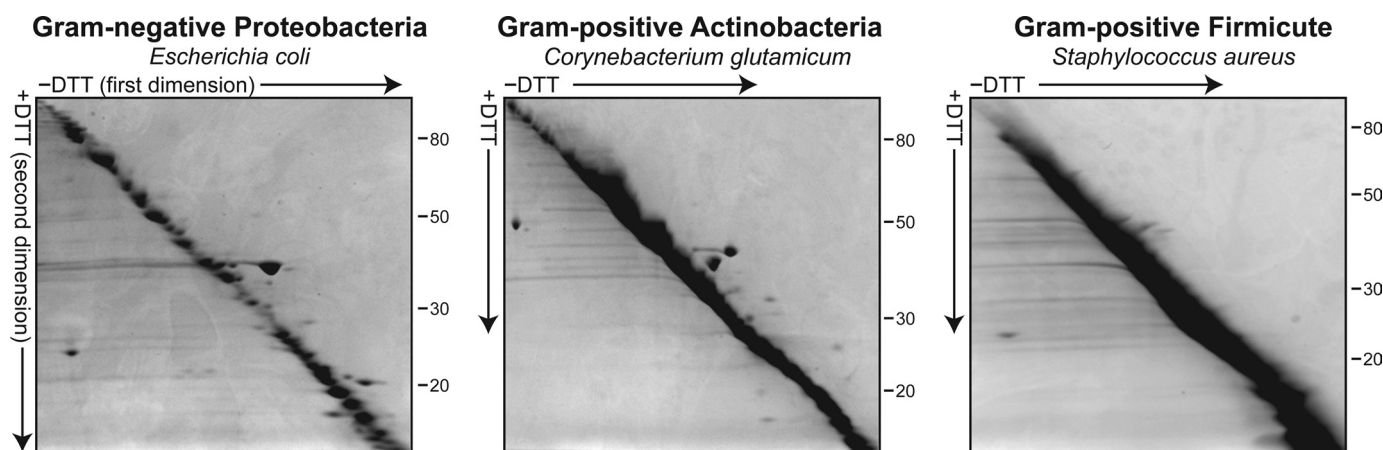


FIGURE 3. Actinobacteria possess disulfide-bonded proteins. Actinobacteria (*C. glutamicum*) and Gram-negative Proteobacteria (*E. coli*) express proteins with both inter- and intramolecular disulfide bonds. Whole cell lysates from the indicated bacteria were separated by two-dimensional nonreducing/reducing SDS-PAGE and visualized by Coomassie Blue staining. Intramolecular disulfide-bonded proteins lie above the *diagonal*, and intermolecular disulfide-bonded proteins lie below the *diagonal*.

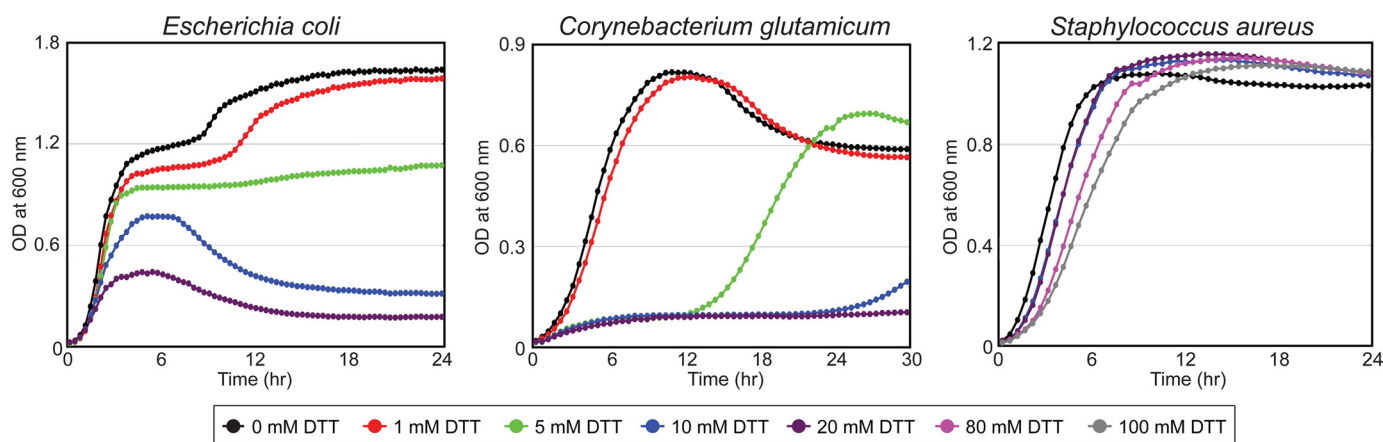


FIGURE 4. Firmicutes have a unique tolerance for high levels of reductant. The reductant DTT severely impaired the growth of aerobic Gram-negative Proteobacteria and Actinobacteria, whereas aerobic Firmicutes were capable of growth even at high DTT concentrations (for additional species, see [supplemental Fig. S9](#)). Representative growth curves from three independent experiments are displayed as changes in OD_{600 nm}. The indicated bacterial strains were grown in brain-heart infusion medium in the presence of 0, 1, 5, 10, and 20 mM DTT (*E. coli* and *C. glutamicum*) or 0, 10, 20, 80, and 100 mM DTT (*S. aureus*).

tional analysis and examined their respective cysteine incorporation patterns to identify potential strategies these bacteria have evolved in response to oxidative pressure on their proteins. We further determined the sensitivity of a range of bacterial species to the reductant DTT, looked for disulfide-bonded proteins by two-dimensional nonreducing/reducing SDS-PAGE, and identified two dimeric actinobacterial Dsb homologs capable of productive oxidation. Together, our predictive and experimental results, combined with those from previous studies (16, 27, 28), strongly suggest that aerobic Gram-positive Actinobacteria possess the capacity for regulated disulfide bond formation, whereas Firmicutes do not. Instead, aerobic Firmicutes largely exclude cysteine residues from both their exported and cytoplasmic proteins.

The complexity of the folding process is largely dependent on the environment and the amino acid composition of a protein. Oxidative environments present a challenge for proteins that contain cysteines due to their susceptibility to attack by reactive oxygen species (15, 40, 41). To alleviate this problem, Gram-negative bacteria are known to maintain their cytoplasm in a reducing state by the thioredoxin and glutaredoxin systems (42,

43). In their periplasm, they localize a soluble enzyme (DsbA) that functions together with the plasma membrane-bound DsbB to catalyze disulfide bond formation in their exported proteins, which increases their stability and protects them from deleterious oxidation (16, 17).

Two different strategies were found for how Gram-positive bacteria address the environmental pressure that accompanies the localization of their proteins. As noted previously by Dutton *et al.* (27), each maturation strategy correlates with a distinct cysteine incorporation pattern that depends on whether the proteomes originate from the Firmicute or Actinobacteria phyla. Firmicutes appear to use a simple and unique method to cope with the oxidative pressure on their exported proteins that we term “cysteine exclusion.”

Cysteine exclusion could have evolved by a selective growth advantage; however, its implementation as a protective strategy is evident based on the following findings. (i) Cysteine incorporation patterns are different between aerobic and anaerobic Firmicutes; (ii) compared with Actinobacteria and Gram-negative bacteria, Firmicutes have a high tolerance for reductant in the growth medium; and (iii) conserved cysteines that form disul-

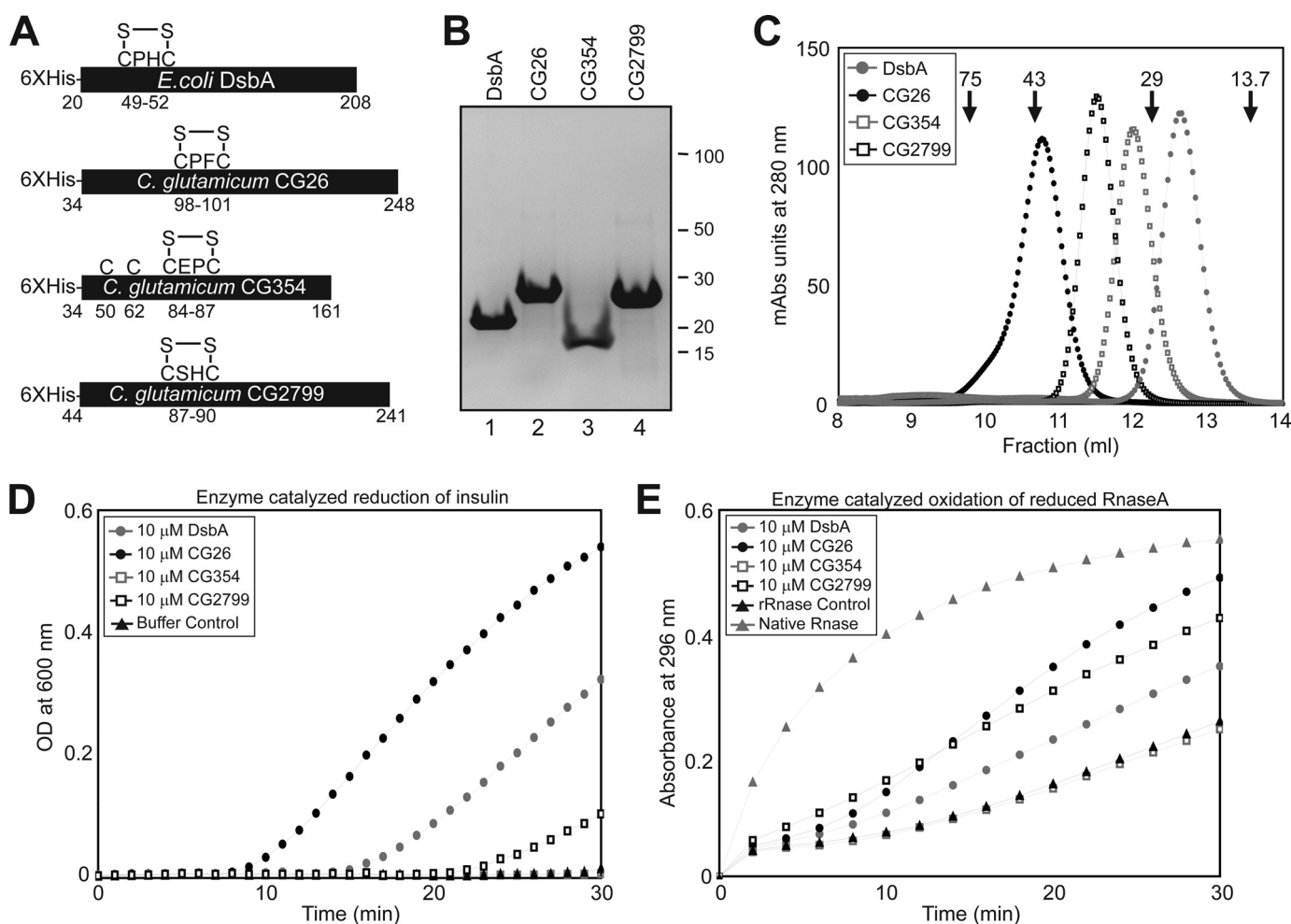


FIGURE 5. The Actinobacterium *C. glutamicum* possesses two secreted dimeric oxidoreductases. *A*, schematic representations and characteristics of the three putative oxidoreductases from *C. glutamicum* and DsbA from *E. coli*. The recombinant proteins were expressed with an N-terminal His tag replacing the signal peptide. The amino acid numbering refers to the unprocessed wild-type proteins, and the active-site Cys-X-X-Cys motifs are displayed. *B*, Coomassie Blue-stained gel of the recombinant proteins that were purified following expression in *E. coli*. *C*, size exclusion profiles of the purified proteins separated on a Superdex 75 column and monitored by the absorbance at 280 nm. The relative size of each standard is indicated with an arrow. mAbs, milli-absorbance. *D*, the actinobacterial enzyme CG26 from *C. glutamicum* efficiently reduces insulin *in vitro*, whereas CG2799 shows slight activity. Each enzyme was analyzed for the capacity to reduce insulin at 27 °C by monitoring precipitation of the reduced B chain observed by changes in OD_{600 nm}. *E*, CG26 and CG2799 can correctly oxidize reduced RNase A *in vitro*. The ability of each enzyme to properly oxidize reduced RNase A (rRNase) in a glutathione redox buffer was assayed at 27 °C by monitoring the change in the absorbance at 296 nm that results from cleavage of cCMP by RNase A. The activities of native RNase A and reduced RNase A were included as controls.

fide bonds in class A β -lactamases and alkaline phosphatases from Gram-negative bacteria are not present in many of the aerobic Firmicute homologs (27, 44–47). Thus, the exported proteins in aerobic Firmicutes that contain cysteines, such as lipoproteins and the sortase enzymes involved in pilus biogenesis, do so as a functional requirement; otherwise, they would have been excluded.

Next, we characterized two disulfide bond-forming proteins in *C. glutamicum* and found that one (CG26) is conserved within the phylum of Actinobacteria. Like the oxidoreductase in the Actinobacterium *Mycobacterium tuberculosis*, which was identified by DsbE homology (48), these enzymes are not entirely homologous to the classical DsbA from Gram-negative bacteria. The differences in sequence (~15% identity) and oligomeric state from the *E. coli* DsbA suggest that, as a group, these enzymes should be classified as actinobacterial or A-DsbA. Together, these findings indicate that, although homology searches are useful to identify potential disulfide

bond-forming enzymes (16, 27, 28), experimental validation is essential to confirm that these proteins function as predicted (41).

The fact that A-DsbA enzymes exist as a dimer and not a monomer like the *E. coli* DsbA has several functional implications. The dimerization of A-DsbA potentially creates specificity for its membrane-bound partner, and it also places two Cys-X-X-Cys active sites in close proximity, which mimics the eukaryotic protein-disulfide isomerases that reside in the endoplasmic reticulum (49). The major difference is that protein-disulfide isomerases are monomeric proteins with two Cys-X-X-Cys active sites. It has recently been proposed that homologs of the vitamin K epoxide reductase enzyme recharge actinobacterial DsbA at the plasma membrane (27, 50). The vitamin K epoxide reductase enzyme that has been proposed to recycle A-DsbA also has potential analogy to eukaryotic oxidation, as homologs exist in the endoplasmic reticulum (27, 50). Thus, disulfide bond formation in Actinobacteria may be more evo-

Cysteine Exclusion and Oxidation in Gram-positive Bacteria

lutionarily related to eukaryotes than to Gram-negative bacteria, suggesting that eukaryotic vitamin K epoxide reductase homologs may be responsible for recharging protein-disulfide isomerase.

One intriguing concept that arose from this study is that Firmicutes also favor cysteine exclusion in their cytoplasmic proteins. This suggests that aerobic Firmicutes may have a high level of oxidative pressure on their cytoplasmic proteins as well. A possible explanation for this is that the outer membrane, which is absent in Firmicutes, provides an additional permeability barrier that aids in the regulation of the cytoplasmic redox conditions.

These findings indicate that aerobic Actinobacteria likely possess a regulated compartment that contains the machinery for disulfide bond formation (27, 48). Supporting this, cryo-electron tomography studies have shown that the mycolic acids covalently linked to the peptidoglycan of Actinobacteria create an outer membrane-like structure that is absent in Firmicutes (51, 52). Thus, the presence of an outer membrane appears to be a strong driving force for evolving a system that facilitates disulfide bond formation by creating a regulated folding environment that co-localizes the enzymes and substrates.

Firmicutes such as *B. subtilis*, *S. aureus*, *E. faecalis*, and *Listeria monocytogenes* have previously been reported to encode homologs of DsbA from Gram-negative bacteria (16, 27, 28). Of these, *B. subtilis* is the only Firmicute that also encodes DsbB and for which substrates have been identified, but neither of these two substrates are involved in essential processes (53–57). In the case of *S. aureus*, where no disulfide-bonded substrates have been identified, the lipoprotein homolog of DsbA is thought to function independently by utilizing extracellular oxidants for its recycling in the absence of DsbB (28, 58, 59). Our analysis, combined with the existence of only two validated substrates that possess enzyme-catalyzed disulfide bonds, accentuates that Firmicutes generally exclude cysteines from their exported proteins. Thus, the few Firmicutes that contain DsbA and DsbB homologs likely utilize them for disulfide bond formation in proteins with nonessential niche functions.

From an evolutionary perspective, cysteine exclusion likely provides a selective advantage in extreme redox environments, potentially explaining why Firmicutes are over-represented among the bacteria that colonize and infect oxygen-exposed tissues such as the epidermis and the respiratory tract (60). This is supported by our finding that Firmicutes tolerate strong reductants and by the fact that Firmicute pathogens such as *Streptococcus pneumoniae* actively produce high levels of the powerful oxidant hydrogen peroxide (61). Moreover, the simple strategy of cysteine exclusion may provide Firmicutes with some protection against the oxidation-based immune defenses employed at the host-pathogen interface by neutrophils and macrophages. On the negative side, the inability of Firmicutes to cope with cysteine residues could create a barrier for lateral gene transfer, reducing the variability of their exported proteins, which constitute the majority of bacterial virulence factors. However, recent studies have demonstrated that Firmicutes can also maintain the structural stability of their exported proteins by utilizing alternative covalent bonds, which likely contributes to their ability to exclude cysteines (62). Although it

is clear that Firmicutes in general do not possess the capacity for disulfide bond formation, further investigation into whether they protect their few exported proteins with single cysteines by a reductase system that is functionally analogous to the one recently described in *E. coli* is warranted (19).

Acknowledgments—We thank members of the Normark laboratory for helpful comments and critical reading of the manuscript.

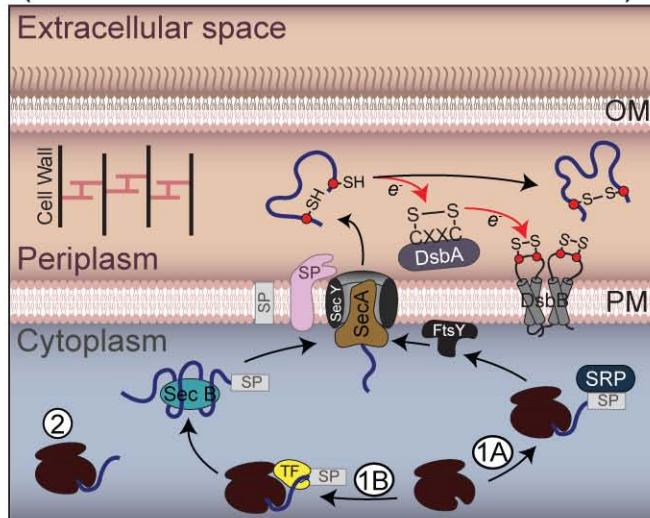
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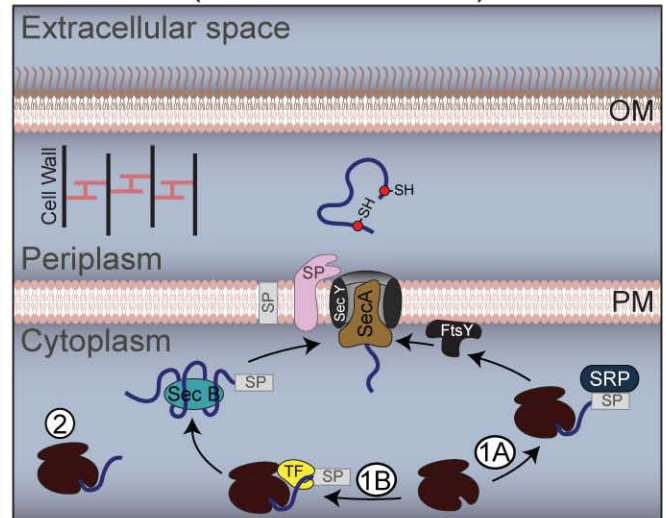
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Gram-negative bacteria

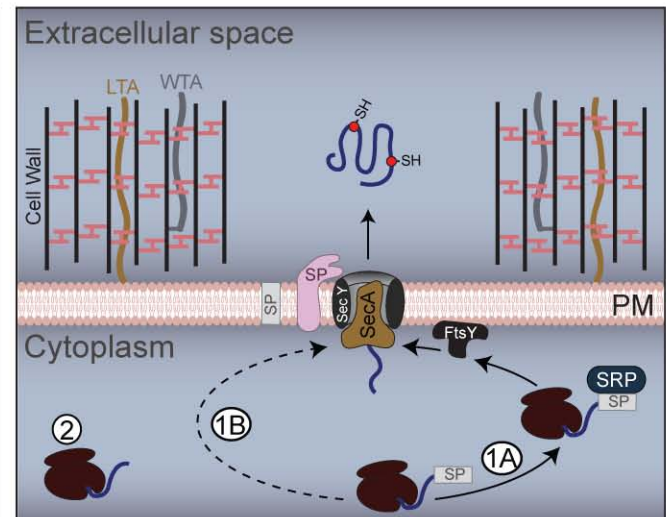
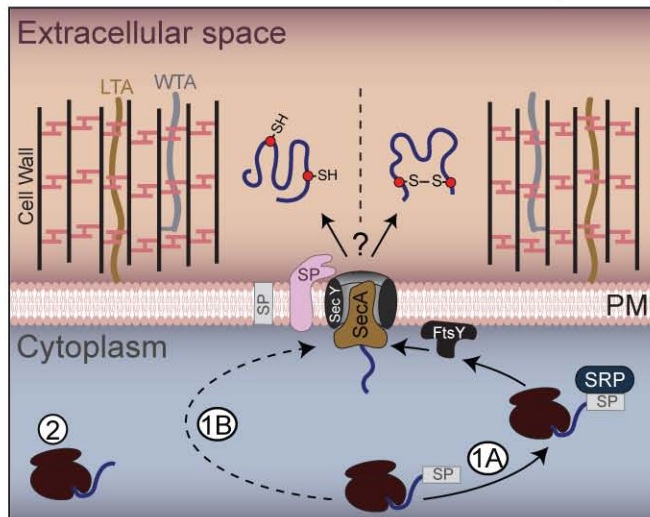
Aerobes
(Aerobes and Facultative Anaerobes)



Anaerobes
(Strict Anaerobes)



Gram-positive bacteria



■ Reducing ■ Oxidizing

Figure S1. Illustrations of the oxidizing and reducing conditions that the cytoplasmic and Sec-dependent exported proteins encounter in Gram-positive and Gram-negative aerobic bacteria (aerobes and facultative anaerobes) and anaerobic bacteria (strict anaerobes). The cytosolic (2) and the two Sec-dependent translocation pathways are depicted: co-translational targeting by SRP (1A) and SecB-dependent post-translocation targeting (1B), which has yet to be identified in Gram-positive bacteria.

Method for the prediction of protein topology

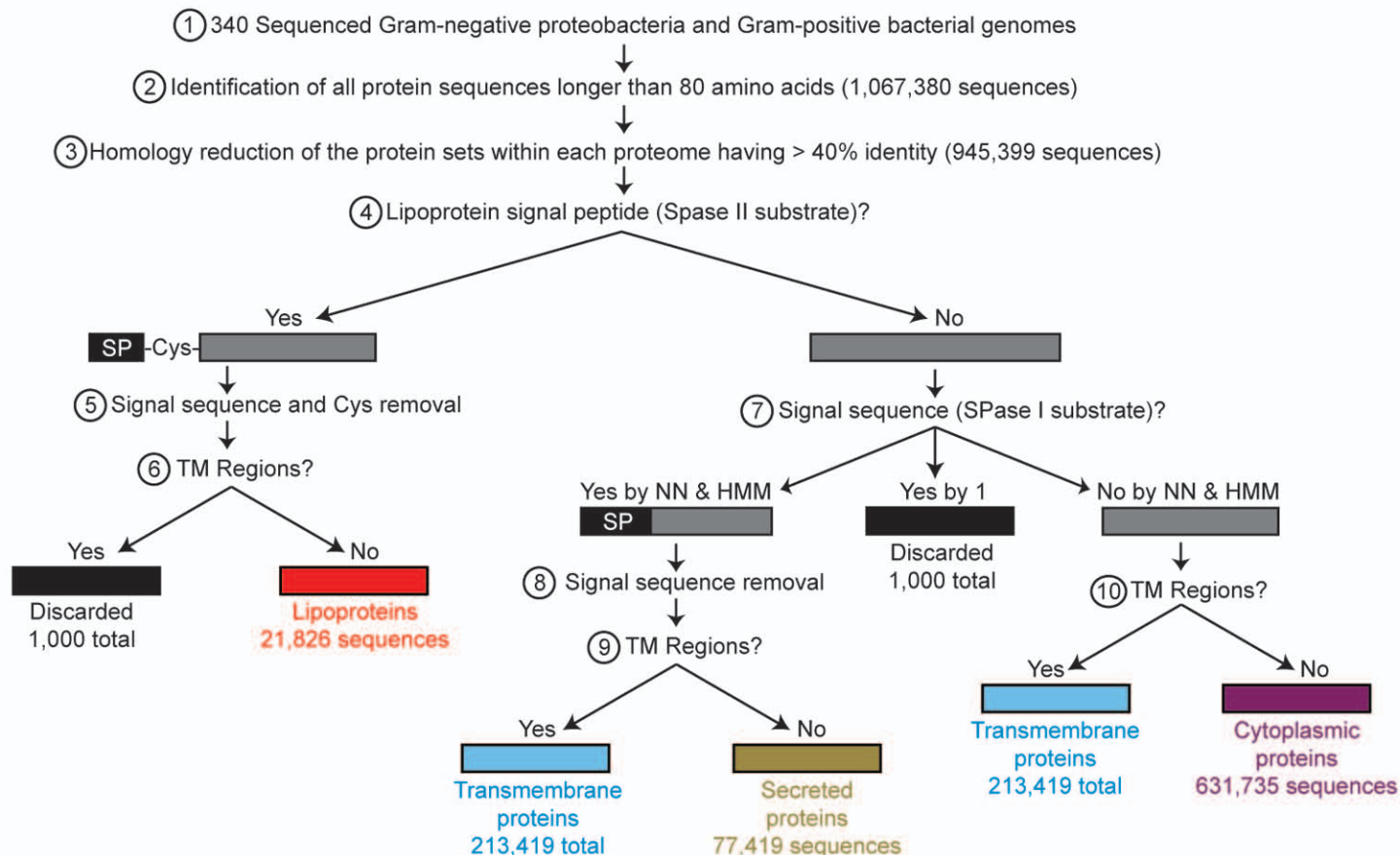


Figure S2. Method outline for the prediction of protein topology. (1) Bacterial proteomes were obtained from EBI (<http://www.ebc.ac.uk/>). (2) Proteins were identified by ORFs that encode proteins 80 amino acids or longer. (3) Protein sets were homology reduced within each genome using a cut-off of 40% identity. (4), LipoP 1.0 was used to predict all potential lipoproteins based on the presence of a SPase II cleavable signal peptide (SP) (Juncker *et al.* 2003). (5 and 6) The lipoprotein signal peptides and N-terminal cysteine residues were removed and all identified lipoproteins underwent a secondary screen to exclude those that contained potential transmembrane (TM) regions using SCAMPI (Bernsel *et al.* 2008). (7) Proteins lacking a lipoprotein signal sequence were analyzed for the presence of a SPase I cleavable signal peptide using SignalP 3.0 (Bendtsen *et al.* 2004). Positive predictions by both the Neural Network and Hidden Markov Model were required to determine the presence of a SPase I signal peptide. Proteins predicted by only one model were discarded. (8 and 9) Signal peptides were removed and the proteins were characterized as secreted proteins or TM proteins based on the absence or presence of potential TM regions identified by SCAMPI. (10) Proteins lacking both a SPase I and SPase II cleavable signal peptide were characterized as cytoplasmic proteins or TM proteins based on the absence or presence of potential TM regions identified by SCAMPI.

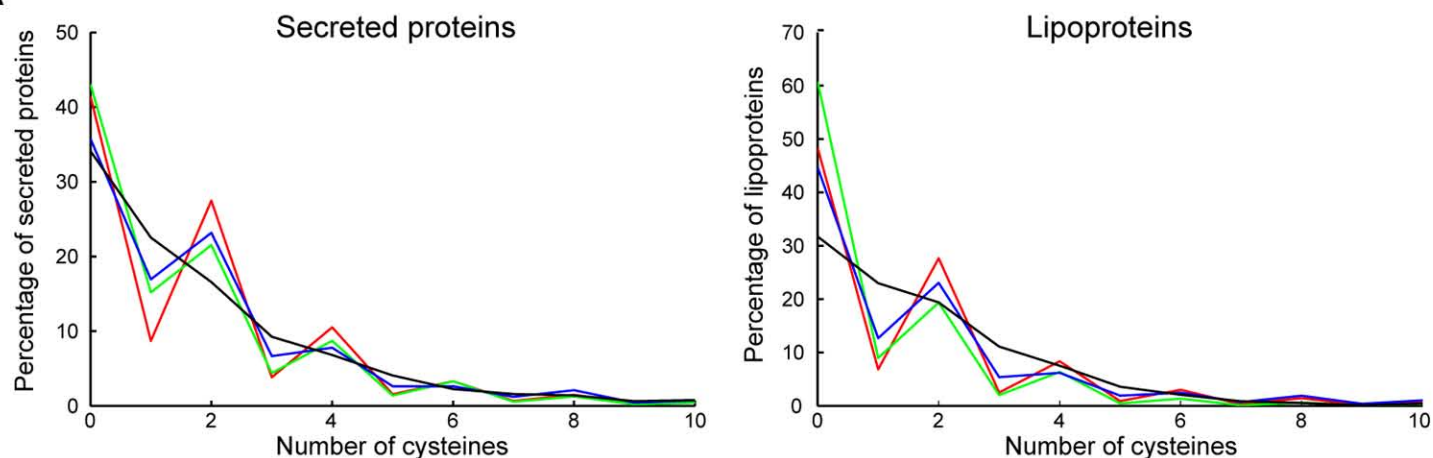
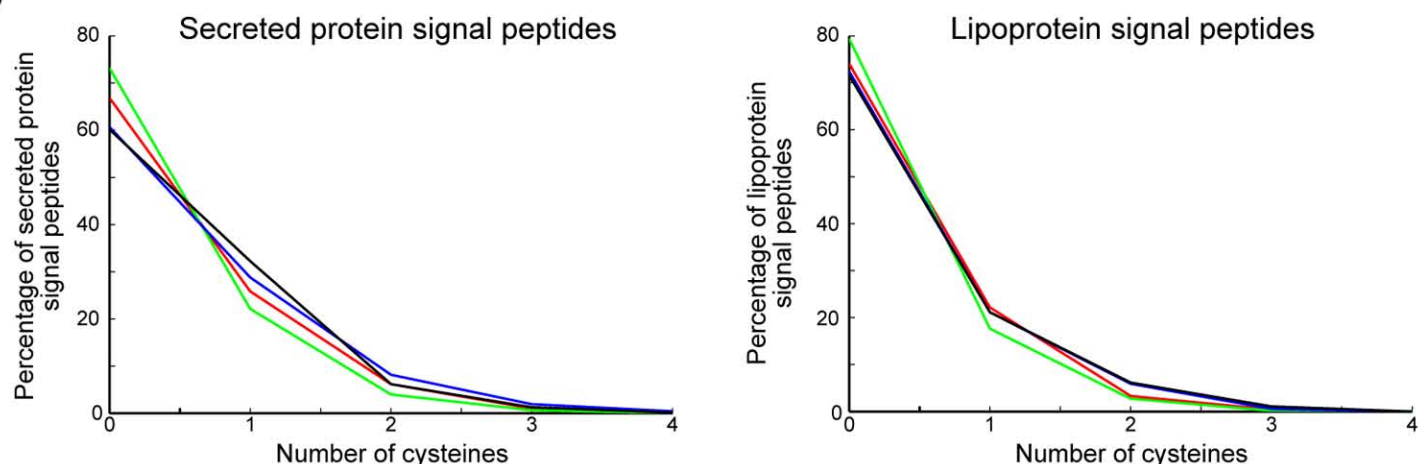
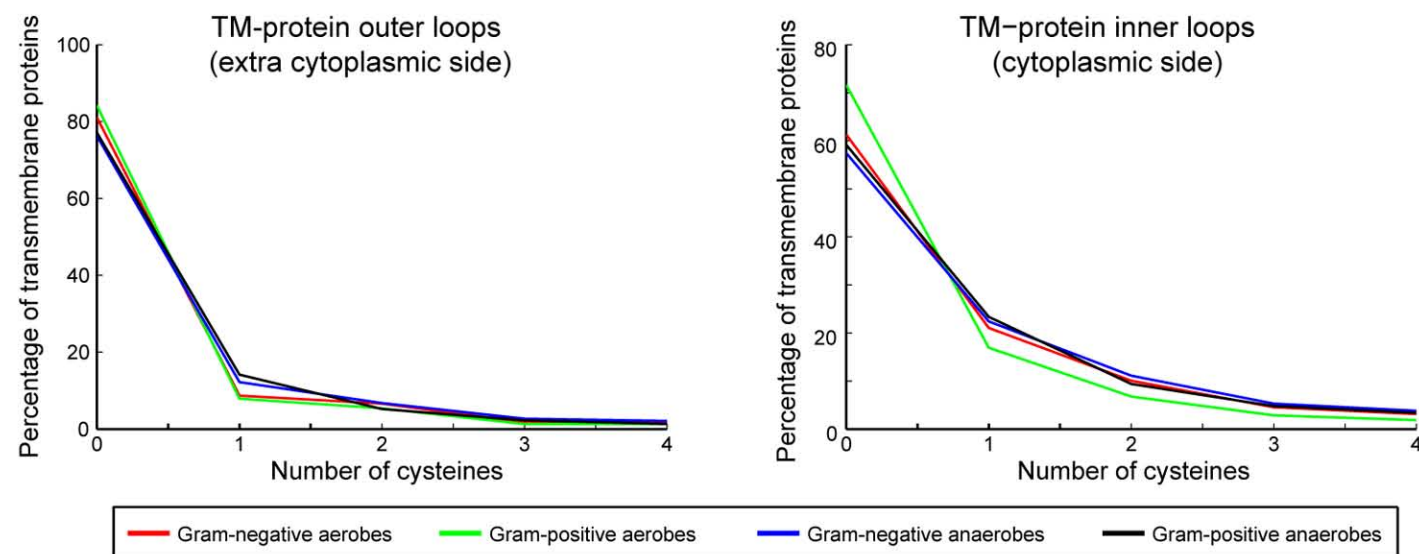
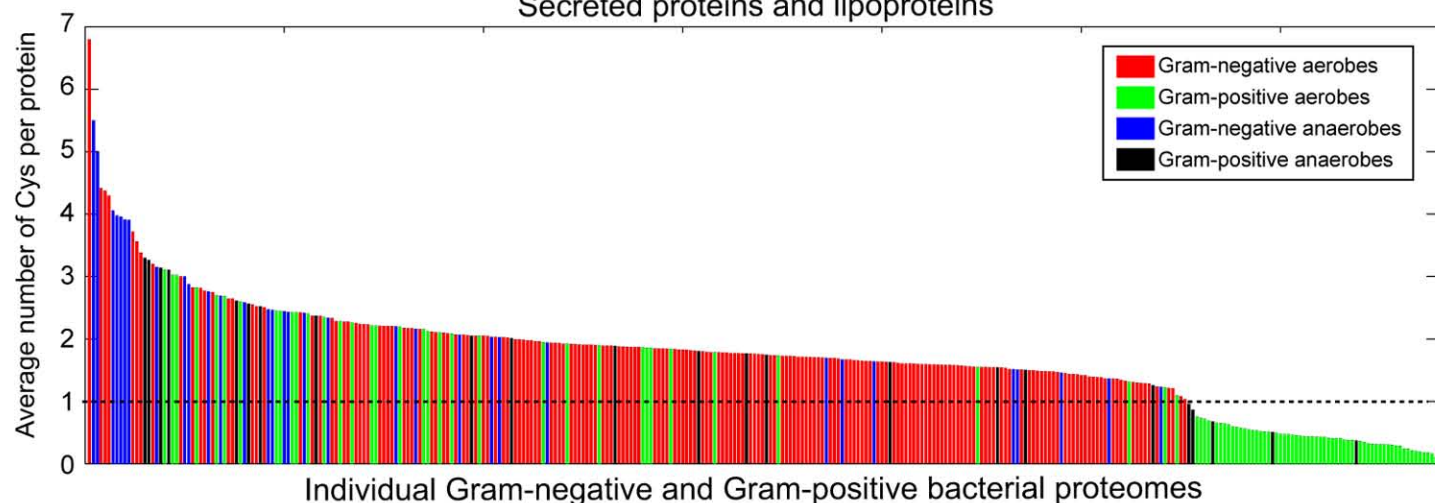
A**B****C**

Figure S3. Average cysteine distribution profiles in the lipoproteins, secreted proteins, their signal peptides and the transmembrane protein loops from aerobic and anaerobic Gram-negative Proteobacteria and Gram-positive bacteria. (A) Gram-negative Proteobacteria and aerobic Gram-positive bacteria encode cysteine residues in an even biased pattern in their secreted proteins and lipoproteins. Graphs displaying the percentage of the bacterial secreted proteins (left panel) and lipoproteins (right panel) that contain the indicated number of cysteine residues. The N-terminal cysteines on the mature lipoproteins were excluded from the calculations. (B) The cysteine frequency in the SPase I (secreted proteins) and SPase II (lipoprotein) signal peptides are shown. (C) The percentage of transmembrane (TM) proteins containing the indicated number of cysteine residues in their outer loops (left panel) and inner loops (right panel) are shown. The inner and outer loops of each TM protein were assigned using SCAMPI.

A

Secreted proteins and lipoproteins

**B**

Exported proteins (secreted proteins and lipoproteins)

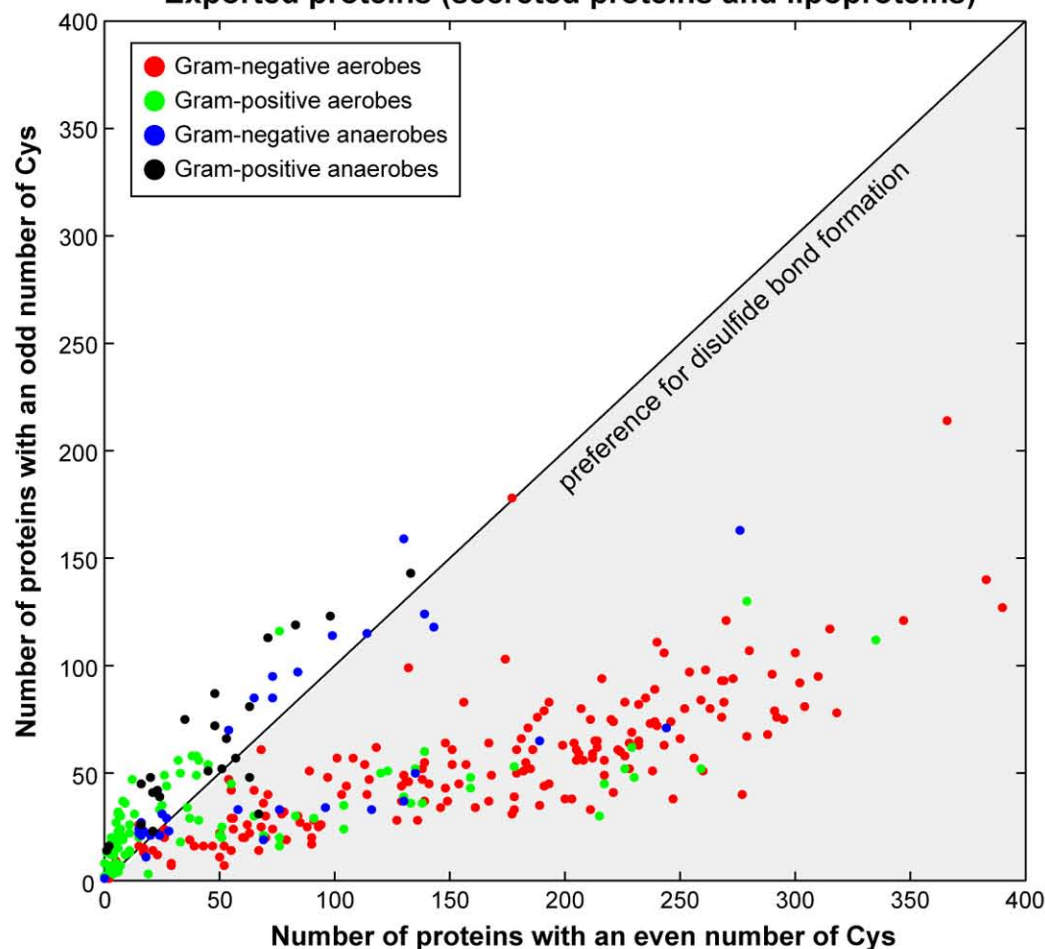


Figure S4. Gram-positive aerobes encode cysteines at a low frequency in their exported proteins and show two different incorporation trends. (A) A distinct subset of aerobic Gram-positive bacteria possess very low cysteine incorporation frequencies (average < 1 Cys per protein) in their secreted proteins and lipoproteins (exported proteins). Proteomes from the aerobic and anaerobic Gram-positive and Gram-negative bacteria were analyzed based on the average number of cysteines in their exported proteins. (B) Individual bacterial proteomes were analyzed for their frequency of encoding an odd versus even number of cysteines within their exported proteins (proteins with 0 Cys were excluded). One cluster of aerobic Gram-positive bacteria is distributed with the aerobic Gram-negative proteobacteria in the lower half of the quadrant (grey) where the majority of their exported proteins contain an even number of cysteines. The other cluster of aerobic Gram-positive bacteria is found in the upper quadrant with a low number of exported proteins encoding cysteine residues in a non-even biased manner.

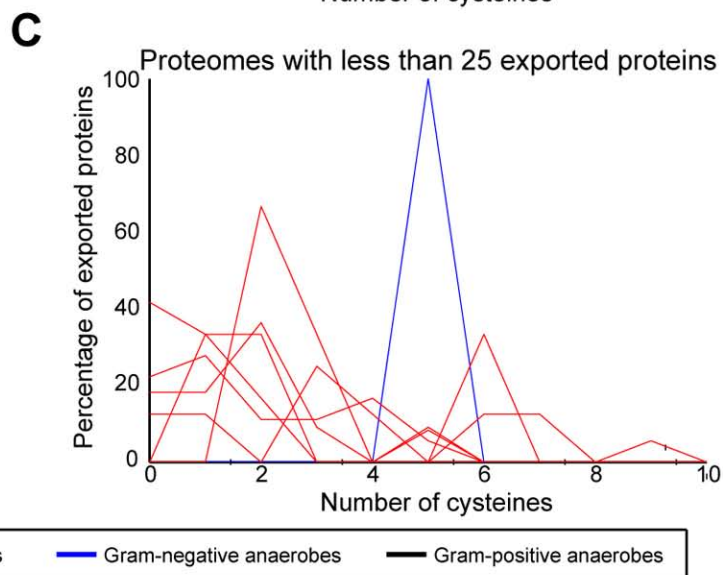
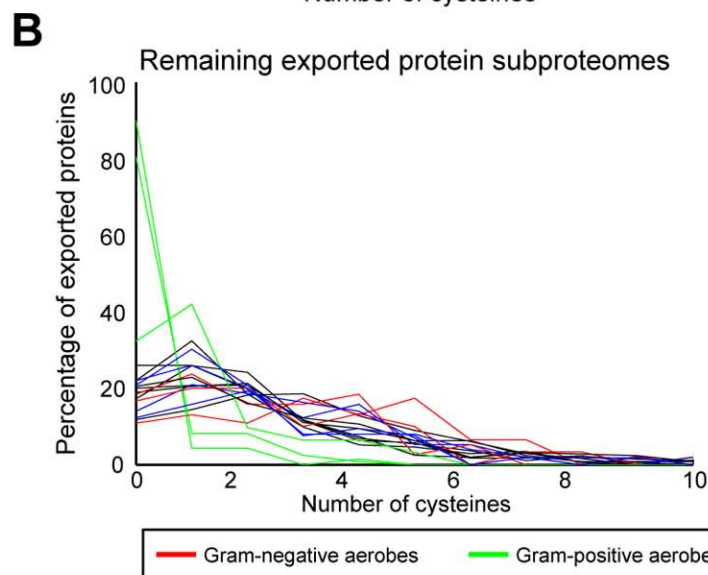
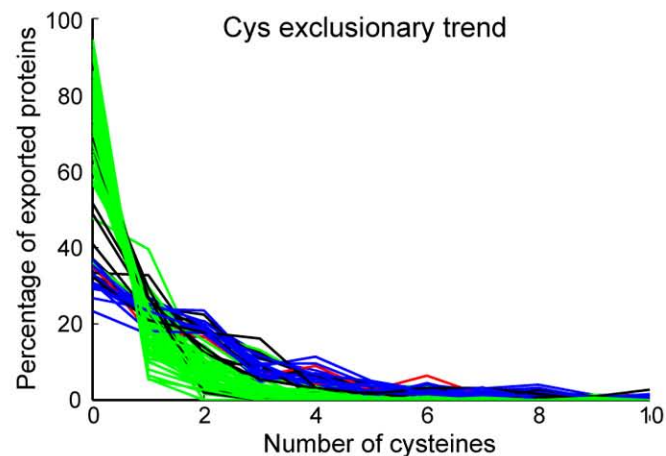
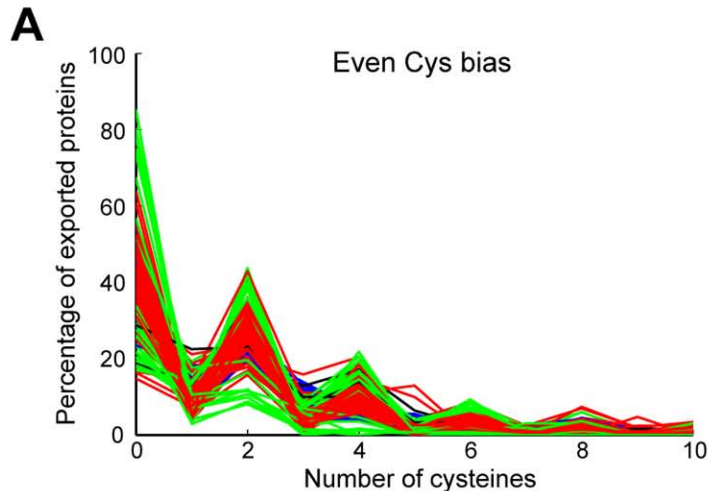


Figure S5. Gram-negative and Gram-positive bacteria have evolved either to exclude cysteine residues from their secreted proteins and lipoproteins or encode them in pairs. (A) The large majority of bacterial proteomes can be separated based on their exported proteins showing a preference for encoding cysteine residues in an even-numbered bias (left panel), or in an exclusionary trend (right panel). The even-numbered bias is defined as the percentage of proteins containing 0 Cys > 1 Cys and 2 Cys > 1 Cys and the exclusionary trend is defined as 0 Cys > 1 Cys > 2 Cys. The cysteines within the SPase I and II signal peptides were analyzed separately (Figure S3), and the N-terminal cysteines on the mature lipoproteins were excluded from the calculations. (B) The few bacterial exported protein subproteomes that did not fit either of the even bias or exclusionary trend are displayed as 'remaining subproteomes', and (C) those encoding less than 25 exported proteins were excluded due to statistically irrelevant data.

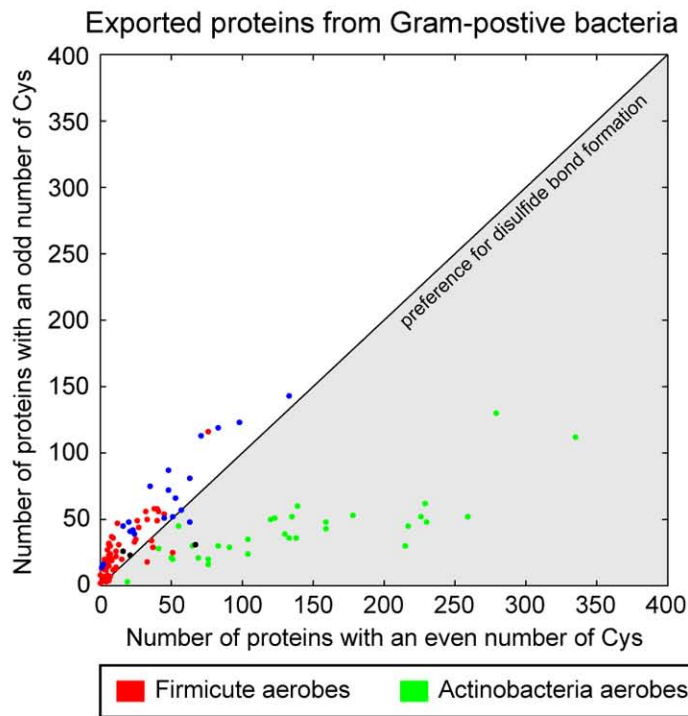
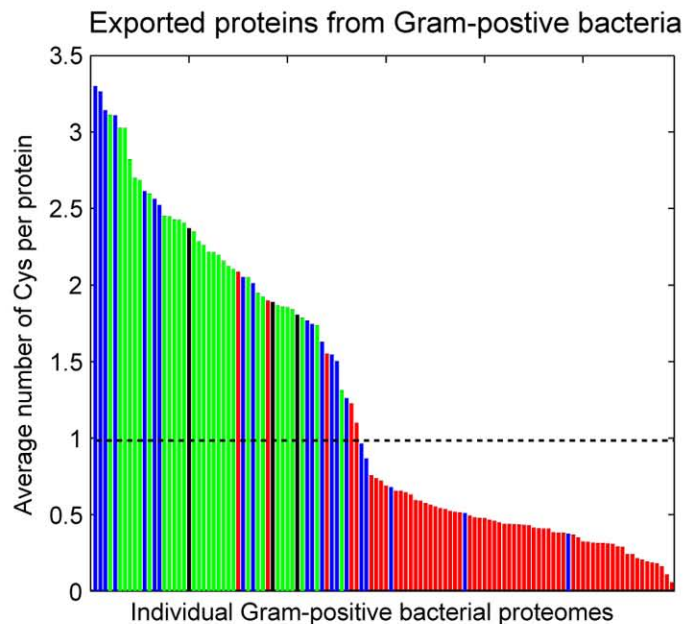
A**B**

Figure S6. The exported protein cysteine incorporation profiles in Gram-positive bacteria have an even bias in Actinobacteria and an exclusionary bias in Firmicutes. (A) All aerobic Actinobacteria favor even numbers of cysteines in their exported proteins. Each dot represents the number of exported cysteine-containing proteins with an even (grey quadrant) versus odd number of cysteines within a given bacterial proteome. (B) Actinobacteria average more than 1 Cys in their exported proteins while 92% of aerobic Firmicutes average less than 1 Cys.

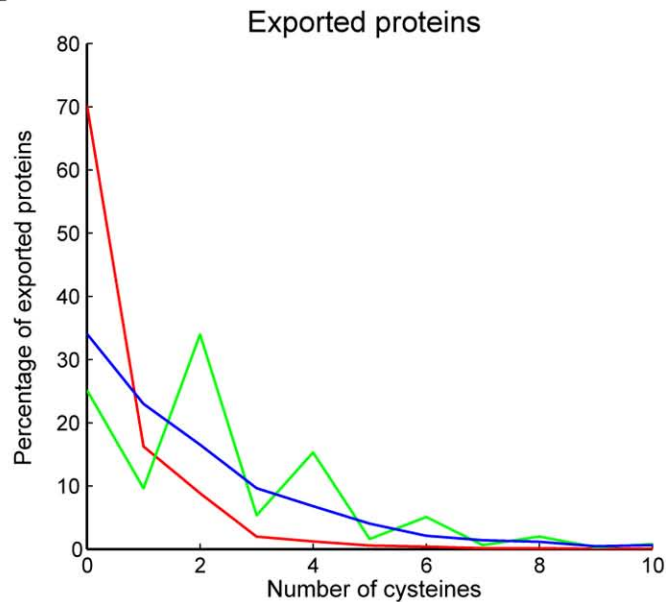
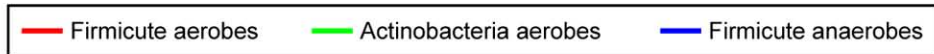
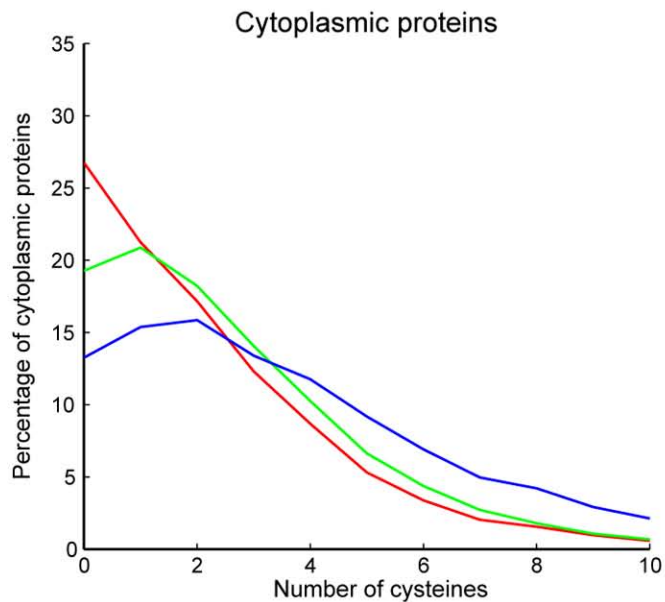
A**B**

Figure S7. Firmicutes favor cysteine exclusion, whereas Actinobacteria have a strong even-number cysteine bias in their exported proteins. (A) The percentages of exported, and (B) cytoplasmic proteins from aerobic and anaerobic Firmicutes and aerobic Actinobacteria that contain the indicated number of cysteine residues are displayed. Anaerobic Actinobacteria were excluded due to the small number (3) of proteomes.

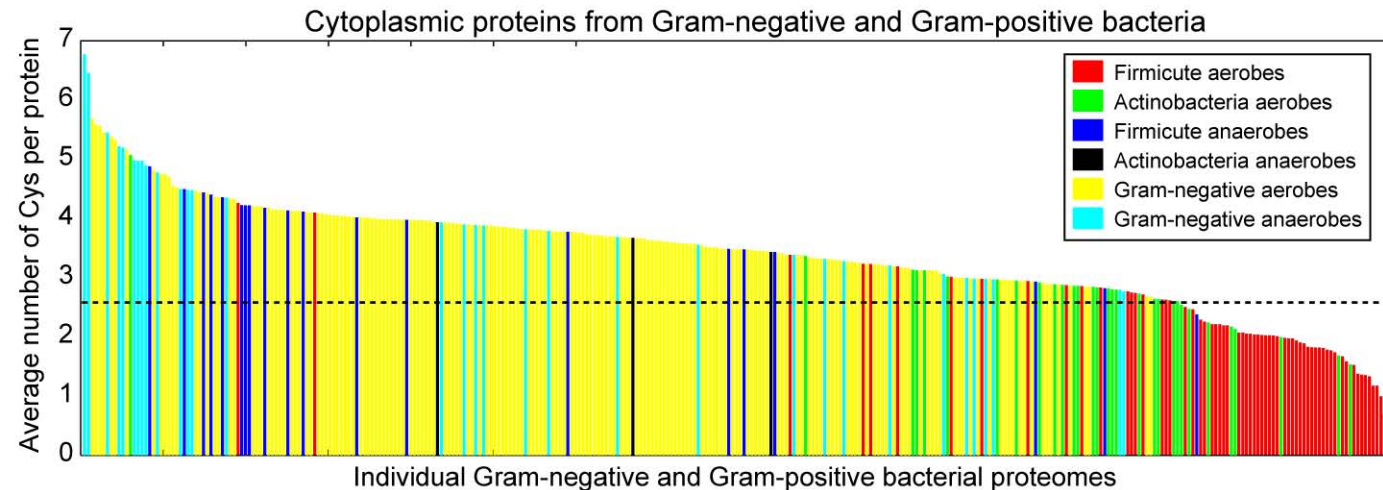


Figure S8. Cytoplasmic proteins from aerobic Gram-positive Firmicutes encode cysteines at a low frequency. Compared to Actinobacteria and Gram-negative bacteria, the cytoplasmic proteins from aerobic Gram-positive Firmicutes contain on average the fewest numbers of cysteine residues.

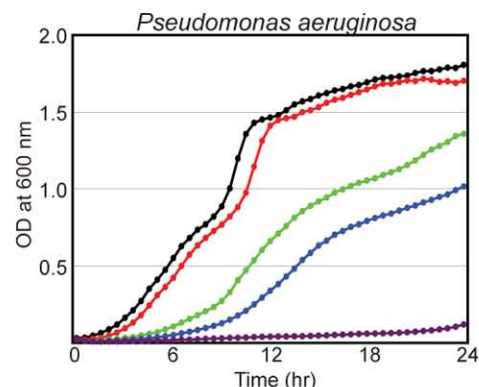
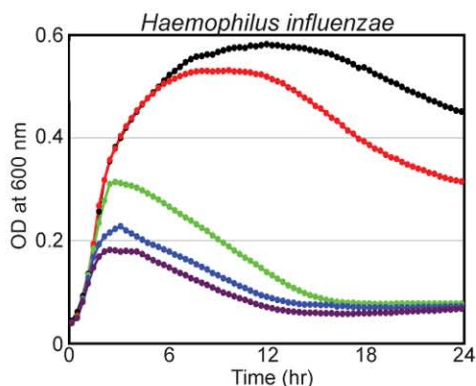
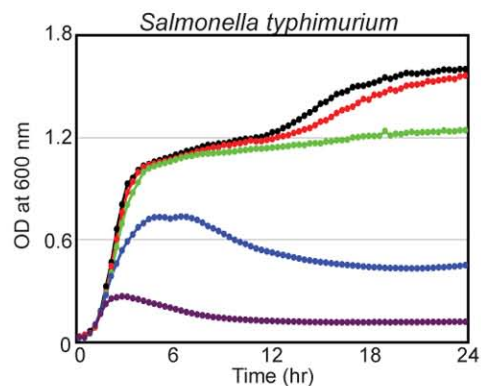
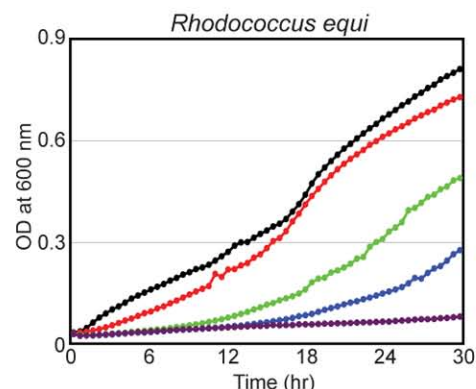
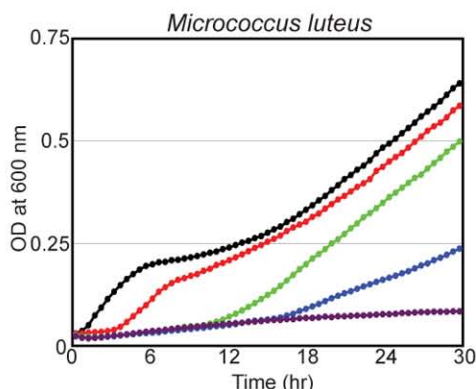
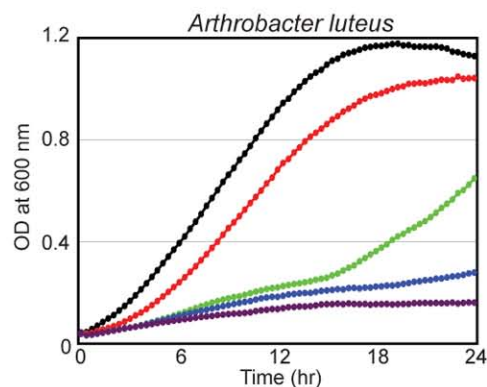
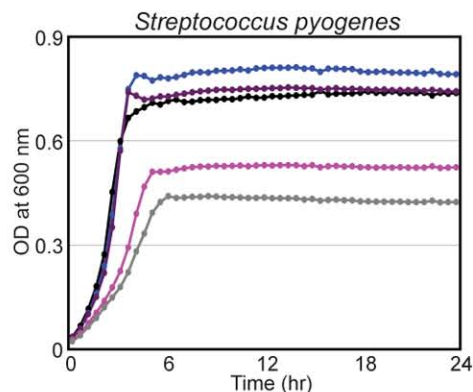
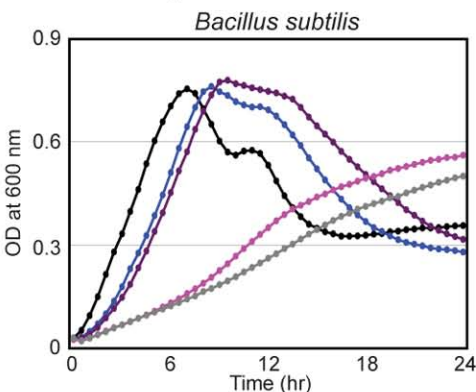
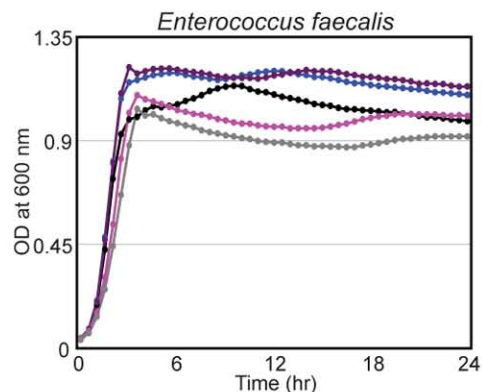
A**Gram-negative Proteobacteria****B****Gram-positive Actinobacteria****C****Gram-positive Firmicutes**

Figure S9. Firmicutes do not possess disulfide-bonded proteins essential for growth like Actinobacteria and Gram-negative Proteobacteria. (A) Additional strains of Gram-negative aerobic Proteobacteria, (B) Gram-positive aerobic Actinobacteria, and (C) aerobic Firmicutes demonstrating how reductant (DTT) severely impairs the growth of Proteobacteria and Actinobacteria, but not Firmicutes. Representative growth curves of 3 independent experiments are displayed as changes in the OD at 600 nm. The indicated bacterial strains were grown at 37°C in BHI in the presence of 0, 1, 5, 10 and 20 mM DTT (A and B) or 0, 10, 20, 80 and 100 mM DTT (C).

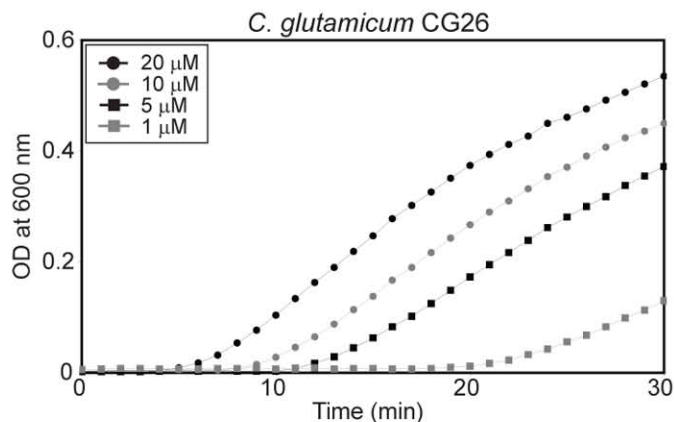
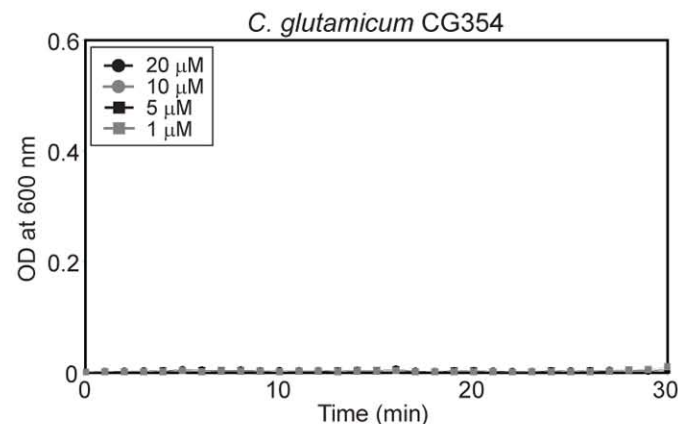
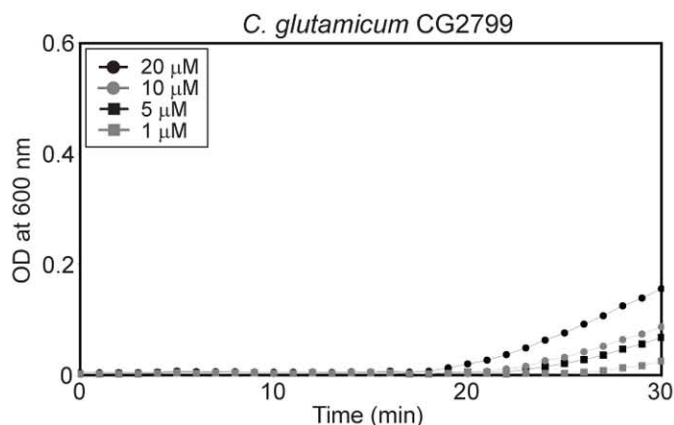
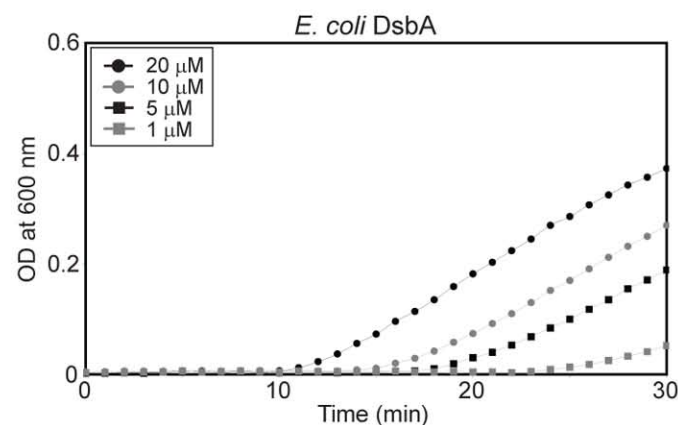
A**B****C****D**

Figure S10. The putative oxidoreductases, CG26 and CG2799, from *C. glutamicum* have reductase activity. (A) The *in vitro* reductase activity of the three purified CG26, (B), CG354, and (C) CG2799 are shown with the control (D) DsbA from *E. coli*. The reductase activity was monitored by measuring the turbidity of insulin that occurs upon precipitation of the reduced B chain. The indicated concentration of each enzyme was assayed at 27°C in a 200 μ l solution containing 100 mM KOAc pH = 7.5, 2 mM EDTA, 320 μ M DTT and 200 μ g of insulin and the OD at 600 nm was followed for 30 min.

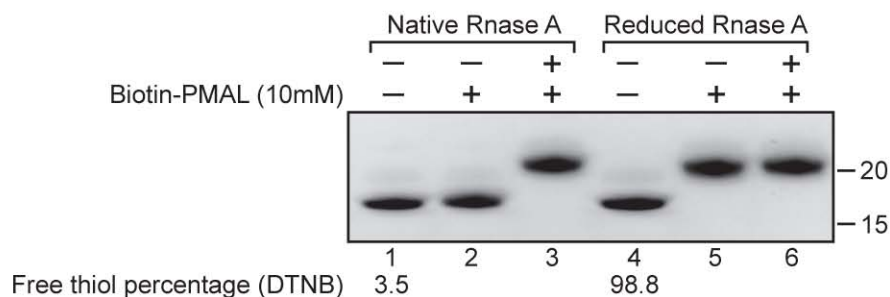
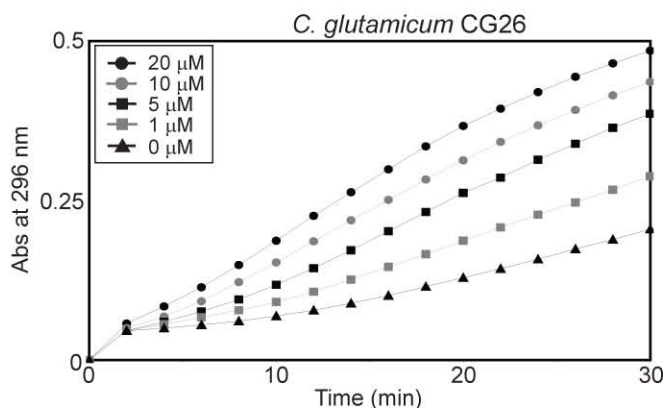
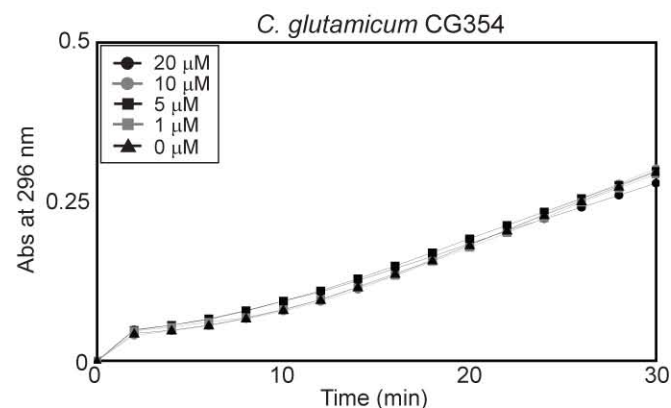
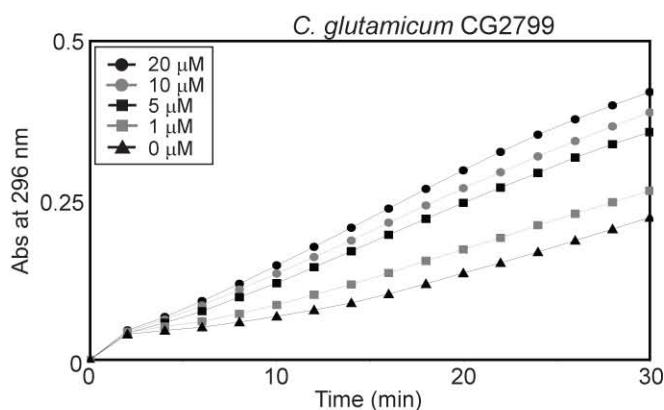
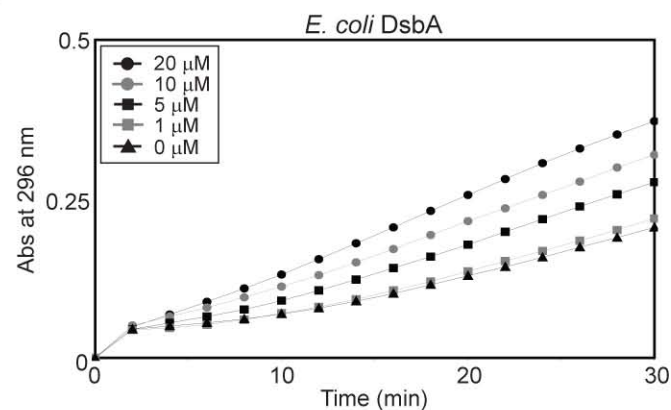
A**B****C****D****E**

Figure S11. CG26 and CG2799 from *C. glutamicum* correctly oxidize reduced Rnase A *in vitro*. (A) Native RnaseA and reduced RnaseA were incubated with the free thiol-modifying agent Maleimide-PEO₂-Biotin (PMAL) for 1 hr at 4°C prior to (lanes 2 and 5) and after reduction with DTT for 1 hr at 27°C (lanes 3 and 6). Samples were resolved by SDS-PAGE followed by coomassie staining. The free thiol percentage was also calculated for samples 1 and 4 using 5,5'-Dithiobis(2nitrobenzoic acid) (DTNB). (B) The *in vitro* oxidase activity of the purified CG26, (C), CG354 and (D), CG2799 are shown in comparison to (E) DsbA from *E. coli*. The oxidase activity was monitored by measuring the cleavage of cCMP by properly oxidized RnaseA. The indicated concentration of each enzyme was assayed at 27°C in a solution containing 100 mM Tris-Acetate pH = 8.0, 2 mM EDTA, 0.2 mM GSSG pH = 8.0, 1 mM GSH pH = 8.0 and 10 μM of reduced RnaseA in a final volume of 1 ml. The reaction was equilibrated for 2 min, adjusted to 4.5 mM cCMP and the Absorbance (Abs) at 296 nm was followed for 30 min.

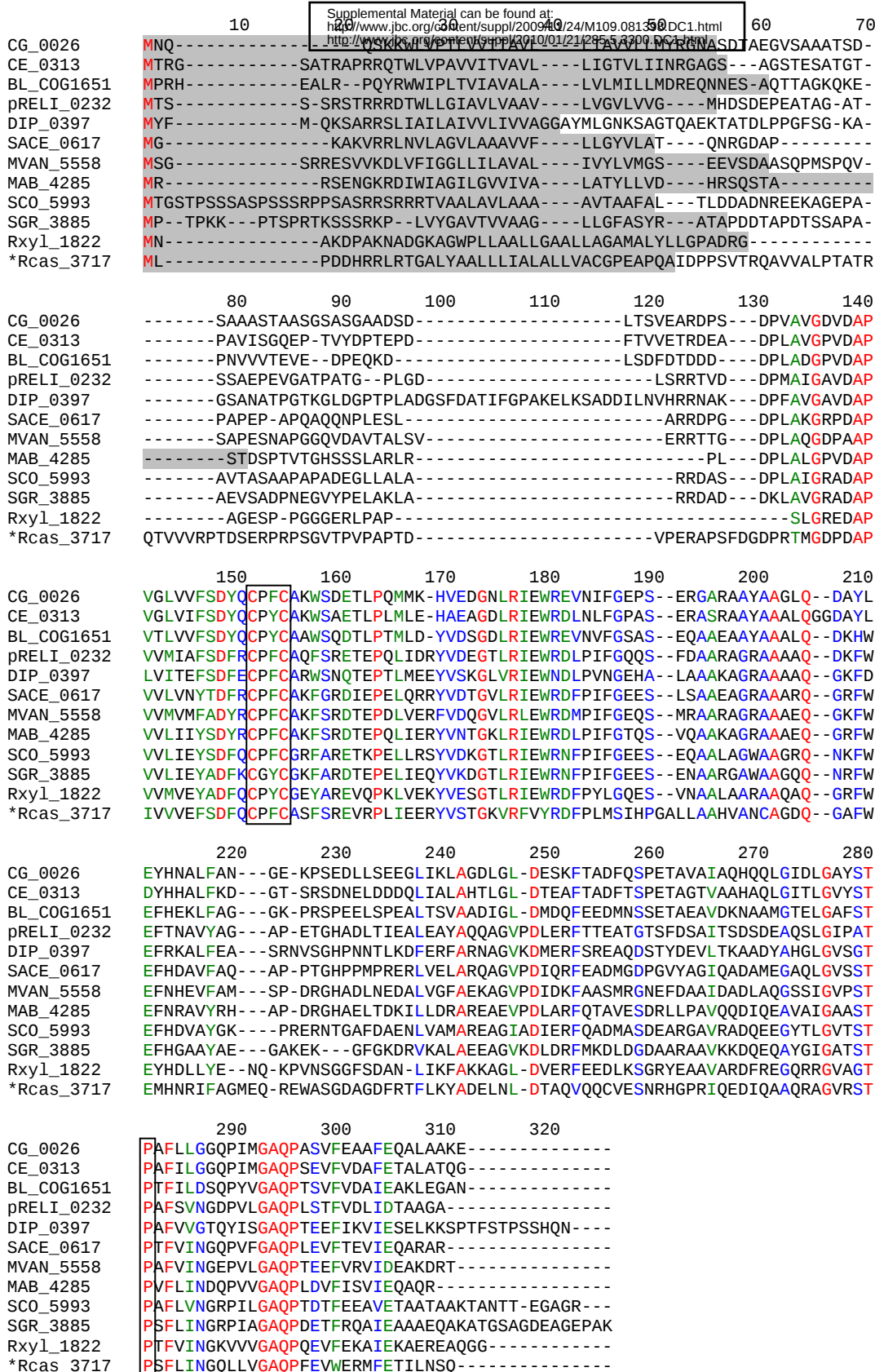


Figure S12. Sequence alignments of homologs to CG26 from *Corynebacterium glutamicum*. ClustalW alignments of the sequences using a homology cutoff of e^{-19} are shown with conserved, strongly similar and weakly similar residues colored in red, green and blue, respectively. The signal sequences are highlighted in grey, and the CXXC and the putative *cisPro* are boxed. CG, *Corynebacterium glutamicum*; CE, *Corynebacterium efficiens*; BL, *Brevibacterium linens*; pRELI, *Rhodococcus erythropolis*; DIP, *Corynebacterium diphtheriae*; SACE, *Saccharopolyspora erythraea*; MVAN, *Mycobacterium vanbaalenii*; MAB, *Mycobacterium abscessus*; SCO, *Streptomyces coelicolor*; SGR, *Streptomyces griseus*; and Rxy1, *Rubrobacter xylanophilus*. The asterisk denotes the only strain, Rcas, or *Roseiflexus castenholzii* that is not an Actinobacteria and comes from the phylum Chloroflexi.

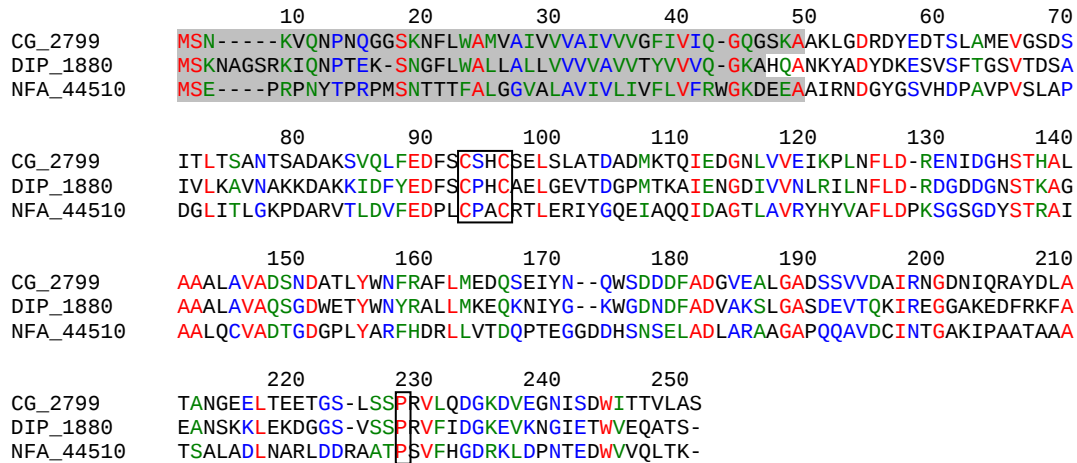


Figure S13. Sequence alignments of homologs to CG2799 from *Corynebacterium glutamicum*. ClustalW alignments of the sequences using a homology cutoff of e^{-11} are shown with conserved, strongly similar and weakly similar residues colored in red, green and blue, respectively. The signal sequences are highlighted in grey, and the CXXC motif and the putative cisPro are boxed. CG, *Corynebacterium glutamicum*; DIP, *Corynebacterium diphtheriae*; and NFA, *Nocardia Farcinica*. All homologs are from the Actinobacteria phylum.

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Mature bacterial exported proteins (secreted proteins and lipoproteins)

	Species	Trend	Number of secreted proteins				Number of lipoproteins			
			0 Cys	1 Cys	2 Cys	3 Cys	0 Cys	1 Cys	2 Cys	3 Cys
Gram-negative Bacteria	Acidiphilium cryptum (strain JF-5)	Even-bias	105	32	65	16	21	2	14	1
	Agrobacterium tumefaciens (strain C58 / ATCC 33970)	Even-bias	166	50	157	17	23	2	15	1
	Azorhizobium caulinodans (strain ATCC 43989 / DSM 5975 / ORS 571)	Even-bias	130	26	104	17	12	1	8	0
	Bartonella bacilliformis (strain ATCC 35685 / KC583)	Even-bias	31	4	16	2	6	0	7	1
	Bartonella henselae (strain Houston 1 / ATCC 49882)	Even-bias	31	7	27	4	9	2	8	2
	Bartonella quintana (strain Toulouse)	Even-bias	25	12	20	1	9	3	7	3
	Bartonella tribocorum CIP 105476	Even-bias	36	10	30	5	9	5	9	2
	Bradyrhizobium japonicum (strain USDA 110)	Even-bias	220	70	227	39	18	7	10	1
	Brucella abortus (strain 2308)	Even-bias	92	18	83	8	15	3	13	0
	Brucella canis ATCC 23365	Even-bias	100	19	85	13	17	3	15	0
	Brucella melitensis (biovar 1, strain NCTC 10094 / ATCC 23456 / 16M)	Even-bias	71	13	59	7	9	2	10	0
	Brucella suis ATCC 23445	Even-bias	96	21	82	13	15	4	14	0
	Caulobacter crescentus (strain CB15 / ATCC 19089)	Even-bias	139	30	118	14	36	4	15	0
	Dinoroseobacter shibae DFL 12	Even-bias	137	23	123	8	26	3	19	0
	Ehrlichia canis (strain Jake)	Even-bias	6	4	13	5	2	0	1	0
	Ehrlichia chaffeensis (strain Arkansas)	Even-bias	7	4	7	6	2	1	2	0
	Ehrlichia ruminantium (strain Gardel)	Even-bias	5	5	17	3	2	1	1	0
	Erythrobacter litoralis (strain HTCC2594)	Even-bias	101	16	86	3	38	5	32	1
	Gluconacetobacter diazotrophicus PAI 5	Even-bias	96	37	76	14	23	6	7	0
	Gluconobacter oxydans (strain 621H)	Even-bias	80	30	78	11	28	4	14	0
	Granulobacter thebesdensis (strain ATCC BAA-1260 / CGDNIH1)	Even-bias	53	14	44	10	16	1	10	0
	Hyphomonas neptunium (strain ATCC 15444)	Even-bias	117	19	101	9	74	7	68	0
	Jannaschia sp. (strain CCS1)	Even-bias	137	18	145	12	26	3	18	0
	Magnetospirillum magneticum (strain AMB-1 / ATCC 700264)	Even-bias	104	26	76	11	28	7	20	0
	Maricaulis maris (strain MCS10)	Even-bias	142	19	126	10	52	4	41	0
	Mesorhizobium sp. (strain BNC1)	Even-bias	152	31	138	17	20	4	17	0
	Methylobacterium extorquens PA1	Even-bias	208	45	128	16	19	4	12	0
	Neorickettsia sennetsu (strain Miyayama)	Even-bias	9	4	9	3	1	0	0	0
	Nitrobacter hamburgensis (strain X14 / DSM 10229)	Even-bias	134	37	124	18	17	2	13	0
	Nitrobacter winogradskyi (strain Nb-255 / ATCC 25391)	Even-bias	91	27	89	19	13	4	11	0
	Novosphingobium aromaticivorans (strain DSM 12444)	Even-bias	142	21	122	15	30	6	28	0
	Orientia tsutsugamushi (strain Boryong)	Remaining	4	6	5	7	1	0	0	0
	Paracoccus denitrificans (strain Pd 1222)	Even-bias	184	38	136	18	25	4	24	0
	Parvibaculum lavamentivorans (strain DS-1 / DSM 13023 / NCIMB 13966)	Even-bias	120	18	78	9	27	2	15	0
	Pelagibacter ubique (strain HTCC1062)	Even-bias	16	4	30	2	11	0	5	0
	Rhizobium etli (strain CFN 42 / ATCC 51251)	Even-bias	156	55	159	17	26	1	21	0
	Rhizobium leguminosarum bv. viciae (strain 3841)	Even-bias	194	72	196	24	26	3	23	0
	Rhizobium loti (strain MAFF303099)	Even-bias	150	38	185	20	25	2	17	0
	Rhizobium meliloti (strain 1021)	Even-bias	181	44	187	17	19	4	14	0
	Rhodobacter sphaeroides (strain ATCC 17023 / 2.4.1 / NCIB 8253 / DSM 158)	Even-bias	115	24	116	15	22	6	21	0
	Rhodopseudomonas palustris (strain ATCC BAA-98 / CGA009)	Even-bias	167	44	134	21	25	2	14	0
	Rhodospirillum rubrum (strain ATCC 11170 / NCIB 8255)	Even-bias	127	25	58	14	32	2	11	0
	Roseobacter denitrificans (strain OCH 114 / ATCC 33942)	Even-bias	131	30	121	15	21	5	13	0
	Silicibacter pomeroyi (strain ATCC 700808 / DSS-3 / DSM 15171)	Even-bias	103	27	120	14	24	5	14	0
	Silicibacter sp. (strain TM1040)	Even-bias	112	17	103	12	23	2	18	1
	Sinorhizobium medicae (strain WSM419)	Even-bias	182	48	190	28	24	5	21	1
	Sphingomonas wittichii (strain RW1 / DSM 6014 / JCM 10273)	Even-bias	197	35	163	19	39	2	16	1
	Sphingopyxis alaskensis	Even-bias	130	27	105	13	40	2	34	0
	Wolbachia pipientis wMel	Remaining	7	8	5	5	0	1	1	1
	Wolbachia sp. (subsp. Brugia malayi, strain TRS)	Remaining	4	5	6	2	1	1	0	1
	Xanthobacter sp.	Even-bias	147	32	134	17	14	2	13	0
	Zymomonas mobilis (strain CP4 / ZM4 / ATCC 31821)	Even-bias	56	16	32	4	16	4	2	0
Alpha proteobacteria										
Aerobic										

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Beta	Anaerobic	Anaplasma marginale (strain St. Maries)	Remaining	4	8	9	6	3	1	2	1
		Anaplasma phagocytophilum (strain HZ)	Remaining	4	8	7	5	2	1	1	2
		Brucella ovis (strain ATCC 25840 / 63/290 / NCTC 10512)	Even-bias	81	17	66	8	11	3	14	0
		Ochrobactrum anthropi (strain ATCC 49188 / DSM 6882 / NCTC 12168)	Even-bias	173	42	145	19	22	2	22	0
		Rickettsia akari (strain Hartford)	Even-bias	10	6	14	6	7	4	3	2
		Rickettsia bellii (strain OSU 85-389)	Even-bias	19	12	13	6	5	2	4	5
		Rickettsia canadensis (strain McKiel)	Exclusionary	12	10	6	4	4	3	4	1
		Rickettsia conorii (strain ATCC VR-613 / Malish 7)	Exclusionary	9	13	8	5	9	1	3	2
		Rickettsia felis (strain ATCC VR-1525 / URRWXC2)	Exclusionary	17	14	11	4	7	3	4	3
		Rickettsia massiliae (strain Mtu5)	Even-bias	10	8	8	5	7	2	5	2
	Aerobic	Rickettsia prowazekii (strain Madrid E)	Remaining	5	13	9	3	6	3	2	1
		Rickettsia rickettsii Iowa	Exclusionary	12	11	9	4	8	2	3	3
		Rickettsia typhi (strain Wilmington / ATCC VR-144)	Remaining	5	10	8	3	6	3	2	0
		Acidovorax avenae (subsp. citrulli, strain AAC00-1)	Even-bias	158	43	94	13	38	3	26	0
		Acidovorax sp. (strain JS42)	Even-bias	146	39	91	16	43	2	37	0
		Bordetella avium (strain 197N)	Even-bias	130	12	86	12	34	4	23	0
		Bordetella bronchiseptica (strain NCTC 13252 / RB50 / ATCC BAA-588)	Even-bias	246	48	102	19	44	1	22	0
		Bordetella parapertussis (strain NCTC 13253 / ATCC BAA-587 / 12822)	Even-bias	194	41	86	16	37	1	20	0
		Bordetella pertussis (strain ATCC BAA-589 / Tohama I / NCTC 13251)	Even-bias	149	25	70	13	33	3	16	0
		Bordetella petrii	Even-bias	187	22	108	19	46	5	22	0
	Anaerobic	Burkholderia cenocepacia (strain AU 1054)	Even-bias	219	46	142	24	61	9	27	0
		Burkholderia cepacia (strain ATCC 53795 / AMMD)	Even-bias	231	54	154	20	69	4	38	0
		Burkholderia mallei (strain ATCC 23344)	Even-bias	120	40	84	21	41	9	20	0
		Burkholderia multivorans ATCC 17616	Even-bias	209	42	129	21	66	3	26	0
		Burkholderia pseudomallei (strain 668)	Even-bias	182	41	119	20	60	7	34	0
		Burkholderia thailandensis (strain E264 / ATCC 700388 / DSM 13276 / CIP 106301)	Even-bias	151	41	106	16	47	5	16	0
		Burkholderia vietnamiensis (strain R1808 / G4 / LMG 22486) / LMG 22486)	Even-bias	245	54	161	27	65	8	38	0
		Burkholderia xenovorans (strain LB400)	Even-bias	272	51	145	31	54	6	29	0
		Chromobacterium violaceum (strain IFO 12614 / ATCC 12472 / DSM 30191 / NCIB 9131 / JCM 1249)	Even-bias	151	27	117	14	38	8	14	0
		Dechloromonas aromatica (strain RCB)	Even-bias	169	27	120	16	48	4	32	0
Beta	Aerobic	Delftia acidovorans SPH-1	Even-bias	249	46	138	22	64	6	29	0
		Janthinobacterium sp. (strain Marseille)	Even-bias	183	27	92	12	55	3	24	0
		Methylobium petroleiphilum (strain PM1)	Even-bias	152	45	127	14	57	4	18	0
		Methylobacillus flagellatus (strain KT / ATCC 51484 / DSM 6875)	Even-bias	125	25	54	8	41	6	12	0
		Neisseria gonorrhoeae (strain ATCC 700825 / FA 1090)	Even-bias	49	12	28	7	29	4	18	0
		Neisseria meningitidis (serogroup C / , serovar 2a, strain ATCC 700532 / FAM18)	Even-bias	54	8	28	9	26	3	20	0
		Nitrosomonas europaea (strain IFO 14298 / ATCC 19718)	Even-bias	71	14	45	6	42	3	12	0
		Nitrosomonas eutropha (strain C71)	Even-bias	78	16	44	4	32	4	11	0
		Nitrospira multiformis (strain ATCC 25196 / NCIMB 11849)	Even-bias	97	27	42	8	38	6	17	0
		Polaromonas naphthalenivorans (strain CJ2)	Even-bias	165	41	104	19	51	3	21	0
		Polaromonas sp. (strain JS666 / ATCC BAA-500)	Even-bias	201	38	126	20	45	1	18	0
		Polynucleobacter sp. (strain QLW-P1DMWA-1)	Even-bias	93	19	57	10	20	1	22	0
		Ralstonia eutropha (strain ATCC 17699 / H16 / DSM 428 / Stanier 337)	Even-bias	267	64	121	16	55	2	36	0
Beta	Aerobic	Ralstonia metallidurans (strain CH34 / ATCC 43123)	Even-bias	270	52	119	15	57	6	42	0
		Ralstonia solanacearum (strain GMI1000)	Even-bias	181	65	119	19	47	6	29	0
		Thiobacillus denitrificans (strain ATCC 25259)	Even-bias	134	27	83	10	41	3	14	0
		Verminephrobacter eiseniae (strain EF01-2)	Even-bias	149	58	109	26	20	2	17	0
	Anaerobic	Hermineimonas arsenicoxydans (strain ULPAs1)	Even-bias	122	26	73	14	40	5	20	0
		Rhodoferrax ferrireducens (strain DSM 15236 / ATCC BAA-621 / T118)	Even-bias	140	41	99	13	46	3	19	0

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Delta Proteobacteria	Aerobic	Bdellovibrio bacteriovorus (strain DSM 50701 / NCIB 9529 / HD100 / ATCC 15356)	Even-bias	191	60	183	29	57	19	44	8
		Geobacter uraniumreducens (strain Rf4)	Exclusionary	149	92	75	26	35	15	19	4
		Lawsonia intracellularis (strain PHE/MN1-00)	Exclusionary	18	11	8	3	10	3	5	2
		Myxococcus xanthus (strain DK 1622)	Even-bias	245	81	159	49	9	8	7	1
		Sorangium cellulosum (strain So ce56)	Even-bias	110	71	112	43	103	43	108	31
	Anaerobic	Anaeromyxobacter dehalogenans (strain 2CP-C)	Even-bias	147	72	93	24	48	23	27	7
		Desulfococcus oleovorans Hxd3	Exclusionary	47	37	33	24	22	14	16	8
		Desulfotalea psychrophila (strain LSV54 / DSM 12343)	Exclusionary	64	60	44	17	1	0	0	0
		Desulfovibrio desulfuricans (strain G20)	Exclusionary	46	47	48	18	20	12	10	2
		Desulfovibrio vulgaris (strain Hildenborough / ATCC 29579 / NCIMB 8303)	Exclusionary	63	54	44	21	1	0	0	0
		Geobacter metallireducens (strain GS-15 / ATCC 53774 / DSM 7210)	Exclusionary	125	82	70	22	1	0	0	0
		Geobacter sulfurreducens (strain ATCC 51573 / DSM 12127 / PCA)	Exclusionary	113	67	65	23	1	0	0	0
		Pelobacter carbinolicus (strain DSM 2380 / Gra Bd 1)	Exclusionary	57	40	38	22	41	14	12	0
		Pelobacter propionicus (strain DSM 2379)	Exclusionary	92	58	45	12	29	21	15	0
		Syntrophobacter fumaroxidans (strain DSM 10017 / MPOB)	Even-bias	62	59	59	39	27	10	12	0
		Syntrophus aciditrophicus (strain SB)	Exclusionary	47	31	26	11	20	12	7	0
		Arcobacter butzleri (strain RM4018)	Even-bias	68	17	34	5	21	5	19	0
Epsilon Proteobacteria	Aerobic	Campylobacter concisus (strain 13826)	Even-bias	62	19	38	10	21	1	20	0
		Campylobacter curvus (strain 525.92)	Even-bias	66	17	32	5	22	3	12	0
		Campylobacter fetus (subsp. fetus, strain 82-40)	Even-bias	52	12	25	1	18	4	8	0
		Campylobacter hominis (strain ATCC BAA-381 / LMG 19568 / NCTC 13146 / CH001A)	Even-bias	28	10	25	3	13	5	9	0
		Campylobacter jejuni (subsp. doylei, strain ATCC BAA-1458 / RM4099 / 269.97)	Even-bias	33	8	18	2	11	3	9	0
		Helicobacter acinonychis (strain Sheeba)	Even-bias	43	7	33	3	15	3	10	0
		Helicobacter hepaticus (strain 3B1 / ATCC 51449)	Even-bias	47	7	20	3	18	8	18	0
		Helicobacter pylori (strain 26695 / ATCC 700392)	Even-bias	48	13	41	3	18	2	11	0
		Nitratiruptor sp. (strain SB155-2)	Even-bias	39	15	29	11	8	6	9	0
		Sulfurimonas denitrificans (strain ATCC33889/DSM 1251)	Even-bias	47	36	34	9	13	4	11	0
		Sulfurovum sp. (strain NBC37-1)	Even-bias	87	41	66	27	24	3	19	0
	Anaerobic	Wolinella succinogenes (strain DSMZ 1740)	Even-bias	56	15	24	4	20	5	12	0
Gamma Proteobacteria	Aerobic	Acinetobacter baumannii (strain ATCC 17978)	Even-bias	26	3	8	3	15	0	8	0
		Acinetobacter sp. (strain ADP1)	Even-bias	129	15	52	1	44	5	26	0
		Actinobacillus pleuropneumoniae (serovar5b, strain L20)	Even-bias	105	11	43	6	1	0	0	0
		Actinobacillus succinogenes (strain ATCC 55618 / 130Z)	Even-bias	91	21	38	3	29	2	14	0
		Aeromonas hydrophila (subsp. hydrophila, strain ATCC 7966 / NCIB 9240)	Even-bias	140	31	94	11	53	3	26	0
		Aeromonas salmonicida (strain A449)	Even-bias	173	39	116	15	32	2	11	0
		Alcanivorax borkumensis (strain SK2 / ATCC 700651 / DSM 11573)	Even-bias	105	19	56	11	49	4	21	0
		Baumannia cicadellinicola coagulata (subsp. Homalodisca)	<25 proteins	3	1	1	2	1	4	1	0
		Blochmannia floridanus	<25 proteins	0	0	0	1	0	0	2	0
		Blochmannia pennsylvanicus (strain BPEN)	<25 proteins	0	0	0	1	1	1	0	1
		Buchnera aphidicola (subsp. Acyrthosiphon pisum, strain Tokyo 1998)	<25 proteins	0	2	4	1	2	0	0	0
		Buchnera aphidicola cedri (subsp. Cinara)	<25 proteins	0	1	1	0	0	0	0	0
		Chromohalobacter salexigens (strain DSM 3043 / ATCC BAA-138 / NCIMB 13768)	Even-bias	139	7	54	6	32	3	13	1
		Citrobacter koseri (strain ATCC BAA-895 / CDC 4225-83 / SGSC4696)	Even-bias	160	20	99	11	41	9	19	0
		Colwellia psychrerythraea	Even-bias	217	41	140	12	82	13	50	4
		Coxiella burnetii (strain Dugway 5J108-111)	Even-bias	42	14	21	6	17	3	6	1
		Dichelobacter nodosus (strain VCS1703A)	Even-bias	33	8	18	4	25	1	9	1
		Enterobacter sakazakii (strain ATCC BAA-894)	Even-bias	134	25	100	8	46	5	21	2
		Erwinia carotovora (subsp. atroseptica, strain ATCC BAA-672 / SCRI 1043)	Even-bias	184	21	110	8	58	3	33	2

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	Escherichia coli (strain K12)	Even-bias	150	27	114	12	54	9	30	4	
	Francisella tularensis (subsp. tularensis, strain FSC 198)	Even-bias	37	10	22	4	25	1	13	2	
	Haemophilus ducreyi (strain ATCC 700724 / 35000HP)	Even-bias	51	8	34	3	2	0	0	0	
	Haemophilus influenzae (strain ATCC 51907 / DSM 11121 / KW20 / Rd)	Even-bias	62	5	31	4	9	3	4	0	
	Haemophilus somnus (strain 129Pt)	Even-bias	46	3	18	1	19	2	12	0	
	Hahella chejuensis (strain KCTC 2396)	Even-bias	237	56	165	23	74	13	52	6	
	Halorhodospira halophila (strain DSM 244 / SL1)	Even-bias	106	11	23	3	36	2	12	1	
	Idiomarina loihiensis (strain L2-TR / DSM 15497 / ATCC BAA-735)	Even-bias	146	15	67	4	57	2	18	0	
	Klebsiella pneumoniae (subsp. pneumoniae, strain ATCC 700721 / MGH 78578)	Even-bias	184	18	107	4	47	3	21	3	
	Legionella pneumophila (strain Paris)	Even-bias	88	27	59	18	33	9	12	1	
	Marinobacter aquaeolei (strain DSM 11845 / ATCC 700491 / VT8) / VT8)	Even-bias	158	21	93	17	66	3	24		Downloaded from jbc.org at National Institute of Health on October 7, 2015
	Marinomonas sp. MWYL1	Even-bias	162	17	92	10	56	2	21		
	Methylococcus capsulatus (strain Bath / NCIMB 11132)	Even-bias	79	26	32	9	27	10	15		
	Nitrosococcus oceani (strain ATCC 19707 / NCIMB 11848)	Even-bias	84	24	45	12	46	3	11		
	Pasteurella multocida (strain Pm70)	Even-bias	65	8	41	5	33	3	23		
	Photobacterium profundum (strain SS9)	Even-bias	254	32	138	19	1	0	0		
	Photorhabdus luminescens laumondii (strain TT01)	Even-bias	91	8	78	6	38	9	14		
	Pseudoalteromonas atlantica (strain T6c / BAA-1087)	Even-bias	211	46	119	16	69	16	56		
	Pseudoalteromonas haloplanktis (strain TAC 125)	Even-bias	166	21	94	7	75	7	33		
	Pseudomonas aeruginosa (strain LMG 12228 / ATCC 15692 / PRS 101 / 1C / PAO1)	Even-bias	235	36	149	15	80	10	52		
	Pseudomonas entomophila (strain L48)	Even-bias	221	33	114	17	74	11	39		
	Pseudomonas fluorescens (strain Pf-5 / ATCC BAA-477)	Even-bias	232	46	142	17	83	11	46		
	Pseudomonas mendocina (strain ymp)	Even-bias	216	39	104	13	69	8	38		
	Pseudomonas putida (strain F1 / ATCC 700007)	Even-bias	230	26	124	11	68	15	37		
	Pseudomonas stutzeri (strain A1501)	Even-bias	128	26	90	7	48	6	25		
	Pseudomonas syringae (pathovar phaseolicola, strain 1448A / Race 6)	Even-bias	177	29	98	12	68	9	29		
	Psychrobacter arcticum (strain 273-4)	Even-bias	45	12	21	2	32	1	20		
	Psychrobacter cryohalolentis (strain K5)	Even-bias	59	13	25	4	42	1	31		
	Psychrobacter sp. (strain PRwf-1)	Even-bias	65	11	31	0	41	2	31		
	Saccharophagus degradans (strain 2-40 / ATCC 43961 / DSM 17024)	Even-bias	177	53	116	19	74	13	57		
	Salmonella arizonae (strain ATCC BAA-731 / CDC346-86 / RSK2980)	Even-bias	120	25	85	9	43	6	23		
	Salmonella choleraesuis (strain SC-B67)	Even-bias	142	25	113	14	46	8	25		
	Salmonella paratyphi A (strain SARB42 / ATCC 9150)	Even-bias	138	22	109	12	42	8	25		
	Salmonella paratyphi B (strain ATCC BAA-1250 / SPB7)	Even-bias	145	22	111	11	40	10	23		
	Salmonella typhi (strain ATCC 700931 / Ty2)	Even-bias	149	27	114	14	46	10	25		
	Salmonella typhimurium (strain ATCC 700720 / SGSC1412 / LT2)	Even-bias	161	25	118	10	45	10	26		
	Serratia proteamaculans (strain 568)	Even-bias	178	31	132	7	62	4	27		
	Shewanella amazonensis (strain ATCC BAA-1098 / SB2B)	Even-bias	199	35	101	13	60	11	30		
	Shewanella baltica (strain OS155 / ATCC BAA-1091)	Even-bias	202	30	101	11	57	13	30		
	Shewanella denitrificans (strain OS217 / ATCC BAA-1090 / DSM 15013)	Even-bias	196	32	94	7	66	6	30		
	Shewanella frigidimarina (strain NCIMB 400)	Even-bias	199	31	93	14	67	11	32		
	Shewanella loihica (strain BAA-1088 / PV-4)	Even-bias	212	33	114	11	65	13	33		
	Shewanella oneidensis (strain MR-1)	Even-bias	171	29	105	13	62	10	29		
	Shewanella pealeana ATCC 700345	Even-bias	226	25	125	17	71	14	37		
	Shewanella putrefaciens (strain CN-32/ATCC BAA-453)	Even-bias	191	21	84	10	64	12	24		
	Shewanella sediminis HAW-EB3	Even-bias	226	44	111	16	82	15	27		
	Shewanella sp. (strain ANA-3)	Even-bias	215	27	120	14	69	10	37		
	Shigella boydii (serovar 4, strain Sb227)	Even-bias	106	27	68	7	37	7	19		
	Shigella dysenteriae (serovar 1, strain Sd97 / Sd197)	Even-bias	101	25	68	7	34	8	15		
	Shigella flexneri (serovar 2a, strain 2457T/ATCC 700930)	Even-bias	104	19	71	11	43	9	18		
	Shigella sonnei (strain Ss046)	Even-bias	120	23	77	12	41	11	19		
	Sodalis glossinidius (strain morsitans)	Even-bias	48	21	37	3	21	10	5		
	Vesicomysocius okutanii (subsp. Calyptogena okutanii, strain HA)	Even-bias	17	5	12	2	6	2	1		
	Vibrio cholerae (strain ATCC 39541 / O395)	Even-bias	141	25	68	7	42	2	23		

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Gram-positive Bacteria	Actinobacteria	Aerobic	Vibrio fischeri (strain ATCC 700601 / ES114)	Even-bias	171	19	86	5	68	5	49	2
			Vibrio harveyi (strain ATCC BAA-1116 / BB120)	Even-bias	196	36	118	9	60	6	54	9
			Vibrio parahaemolyticus (serovar O3:K6, strain RIMD 2210633)	Even-bias	201	30	110	9	69	3	52	4
			Vibrio vulnificus (strain CMCP6)	Even-bias	170	19	92	6	49	1	32	2
			Wigglesworthia glossinidia brevipalpis	<25 proteins	2	0	2	0	3	4	0	0
			Xanthomonas axonopodis citri (strain 306)	Even-bias	173	44	113	11	48	10	38	2
			Xanthomonas campestris (strain ATCC 33913)	Even-bias	168	43	112	19	47	9	47	0
			Xanthomonas oryzae (pathovar oryzae, MAFF 311018)	Even-bias	146	39	87	26	47	7	37	1
			Xylella fastidiosa (strain 9a5c)	Even-bias	58	18	24	11	27	6	12	0
			Xylella fastidiosa (strain Temecula1 / ATCC 700964)	Even-bias	49	15	28	11	27	6	13	1
			Yersinia enterocolitica (serovar O:8 / strain 8081)	Even-bias	139	15	98	7	55	4	17	0
			Yersinia pestis (biovar Antiqua Antiqua, strain Antiqua)	Even-bias	154	19	106	6	54	3	14	1
			Yersinia pseudotuberculosis (serovar I, strain IP32953)	Even-bias	162	21	108	4	53	5	17	1
			Alkalilimnicola ehrlichei (strain MLHE-1)	Even-bias	108	17	48	8	36	1	15	1
			Carsonella ruddii (strain PV)	<25 proteins	0	0	0	0	0	0	0	0
			Mannheimia succiniciproducens (strain MBEL55E)	Even-bias	63	9	30	2	35	2	18	3
			Psychromonas ingrahamii (strain 37)	Even-bias	123	18	68	7	42	5	19	1
			Ruthia magnifica magnifica (subsp. Calyptogena)	Even-bias	22	4	10	3	10	0	2	2
			Thiomicrospira crunogena (strain XCL-2)	Even-bias	100	19	39	3	27	3	18	2
		Anaerobic	Acidothermus cellulolyticus (strain ATCC 43068 / 11B)	Even-bias	18	8	20	7	4	3	14	1
			Arthrobacter aurescens (strain TC1)	Even-bias	38	16	44	8	32	4	39	4
			Clavibacter michiganensis (subsp. michiganensis, strain NCPPB 382)	Even-bias	41	16	41	4	28	2	26	1
			Corynebacterium diphtheriae (biovar gravis, strain NCTC 13129 / ATCC 700971)	Even-bias	19	8	33	5	15	3	21	1
			Corynebacterium efficiens (strain DSM 44549 / NBRC 100395 / JCM 11189 / YS-314 / AJ 12310)	Even-bias	28	7	40	5	15	2	14	1
			Corynebacterium glutamicum (strain R)	Even-bias	28	11	32	9	33	4	30	1
			Corynebacterium jeikeium (strain K411)	Even-bias	32	6	32	10	14	2	17	1
			Frankia alni (strain ACN14a)	Even-bias	40	13	50	8	13	1	11	1
			Kineococcus radiotolerans SRS30216	Even-bias	60	19	63	4	19	7	26	1
			Leifsonia xyli xyli (strain CTCB07)	Even-bias	17	13	18	2	12	3	12	1
	Mycobacterium avium (strain 104)		Even-bias	29	20	76	9	24	7	23	1	
	Mycobacterium bovis (strain ATCC BAA-935)		Even-bias	24	23	43	12	11	5	21	1	
	Mycobacterium gilvum PYR-GCK		Even-bias	56	15	104	8	28	2	40	1	
	Mycobacterium leprae (strain TN)		Even-bias	14	8	26	6	4	4	6	1	
	Mycobacterium paratuberculosis (strain ATCC BAA-968 / K-10)		Even-bias	26	26	62	16	22	9	16	1	
	Mycobacterium smegmatis (strain ATCC 700084 / mc(2 / mc(2)155)		Even-bias	70	21	94	13	45	7	56	1	
	Mycobacterium tuberculosis (strain ATCC 25177 / H37Ra)		Even-bias	26	21	48	12	12	5	21	1	
	Mycobacterium ulcerans (strain Agy99)		Even-bias	34	20	54	15	14	5	25	1	
	Mycobacterium vanbaalenii (strain DSM 7251 / PYR-1)		Even-bias	63	25	125	13	34	3	38	1	
	Nocardia farcinica (strain IFM 10152)		Even-bias	40	22	72	16	25	7	43	1	
	Nocardioideis sp. (strain BAA-499 / JS614)	Even-bias	52	23	81	10	29	2	39	1		
	Renibacterium salmoninarum (strain ATCC 33209)	Even-bias	29	18	24	9	16	10	16	1		
	Rhodococcus sp. (strain RHA1)	Even-bias	65	27	85	14	53	8	54	1		
	Rubrobacter xylanophilus (strain DSM 9941 / NBRC 16129)	Even-bias	39	18	19	5	20	3	9	1		
	Saccharopolyspora erythraea (strain NRRL 23338)	Even-bias	62	21	89	20	23	6	32	1		
	Salinispora arenicola CNS-205	Even-bias	37	15	47	8	15	9	29	1		
	Salinispora tropica CNB-440	Even-bias	27	19	32	6	13	7	26	0		
	Streptomyces avermitilis (strain DSM 46492 / NRRL 8165 / NCIMB 12804 / ATCC 31267 / JCM 5070)	Even-bias	68	48	99	36	38	7	42	6		
	Streptomyces coelicolor (strain ATCC BAA-471 / M145 / A3(2))	Even-bias	75	40	108	37	49	12	57	3		
	Thermobifida fusca (strain YX)	Even-bias	25	13	34	4	17	6	17	0		
	Tropheryma whipplei (strain Twist)	Even-bias	6	1	8	1	4	0	2	0		
	Anaerobic	Bifidobacterium adolescentis (strain ATCC 15703 / DSM 20083)	Exclusionary	14	9	8	5	5	5	5	2	
		Bifidobacterium longum (strain NCC 2705)	Exclusionary	8	9	6	6	12	4	5	4	
		Propionibacterium acnes (strain DSM 16379 / KPA171202)	Even-bias	9	8	30	4	14	8	13	5	

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Firmicutes	Aerobic	Acholeplasma laidlawii (strain PG-8A)	Exclusionary	27	1	0	0	57	5	2	0
		Bacillus amyloliquefaciens (strain FZB42)	Exclusionary	89	34	31	5	62	14	5	0
		Bacillus anthracis (strain Ames, isolate Porton)	Exclusionary	111	34	21	1	102	11	9	2
		Bacillus cereus (strain ATCC 10987)	Exclusionary	112	35	26	2	109	17	7	2
		Bacillus clausii (strain KSM-K16)	Even-bias	74	20	18	0	123	8	13	0
		Bacillus halodurans (strain C-125 / ATCC BAA-125 / JCM 9153 / FERM 7344 / DSM 18197)	Even-bias	90	21	19	4	103	7	12	0
		Bacillus licheniformis (strain DSM 13 / ATCC 14580, sub strain Goettingen)	Exclusionary	101	37	33	5	85	7	3	0
		Bacillus pumilus (strain SAFR-032)	Exclusionary	107	23	19	1	73	7	4	1
		Bacillus subtilis (strain 168)	Exclusionary	104	35	28	2	72	14	5	3
		Bacillus thuringiensis (strain Al Hakam)	Exclusionary	119	34	18	1	98	10	6	2
		Bacillus weihenstephanensis KBAB4	Exclusionary	103	35	22	3	119	14	7	3
		Desulfitobacterium hafniense (strain Y51)	Exclusionary	52	44	26	11	56	30	21	0
		Enterococcus faecalis (strain V583 / ATCC 700802)	Exclusionary	68	14	10	1	62	4	2	0
		Geobacillus kaustophilus (strain HTA426)	Exclusionary	63	30	14	4	49	8	8	0
		Geobacillus thermodenitrificans (strain NG80-2)	Exclusionary	53	19	16	6	60	9	6	0
		Lactobacillus acidophilus (strain NCFM)	Exclusionary	37	24	8	5	25	5	1	0
		Lactobacillus brevis (strain ATCC 367 / JCM 1170)	Exclusionary	64	23	11	2	18	1	0	0
		Lactobacillus casei (strain ATCC 334)	Exclusionary	75	10	4	1	31	3	0	0
		Lactobacillus delbrueckii (subsp. bulgaricus, strain ATCC BAA-365)	Exclusionary	36	16	5	2	14	2	1	0
		Lactobacillus gasserii (strain ATCC 33323 / DSM 20243)	Exclusionary	25	8	2	2	19	6	2	0
		Lactobacillus helveticus (strain DPC 4571)	Exclusionary	31	18	5	1	14	3	1	0
		Lactobacillus johnsonii (strain NCC 533)	Exclusionary	27	8	4	1	20	9	1	0
		Lactobacillus plantarum (strain WCFS1 / ATCC BAA-793 / NCIMB 8826)	Exclusionary	60	21	4	1	35	8	1	0
		Lactobacillus reuteri (strain ATCC 23272 / DSM 20016 / F275)	Exclusionary	35	17	6	0	9	4	0	0
		Lactobacillus sakei (subsp. sakei, strain 23K)	Exclusionary	67	37	10	6	22	2	0	0
		Lactobacillus salivarius (subsp. salivarius, strain UCC118)	Exclusionary	34	15	3	1	18	1	0	0
		Lactococcus lactis (subsp. lactis, strain IL1403)	Exclusionary	57	23	5	0	26	4	0	0
		Leuconostoc mesenteroides (subsp. mesenteroides, strain ATCC 8293 / NCDO 523)	Exclusionary	73	7	3	0	2	0	0	0
		Listeria innocua (serovar 6a, strain CLIP 11262)	Exclusionary	65	18	5	2	50	9	1	0
		Listeria monocytogenes (serovar 1/2a, strain EGD-e / ATCC BAA-679)	Exclusionary	55	18	8	4	52	6	1	0
		Listeria welshimeri (serovar 6b, strain ATCC 35897 / DSM 20650 / SLCC5334)	Exclusionary	58	17	5	4	51	9	1	0
		Mesoplasma florum (strain ATCC 33453 / NCTC 11704 / L1 / NBRC 100688)	Exclusionary	12	10	2	1	9	1	2	0
		Mycoplasma agalactiae	Exclusionary	19	33	6	3	22	1	1	0
		Mycoplasma capricolum (subsp. capricolum, strain California kid / ATCC 27343 / NCTC 10154)	Exclusionary	18	14	2	0	24	3	1	0
		Mycoplasma gallisepticum (strain R(low))	Exclusionary	25	5	1	0	18	0	1	0
		Mycoplasma genitalium (strain ATCC 33530 / NCTC 10195 / G-37)	Even-bias	9	0	0	1	12	2	3	0
		Mycoplasma hyopneumoniae (strain J / ATCC 25934 / NCTC 10110)	Remaining	37	2	0	0	26	1	3	0
		Mycoplasma mobile (strain 163K / NCTC 11711 / ATCC 43663)	Exclusionary	25	8	0	0	6	0	0	0
		Mycoplasma pneumoniae (strain M129 / ATCC 29342)	Even-bias	10	2	4	3	19	2	1	0
		Mycoplasma pulmonis (strain UAB CTIP)	Exclusionary	33	4	2	0	35	0	0	0
		Mycoplasma synoviae (strain 53)	Exclusionary	19	2	0	0	15	0	0	0
		Oceanobacillus iheyensis (strain KCTC 3954 / DSM 14371 / JCM 11309 / HTE831)	Even-bias	80	11	18	0	102	5	10	1
		Oenococcus oeni (strain BAA-331 / PSU-1)	Exclusionary	49	11	2	2	3	0	0	0
		Onion yellows phytoplasma (strain OY-M)	Remaining	10	12	3	2	0	1	0	0
		Pediococcus pentosaceus (strain ATCC25745 / 183-1w)	Exclusionary	34	6	4	0	18	2	0	0
		Pelotomaculum thermopropionicum (strain DSM 13744 / JCM 10971 / SI)	Exclusionary	30	26	16	13	11	5	8	5
		Staphylococcus aureus (strain Mu50 / ATCC 700699)	Exclusionary	66	16	4	3	43	5	3	0
		Staphylococcus epidermidis (strain ATCC 12228)	Exclusionary	46	14	4	1	40	4	2	0
		Staphylococcus haemolyticus (strain JCSC1435)	Exclusionary	34	15	4	0	34	4	1	0
		Staphylococcus saprophyticus (subsp. saprophyticus, strain ATCC 15305 / DSM 20229)	Exclusionary	44	10	1	0	34	2	2	0

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Anaerobic	Streptococcus agalactiae Ia (serovar Ia, strain A909 / CDC SS700 / ATCC 27591)	Exclusionary	59	15	4	1	30	1	0	0
	Streptococcus agalactiae III (serovar III, strain NEM316)	Exclusionary	56	18	5	1	27	3	0	0
	Streptococcus agalactiae V (serovar V, strain 2603 V/R / ATCC BAA-611)	Exclusionary	57	15	3	0	27	1	0	0
	Streptococcus gordonii (strain Challis / ATCC 35105 / CH1 / DL1 / V288)	Remaining	52	5	8	2	47	5	2	1
	Streptococcus mutans (serovar c, strain ATCC 700610 / UA159)	Exclusionary	40	10	6	1	22	1	0	0
	Streptococcus pneumoniae (serovar 2, strain D39 / NCTC 7466)	Even-bias	31	2	4	0	27	0	2	1
	Streptococcus pneumoniae (strain ATCC BAA-255 / R6)	Even-bias	32	5	4	0	25	0	3	1
	Streptococcus pneumoniae (strain TIGR4 / ATCC BAA-334)	Even-bias	35	3	4	0	28	0	2	1
	Streptococcus pyogenes M1 (serovar M1, strain MGAS5005 / ATCC BAA-947)	Exclusionary	40	16	8	4	21	2	2	0
	Streptococcus sanguinis (strain SK36)	Exclusionary	61	8	2	2	38	7	3	0
	Streptococcus suis (strain 05ZYH33)	Exclusionary	38	6	5	2	26	5	1	0
	Streptococcus thermophilus (strain ATCC BAA-491 / LMD-9)	Exclusionary	35	7	1	0	13	6	0	0
	Symbiobacterium thermophilum (strain IAM 14863 / T)	Even-bias	100	16	29	4	0	0	0	0
	Ureaplasma parvum (serovar 3, strain ATCC 700970)	Even-bias	11	7	6	3	15	2	4	0
	Alkaliphilus metalliredigens (strain QYMF)	Exclusionary	185	39	11	3	0	0	0	0
	Alkaliphilus oremlandii (strain OhILAs)	Exclusionary	115	30	14	5	10	8	0	0
	Caldicellulosiruptor saccharolyticus (strain ATCC 43494 / DSM 8903)	Remaining	27	37	36	18	3	8	6	0
	Carboxydotherrmus hydrogenoformans (strain Z-2901 / DSM 6008)	Exclusionary	45	25	11	10	0	0	0	0
	Clostridium acetobutylicum (strain DSM 792 / JCM 1419 / LMG 5710 / ATCC 824 / VKM B-1787)	Remaining	30	36	20	18	22	22	21	3
	Clostridium beijerinckii (strain ATCC 51743 / NCIMB 8052)	Exclusionary	51	43	33	23	28	25	16	0
	Clostridium botulinum (strain ATCC 19397 / Type A)	Remaining	42	42	32	19	0	0	0	0
	Clostridium difficile (strain 630)	Exclusionary	46	35	32	11	1	0	0	0
	Clostridium kluyveri (strain ATCC 8527 / DSM 555 / NCIMB 10680)	Remaining	23	33	20	15	15	23	12	0
	Clostridium novyi (strain NT)	Remaining	9	14	15	9	15	12	12	0
	Clostridium perfringens (strain ATCC 13124 / NCTC 8237 / Type A)	Exclusionary	50	48	18	13	5	6	2	0
	Clostridium phytofermentans (strain ATCC 700394 / DSM 18823 / ISDg)	Remaining	13	21	25	23	24	24	32	5
	Clostridium tetani (strain Massachusetts / E88)	Even-bias	24	21	21	10	22	15	16	0
	Clostridium thermocellum (strain ATCC 27405 / DSM 1237)	Remaining	37	39	40	22	18	18	17	0
	Desulfotomaculum reducens (strain MI-1)	Exclusionary	42	30	32	10	28	14	3	0
	Moorella thermoacetica (strain ATCC 39073)	Exclusionary	44	25	11	5	16	6	6	0
	Mycoplasma mycoides SC (subsp. mycoides, strain PG1)	Exclusionary	10	11	1	0	23	4	0	0
	Mycoplasma penetrans (strain HF-2)	Exclusionary	19	10	1	1	14	3	0	0

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Exported protein signal peptides (secreted proteins and lipoproteins)

	Species	Number of secreted protein signal peptides			Number of lipoprotein signal peptides		
		0 Cys	1 Cys	2 Cys	0 Cys	1 Cys	2 Cys
Gram-negative Bacteria	Acidiphilium cryptum (strain JF-5)	179	61	6	35	7	1
	Agrobacterium tumefaciens (strain C58 / ATCC 33970)	354	89	15	34	8	0
	Azorhizobium caulinodans (strain ATCC 43989 / DSM 5975 / ORS 571)	257	78	18	19	6	0
	Bartonella bacilliformis (strain ATCC 35685 / KC583)	32	21	5	6	7	1
	Bartonella henselae (strain Houston 1 / ATCC 49882)	47	23	10	17	4	2
	Bartonella quintana (strain Toulouse)	42	16	7	14	6	2
	Bartonella tribocorum CIP 105476	54	23	10	15	11	1
	Bradyrhizobium japonicum (strain USDA 110)	497	171	41	33	10	1
	Brucella abortus (strain 2308)	172	54	11	27	8	0
	Brucella canis ATCC 23365	191	52	12	30	9	0
	Brucella melitensis (biovar 1, strain NCTC 10094 / ATCC 23456 / 16M)	125	42	10	18	5	0
	Brucella suis ATCC 23445	187	53	12	27	8	0
	Caulobacter crescentus (strain CB15 / ATCC 19089)	279	86	4	58	10	0
	Dinoroseobacter shibae DFL 12	268	88	16	40	11	0
	Ehrlichia canis (strain Jake)	18	19	4	6	6	0
	Ehrlichia chaffeensis (strain Arkansas)	17	13	4	2	6	1
	Ehrlichia ruminantium (strain Gardel)	16	14	3	4	3	1
	Erythrobacter litoralis (strain HTCC2594)	198	49	9	75	18	1
	Gluconacetobacter diazotrophicus PAI 5	162	68	21	28	11	1
	Gluconobacter oxydans (strain 621H)	142	79	18	39	18	3
	Granulobacter thebesdensis (strain ATCC BAA-1260 / CGDNIH1)	95	39	13	18	11	2
	Hyphomonas neptunium (strain ATCC 15444)	212	91	12	152	30	4
	Jannaschia sp. (strain CCS1)	312	89	27	42	8	1
	Magnetospirillum magneticum (strain AMB-1 / ATCC 700264)	228	58	15	54	11	2
	Maricaulis maris (strain MCS10)	267	72	20	96	20	4
	Mesorhizobium sp. (strain BNC1)	307	92	14	36	9	3
	Methylobacterium extorquens PA1	368	99	14	27	11	0
	Neorickettsia sennetsu (strain Miyayama)	18	11	5	2	1	1
	Nitrobacter hamburgensis (strain X14 / DSM 10229)	287	85	32	27	12	1
	Nitrobacter winogradskyi (strain Nb-255 / ATCC 25391)	204	63	16	23	8	0
	Novosphingobium aromaticivorans (strain DSM 12444)	276	76	18	67	9	3
	Orientia tsutsugamushi (strain Boryong)	28	10	1	4	0	0
	Paracoccus denitrificans (strain Pd 1222)	346	85	17	50	7	0
	Parvibaculum lavamentivorans (strain DS-1 / DSM 13023 / NCIMB 13966)	196	66	8	40	11	1
	Pelagibacter ubique (strain HTCC1062)	63	7	0	15	0	1
	Rhizobium etli (strain CFN 42 / ATCC 51251)	364	94	10	45	7	2
	Rhizobium leguminosarum bv. viciae (strain 3841)	458	118	18	49	5	0
	Rhizobium loti (strain MAFF303099)	382	107	17	38	10	2
	Rhizobium meliloti (strain 1021)	410	90	20	38	6	0
	Rhodobacter sphaeroides (strain ATCC 17023 / 2.4.1 / NCIB 8253 / DSM 158)	270	63	12	41	14	1
	Rhodopseudomonas palustris (strain ATCC BAA-98 / CGA009)	350	93	23	34	11	0
	Rhodospirillum rubrum (strain ATCC 11170 / NCIB 8255)	195	58	13	40	14	0
	Roseobacter denitrificans (strain OCh 114 / ATCC 33942)	254	93	18	33	11	0
	Silicibacter pomeroyi (strain ATCC 700808 / DSS-3 / DSM 15171)	263	65	11	38	11	0
	Silicibacter sp. (strain TM1040)	224	74	20	41	9	0
	Sinorhizobium medicae (strain WSM419)	409	117	24	43	13	3
	Sphingomonas wittichii (strain RW1 / DSM 6014 / JCM 10273)	410	99	15	61	6	1
	Sphingopyxis alaskensis	264	63	13	83	8	1
	Wolbachia pipientis wMel	18	12	1	3	3	0
	Wolbachia sp. (subsp. Brugia malayi, strain TRS)	12	11	1	5	0	0
	Xanthobacter sp.	334	69	17	30	6	1
	Zymomonas mobilis (strain CP4 / ZM4 / ATCC 31821)	77	44	9	20	3	2
Alpha proteobacteria							
Aerobic							

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Beta Proteobacteria	Anaerobic	Anaplasma marginale (strain St. Maries)	13	19	7	5	4	1
		Anaplasma phagocytophilum (strain HZ)	14	9	7	6	5	1
		Brucella ovis (strain ATCC 25840 / 63/290 / NCTC 10512)	142	52	16	24	6	1
		Ochrobactrum anthropi (strain ATCC 49188 / DSM 6882 / NCTC 12168)	351	89	15	42	11	1
		Rickettsia akari (strain Hartford)	28	14	2	15	2	1
		Rickettsia bellii (strain OSU 85-389)	45	13	2	15	5	0
		Rickettsia canadensis (strain McKiel)	32	5	1	11	2	1
		Rickettsia conorii (strain ATCC VR-613 / Malish 7)	33	9	2	13	3	1
		Rickettsia felis (strain ATCC VR-1525 / URRWXCal2)	38	20	1	16	4	1
		Rickettsia massiliae (strain Mtu5)	26	13	1	15	2	1
		Rickettsia prowazekii (strain Madrid E)	22	13	3	12	2	2
		Rickettsia rickettsii Iowa	30	10	2	13	3	1
		Rickettsia typhi (strain Wilmington / ATCC VR-144)	23	13	1	9	4	1
		Acidovorax avenae (subsp. citrulli, strain AAC00-1)	233	129	40	56	18	3
	Aerobic	Acidovorax sp. (strain JS42)	226	102	17	72	14	3
		Bordetella avium (strain 197N)	189	82	10	50	15	1
		Bordetella bronchiseptica (strain NCTC 13252 / RB50 / ATCC BAA-588)	303	146	30	56	14	4
		Bordetella parapertussis (strain NCTC 13253 / ATCC BAA-587 / 12822)	257	118	24	47	12	3
		Bordetella pertussis (strain ATCC BAA-589 / Tohama I / NCTC 13251)	198	83	19	39	15	0
		Bordetella petrii	250	124	22	50	23	4
		Burkholderia cenocepacia (strain AU 1054)	339	127	42	78	34	11
		Burkholderia cepacia (strain ATCC 53795 / AMMD)	337	159	40	85	43	10
		Burkholderia mallei (strain ATCC 23344)	190	102	23	57	20	4
		Burkholderia multivorans ATCC 17616	296	133	33	73	37	8
		Burkholderia pseudomallei (strain 668)	275	137	33	82	30	7
		Burkholderia thailandensis (strain E264 / ATCC 700388 / DSM 13276 / CIP 106301)	239	112	33	59	21	4
		Burkholderia vietnamiensis (strain R1808 / G4 / LMG 22486) / LMG 22486)	370	170	32	91	39	5
		Burkholderia xenovorans (strain LB400)	352	191	48	75	38	3
		Chromobacterium violaceum (strain IFO 12614 / ATCC 12472 / DSM 30191 / NCIB 9131 / JCM 1249)	244	120	32	58	13	2
		Dechloromonas aromatica (strain RCB)	287	107	18	73	19	3
		Delftia acidovorans SPH-1	320	157	49	88	34	2
		Janthinobacterium sp. (strain Marseille)	234	93	22	69	13	2
		Methylibium petroleiphilum (strain PM1)	285	119	28	63	20	4
		Methylobacillus flagellatus (strain KT / ATCC 51484 / DSM 6875)	167	60	18	47	13	3
		Neisseria gonorrhoeae (strain ATCC 700825 / FA 1090)	82	25	9	51	8	1
		Neisseria meningitidis (serogroup C / , serovar 2a, strain ATCC 700532 / FAM18)	78	32	8	44	12	3
		Nitrosomonas europaea (strain IFO 14298 / ATCC 19718)	106	48	10	46	14	3
		Nitrosomonas eutropha (strain C71)	109	47	10	39	12	0
		Nitrospira multiformis (strain ATCC 25196 / NCIMB 11849)	146	50	13	55	13	1
		Polaromonas naphthalenivorans (strain CJ2)	255	110	33	58	23	5
		Polaromonas sp. (strain JS666 / ATCC BAA-500)	297	129	30	50	17	5
		Polynucleobacter sp. (strain QLV-P1DMWA-1)	138	68	12	29	13	3
		Ralstonia eutropha (strain ATCC 17699 / H16 / DSM 428 / Stanier 337)	359	149	42	81	26	3
		Ralstonia metallidurans (strain CH34 / ATCC 43123)	383	136	35	89	29	6
		Ralstonia solanacearum (strain GMI1000)	276	155	41	73	30	1
		Thiobacillus denitrificans (strain ATCC 25259)	227	59	12	55	10	0
		Verminephrobacter eiseniae (strain EF01-2)	236	115	37	31	8	2
	Anaerobic	Hermineimonas arsenicoxydans (strain ULPAs1)	180	72	18	50	18	3
		Rhodoferrax ferrireducens (strain DSM 15236 / ATCC BAA-621 / T118)	229	103	17	71	14	2

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Delta Proteobacteria	Aerobic	Bdellovibrio bacteriovorus (strain DSM 50701 / NCIB 9529 / HD100 / ATCC 15356)	556	116	34	171	44	9
		Geobacter uraniumreducens (strain Rf4)	230	168	49	49	23	5
		Lawsonia intracellularis (strain PHE/MN1-00)	30	16	6	11	8	4
		Myxococcus xanthus (strain DK 1622)	382	303	91	33	7	2
		Sorangium cellulosum (strain So ce56)	371	207	86	417	192	28
	Anaerobic	Anaeromyxobacter dehalogenans (strain 2CP-C)	349	93	11	150	24	6
		Desulfococcus oleovorans Hxd3	91	88	33	41	21	13
		Desulfotalea psychrophila (strain LSV54 / DSM 12343)	86	91	33	0	1	0
		Desulfovibrio desulfuricans (strain G20)	81	63	27	28	16	3
		Desulfovibrio vulgaris (strain Hildenborough / ATCC 29579 / NCIMB 8303)	86	73	38	0	1	0
		Geobacter metallireducens (strain GS-15 / ATCC 53774 / DSM 7210)	209	133	39	0	1	0
		Geobacter sulfurreducens (strain ATCC 51573 / DSM 12127 / PCA)	201	125	40	0	1	0
		Pelobacter carbinolicus (strain DSM 2380 / Gra Bd 1)	90	60	27	42	24	13
		Pelobacter propionicus (strain DSM 2379)	128	92	34	40	31	5
		Syntrophobacter fumaroxidans (strain DSM 10017 / MPOB)	155	110	33	28	26	10
		Syntrophus aciditrophicus (strain SB)	80	39	18	26	14	5
		Arcobacter butzleri (strain RM4018)	121	24	4	46	2	0
Epsilon Proteobacteria	Aerobic	Campylobacter concisus (strain 13826)	102	54	8	42	16	0
		Campylobacter curvus (strain 525.92)	88	49	9	40	10	1
		Campylobacter fetus (subsp. fetus, strain 82-40)	65	38	10	33	4	1
		Campylobacter hominis (strain ATCC BAA-381 / LMG 19568 / NCTC 13146 / CH001A)	56	26	4	28	9	1
		Campylobacter jejuni (subsp. doylei, strain ATCC BAA-1458 / RM4099 / 269.97)	48	19	4	22	3	1
		Helicobacter acinonychis (strain Sheeba)	63	36	6	23	7	2
		Helicobacter hepaticus (strain 3B1 / ATCC 51449)	50	33	14	26	19	5
		Helicobacter pylori (strain 26695 / ATCC 700392)	83	38	8	24	9	0
		Nitratiruptor sp. (strain SB155-2)	91	22	3	30	1	1
		Sulfurimonas denitrificans (strain ATCC33889/DSM 1251)	125	18	5	40	0	1
		Sulfurovum sp. (strain NBC37-1)	236	33	3	66	3	1
	Anaerobic	Wolinella succinogenes (strain DSMZ 1740)	99	24	2	38	4	0
Gamma Proteobacteria	Aerobic	Acinetobacter baumannii (strain ATCC 17978)	32	12	3	20	5	3
		Acinetobacter sp. (strain ADP1)	145	70	19	59	24	7
		Actinobacillus pleuropneumoniae (serovar5b, strain L20)	118	70	6	1	0	0
		Actinobacillus succinogenes (strain ATCC 55618 / 130Z)	129	43	8	40	5	4
		Aeromonas hydrophila (subsp. hydrophila, strain ATCC 7966 / NCIB 9240)	226	90	29	70	28	1
		Aeromonas salmonicida (strain A449)	235	142	35	37	14	3
		Alcanivorax borkumensis (strain SK2 / ATCC 700651 / DSM 11573)	166	55	18	80	18	1
		Baumannia cicadellinicola coagulata (subsp. Homalodisca)	7	4	0	5	1	0
		Blochmannia floridanus	1	0	0	2	0	0
		Blochmannia pennsylvanicus (strain BPEN)	1	3	0	2	2	0
		Buchnera aphidicola (subsp. Acyrthosiphon pisum, strain Tokyo 1998)	9	0	0	0	2	0
		Buchnera aphidicola cedri (subsp. Cinara)	3	0	0	0	0	0
		Chromohalobacter salexigens (strain DSM 3043 / ATCC BAA-138 / NCIMB 13768)	149	62	16	38	14	1
		Citrobacter koseri (strain ATCC BAA-895 / CDC 4225-83 / SGSC4696)	250	97	9	59	17	1
		Colwellia psychrethraea	327	132	22	143	36	5
		Coxiella burnetii (strain Dugway 5J108-111)	72	32	8	25	4	2
		Dichelobacter nodosus (strain VCS1703A)	55	16	4	25	10	3
		Enterobacter sakazakii (strain ATCC BAA-894)	221	84	21	63	17	2
		Erwinia carotovora (subsp. atroseptica, strain ATCC BAA-672 / SCRI 1043)	256	102	17	71	30	1

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Escherichia coli (strain K12)	254	89	31	74	30	2
Francisella tularensis (subsp. tularensis, strain FSC 198)	64	29	5	36	12	0
Haemophilus ducreyi (strain ATCC 700724 / 35000HP)	70	39	9	2	1	0
Haemophilus influenzae (strain ATCC 51907 / DSM 11121 / KW20 / Rd)	74	45	4	15	0	1
Haemophilus somnus (strain 129Pt)	69	19	2	31	4	1
Hahella chejuensis (strain KCTC 2396)	368	205	62	131	47	6
Halorhodospira halophila (strain DSM 244 / SL1)	118	35	10	46	10	2
Idiomarina loihiensis (strain L2-TR / DSM 15497 / ATCC BAA-735)	216	57	5	82	6	1
Klebsiella pneumoniae (subsp. pneumoniae, strain ATCC 700721 / MGH 78578)	272	83	13	61	22	1
Legionella pneumophila (strain Paris)	168	85	21	31	25	2
Marinobacter aquaeolei (strain DSM 11845 / ATCC 700491 / VT8) / VT8)	250	67	21	101	16	1
Marinomonas sp. MWYL1	215	85	21	63	23	1
Methylococcus capsulatus (strain Bath / NCIMB 11132)	132	57	18	40	14	2
Nitrosococcus oceanii (strain ATCC 19707 / NCIMB 11848)	153	51	7	41	20	4
Pasteurella multocida (strain Pm70)	102	35	8	49	11	2
Photobacterium profundum (strain SS9)	319	172	36	1	0	0
Photorhabdus luminescens laumondii (strain TT01)	172	51	9	57	16	1
Pseudoalteromonas atlantica (strain T6c / BAA-1087)	282	141	49	132	30	8
Pseudoalteromonas haloplanktis (strain TAC 125)	217	100	16	106	33	3
Pseudomonas aeruginosa (strain LMG 12228 / ATCC 15692 / PRS 101 / 1C / PAO1)	325	177	45	118	39	4
Pseudomonas entomophila (strain L48)	296	136	38	100	32	8
Pseudomonas fluorescens (strain Pf-5 / ATCC BAA-477)	315	166	59	118	37	6
Pseudomonas mendocina (strain ymp)	295	128	37	97	25	6
Pseudomonas putida (strain F1 / ATCC 700007)	299	133	36	91	30	6
Pseudomonas stutzeri (strain A1501)	196	80	20	63	17	6
Pseudomonas syringae (pathovar phaseolicola, strain 1448A / Race 6)	235	113	23	84	25	6
Psychrobacter arcticum (strain 273-4)	71	21	3	56	4	2
Psychrobacter cryohalolentis (strain K5)	92	27	7	80	12	1
Psychrobacter sp. (strain PRwf-1)	91	25	10	74	12	1
Saccharophagus degradans (strain 2-40 / ATCC 43961 / DSM 17024)	264	161	66	129	66	13
Salmonella arizonae (strain ATCC BAA-731 / CDC346-86 / RSK2980)	219	65	9	63	21	1
Salmonella choleraesuis (strain SC-B67)	258	84	19	66	22	3
Salmonella paratyphi A (strain SARB42 / ATCC 9150)	261	76	16	62	23	2
Salmonella paratyphi B (strain ATCC BAA-1250 / SPB7)	270	80	15	60	26	3
Salmonella typhi (strain ATCC 700931 / Ty2)	268	86	20	65	25	2
Salmonella typhimurium (strain ATCC 700720 / SGSC1412 / LT2)	291	79	21	68	21	4
Serratia proteamaculans (strain 568)	290	109	28	78	21	0
Shewanella amazonensis (strain ATCC BAA-1098 / SB2B)	313	103	27	114	24	4
Shewanella baltica (strain OS155 / ATCC BAA-1091)	274	136	28	85	41	6
Shewanella denitrificans (strain OS217 / ATCC BAA-1090 / DSM 15013)	257	117	30	95	23	10
Shewanella frigidimarina (strain NCIMB 400)	277	115	34	115	28	3
Shewanella loihica (strain BAA-1088 / PV-4)	313	115	27	123	27	5
Shewanella oneidensis (strain MR-1)	270	112	29	96	34	2
Shewanella pealeana ATCC 700345	328	130	29	140	28	5
Shewanella putrefaciens (strain CN-32/ATCC BAA-453)	252	114	25	90	33	7
Shewanella sediminis HAW-EB3	332	141	30	140	39	6
Shewanella sp. (strain ANA-3)	292	149	34	110	33	7
Shigella boydii (serovar 4, strain Sb227)	167	64	17	56	15	1
Shigella dysenteriae (serovar 1, strain Sd97 / Sd197)	163	53	14	52	17	1
Shigella flexneri (serovar 2a, strain 2457T/ATCC 700930)	178	59	15	54	23	2
Shigella sonnei (strain Ss046)	199	66	19	57	20	3
Sodalis glossinidius okutanii (subsp. Calyptogenia okutanii, strain HA)	91	26	5	32	9	0
Vesicomyxosocius okutanii (subsp. Calyptogenia okutanii, strain HA)	33	9	0	12	1	0
Vibrio cholerae (strain ATCC 39541 / O395)	204	74	13	62	16	1

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Gram-positive Bacteria	Actinobacteria	Aerobic	Vibrio fischeri (strain ATCC 700601 / ES114)	241	84	13	121	17	3
			Vibrio harveyi (strain ATCC BAA-1116 / BB120)	322	114	16	124	22	0
			Vibrio parahaemolyticus (serovar O3:K6, strain RIMD 2210633)	316	93	21	129	21	1
			Vibrio vulnificus (strain CMCP6)	265	82	13	90	12	0
			Wigglesworthia glossinidia brevipalpis	4	1	0	6	1	0
			Xanthomonas axonopodis citri (strain 306)	247	138	47	79	35	7
			Xanthomonas campestris (strain ATCC 33913)	249	141	47	80	33	10
			Xanthomonas oryzae (pathovar oryzae, MAFF 311018)	213	132	39	66	34	5
			Xylella fastidiosa (strain 9a5c)	91	39	9	33	13	2
			Xylella fastidiosa (strain Temecula1 / ATCC 700964)	80	38	10	29	16	5
			Yersinia enterocolitica (serovar O:8 / strain 8081)	227	79	11	66	16	1
			Yersinia pestis (biovar Antiqua Antiqua, strain Antiqua)	243	78	15	54	21	0
			Yersinia pseudotuberculosis (serovar I, strain IP32953)	253	83	15	59	20	1
			Alkalilimnicola ehrlichei (strain MLHE-1)	153	54	10	50	7	0
			Carsonella ruddii (strain PV)	0	0	0	1	0	0
			Mannheimia succiniciproducens (strain MBEL55E)	95	24	6	55	3	2
			Psychromonas ingrahamii (strain 37)	163	79	13	65	10	2
			Ruthia magnifica magnifica (subsp. Calyptogena)	38	7	2	13	1	0
			Thiomicrospira crunogena (strain XCL-2)	144	30	3	52	6	0
			Acidothermus cellulolyticus (strain ATCC 43068 / 11B)	68	15	1	25	8	0
	Actinobacteria	Aerobic	Arthrobacter aurescens (strain TC1)	103	38	10	78	7	3
			Clavibacter michiganensis (subsp. michiganensis, strain NCPPB 382)	95	33	3	63	3	0
			Corynebacterium diphtheriae (biovar gravis, strain NCTC 13129 / ATCC 700971)	55	24	4	28	14	2
			Corynebacterium efficiens (strain DSM 44549 / NBRC 100395 / JCM 11189 / YS-314 / AJ 12310)	73	24	2	34	2	0
			Corynebacterium glutamicum (strain R)	72	19	1	72	9	0
			Corynebacterium jeikeium (strain K411)	74	20	1	30	7	1
			Frankia alni (strain ACN14a)	118	30	3	34	4	2
			Kineococcus radiotolerans SRS30216	138	40	5	60	5	0
			Leifsonia xyli xyli (strain CTCB07)	52	18	0	29	1	0
			Mycobacterium avium (strain 104)	135	47	11	51	14	1
			Mycobacterium bovis (strain ATCC BAA-935)	102	25	13	43	16	3
			Mycobacterium gilvum PYR-GCK	163	60	12	77	15	1
			Mycobacterium leprae (strain TN)	40	23	5	12	9	0
			Mycobacterium paratuberculosis (strain ATCC BAA-968 / K-10)	116	50	17	43	15	3
			Mycobacterium smegmatis (strain ATCC 700084 / mc(2 / mc(2)155)	187	54	11	116	17	6
			Mycobacterium tuberculosis (strain ATCC 25177 / H37Ra)	102	30	13	45	16	3
			Mycobacterium ulcerans (strain Agy99)	120	36	6	53	17	2
			Mycobacterium vanbaalenii (strain DSM 7251 / PYR-1)	207	74	14	77	32	2
			Nocardia farcinica (strain IFM 10152)	139	55	8	71	18	0
			Nocardioideis sp. (strain BAA-499 / JS614)	202	44	4	85	7	1
			Renibacterium salmoninarum (strain ATCC 33209)	69	25	4	42	4	1
			Rhodococcus sp. (strain RHA1)	176	64	13	116	37	0
			Rubrobacter xylanophilus (strain DSM 9941 / NBRC 16129)	64	19	8	28	7	2
			Saccharopolyspora erythraea (strain NRRL 23338)	204	66	12	69	10	2
			Salinispora arenicola CNS-205	162	20	5	63	4	0
			Salinispora tropica CNB-440	124	23	6	56	3	0
			Streptomyces avermitilis (strain DSM 46492 / NRRL 8165 / NCIMB 12804 / ATCC 31267 / JCM 5070)	296	91	14	88	21	3
			Streptomyces coelicolor (strain ATCC BAA-471 / M145 / A3(2))	326	84	14	116	26	3
			Thermobifida fusca (strain YX)	75	30	4	39	11	2
			Tropheryma whipplei (strain Twist)	13	7	2	7	1	0
Gram-positive Bacteria	Actinobacteria	Anaerobic	Bifidobacterium adolescentis (strain ATCC 15703 / DSM 20083)	23	13	5	13	4	1
			Bifidobacterium longum (strain NCC 2705)	16	12	6	18	4	4
			Propionibacterium acnes (strain DSM 16379 / KPA171202)	46	20	5	36	11	3

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Firmicutes	Aerobic	Acholeplasma laidlawii (strain PG-8A)	23	6	0	57	6	0
		Bacillus amyloliquefaciens (strain FZB42)	102	49	13	56	20	7
		Bacillus anthracis (strain Ames, isolate Porton)	113	49	7	89	30	6
		Bacillus cereus (strain ATCC 10987)	117	48	15	89	41	6
		Bacillus clausii (strain KSM-K16)	78	33	6	102	39	3
		Bacillus halodurans (strain C-125 / ATCC BAA-125 / JCM 9153 / FERM 7344 / DSM 18197)	103	33	4	97	22	4
		Bacillus licheniformis (strain DSM 13 / ATCC 14580, sub strain Goettingen)	123	43	12	73	19	3
		Bacillus pumilus (strain SAFR-032)	107	32	9	57	23	5
		Bacillus subtilis (strain 168)	122	45	9	63	24	8
		Bacillus thuringiensis (strain Al Hakam)	107	45	19	81	29	8
		Bacillus weihenstephanensis KBAB4	98	52	13	99	42	3
		Desulfotobacterium hafniense (strain Y51)	107	42	4	112	24	5
		Enterococcus faecalis (strain V583 / ATCC 700802)	73	22	1	59	8	2
		Geobacillus kaustophilus (strain HTA426)	88	22	3	55	8	6
		Geobacillus thermodenitrificans (strain NG80-2)	70	23	2	56	17	3
		Lactobacillus acidophilus (strain NCFM)	60	13	2	29	2	1
		Lactobacillus brevis (strain ATCC 367 / JCM 1170)	82	17	1	16	3	0
		Lactobacillus casei (strain ATCC 334)	82	9	1	32	2	0
		Lactobacillus delbrueckii (subsp. bulgaricus, strain ATCC BAA-365)	52	8	1	13	4	0
		Lactobacillus gasseri (strain ATCC 33323 / DSM 20243)	28	8	1	24	3	0
		Lactobacillus helveticus (strain DPC 4571)	46	9	0	17	1	0
		Lactobacillus johnsonii (strain NCC 533)	32	7	1	27	3	0
		Lactobacillus plantarum (strain WCFS1 / ATCC BAA-793 / NCIMB 8826)	70	18	0	41	3	0
		Lactobacillus reuteri (strain ATCC 23272 / DSM 20016 / F275)	43	14	1	10	4	0
		Lactobacillus sakei (subsp. sakei, strain 23K)	107	14	2	21	3	0
		Lactobacillus salivarius (subsp. salivarius, strain UCC118)	42	8	4	15	4	0
		Lactococcus lactis (subsp. lactis, strain IL1403)	75	9	1	28	2	0
		Leuconostoc mesenteroides (subsp. mesenteroides, strain ATCC 8293 / NCDO 523)	78	6	1	2	0	0
		Listeria innocua (serovar 6a, strain CLIP 11262)	68	20	4	53	7	0
		Listeria monocytogenes (serovar 1/2a, strain EGD-e / ATCC BAA-679)	67	19	4	54	6	1
		Listeria welshimeri (serovar 6b, strain ATCC 35897 / DSM 20650 / SLCC5334)	66	18	1	55	6	1
		Mesoplasma florum (strain ATCC 33453 / NCTC 11704 / L1 / NBRC 100688)	14	11	0	12	0	0
		Mycoplasma agalactiae	44	15	2	22	2	0
		Mycoplasma capricolum (subsp. capricolum, strain California kid / ATCC 27343 / NCTC 10154)	26	8	1	29	1	0
		Mycoplasma gallisepticum (strain R(low))	16	15	0	17	2	0
		Mycoplasma genitalium (strain ATCC 33530 / NCTC 10195 / G-37)	6	5	0	13	4	0
		Mycoplasma hyopneumoniae (strain J / ATCC 25934 / NCTC 10110)	32	7	1	28	2	0
		Mycoplasma mobile (strain 163K / NCTC 11711 / ATCC 43663)	28	5	0	6	0	0
		Mycoplasma pneumoniae (strain M129 / ATCC 29342)	11	8	0	20	3	0
		Mycoplasma pulmonis (strain UAB CTIP)	27	10	1	35	0	0
		Mycoplasma synoviae (strain 53)	15	6	0	15	0	0
		Oceanobacillus iheyensis (strain KCTC 3954 / DSM 14371 / JCM 11309 / HTE831)	101	13	0	99	17	3
		Oenococcus oeni (strain BAA-331 / PSU-1)	57	6	1	3	0	0
		Onion yellows phytoplasma (strain OY-M)	21	9	0	1	0	0
		Pediococcus pentosaceus (strain ATCC25745 / 183-1w)	39	6	0	18	2	0
		Pelotomaculum thermopropionicum (strain DSM 13744 / JCM 10971 / SI)	74	23	8	18	13	1
		Staphylococcus aureus (strain Mu50 / ATCC 700699)	76	12	1	45	4	2
		Staphylococcus epidermidis (strain ATCC 12228)	57	10	0	38	7	1
		Staphylococcus haemolyticus (strain JCSC1435)	42	10	1	33	6	0
		Staphylococcus saprophyticus (subsp. saprophyticus, strain ATCC 15305 / DSM 20229)	43	9	2	28	6	4

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Anaerobic	Streptococcus agalactiae Ia (serovar Ia, strain A909 / CDC SS700 / ATCC 27591)	59	18	3	21	8	2
	Streptococcus agalactiae III (serovar III, strain NEM316)	59	21	2	22	6	2
	Streptococcus agalactiae V (serovar V, strain 2603 V/R / ATCC BAA-611)	55	18	2	18	9	1
	Streptococcus gordonii (strain Challis / ATCC 35105 / CH1 / DL1 / V288)	55	12	0	41	12	2
	Streptococcus mutans (serovar c, strain ATCC 700610 / UA159)	44	12	3	22	2	0
	Streptococcus pneumoniae (serovar 2, strain D39 / NCTC 7466)	32	5	1	22	7	1
	Streptococcus pneumoniae (strain ATCC BAA-255 / R6)	35	5	1	21	7	1
	Streptococcus pneumoniae (strain TIGR4 / ATCC BAA-334)	36	4	2	23	7	1
	Streptococcus pyogenes M1 (serovar M1, strain MGAS5005 / ATCC BAA-947)	49	16	3	20	6	0
	Streptococcus sanguinis (strain SK36)	66	11	0	39	7	2
	Streptococcus suis (strain 05ZYH33)	39	10	2	26	5	2
	Streptococcus thermophilus (strain ATCC BAA-491 / LMD-9)	40	2	1	14	4	1
	Symbiobacterium thermophilum (strain IAM 14863 / T)	89	73	12	0	0	0
	Ureaplasma parvum (serovar 3, strain ATCC 700970)	20	6	3	20	1	1
	Alkaliphilus metalliredigens (strain QYMF)	103	121	19	0	0	0
	Alkaliphilus oremlandii (strain OhILAs)	83	72	16	17	2	0
	Caldicellulosiruptor saccharolyticus (strain ATCC 43494 / DSM 8903)	109	39	2	17	6	0
	Carboxydotherrnus hydrogenoformans (strain Z-2901 / DSM 6008)	76	31	2	0	0	0
	Clostridium acetobutylicum (strain DSM 792 / JCM 1419 / LMG 5710 / ATCC 824 / VKM B-1787)	117	39	9	61	19	4
	Clostridium beijerinckii (strain ATCC 51743 / NCIMB 8052)	120	47	5	65	22	1
	Clostridium botulinum (strain ATCC 19397 / Type A)	77	64	17	0	0	0
	Clostridium difficile (strain 630)	66	53	19	1	0	0
	Clostridium kluyveri (strain ATCC 8527 / DSM 555 / NCIMB 10680)	80	28	8	39	11	5
	Clostridium novyi (strain NT)	51	18	3	27	19	7
	Clostridium perfringens (strain ATCC 13124 / NCTC 8237 / Type A)	77	63	11	11	1	1
	Clostridium phytofermentans (strain ATCC 700394 / DSM 18823 / ISDg)	83	41	14	108	42	20
	Clostridium tetani (strain Massachusetts / E88)	62	30	4	49	13	2
	Clostridium thermocellum (strain ATCC 27405 / DSM 1237)	113	71	13	55	18	3
	Desulfotomaculum reducens (strain MI-1)	92	38	4	44	6	3
	Moorella thermoacetica (strain ATCC 39073)	78	15	2	25	3	0
	Mycoplasma mycoides SC (subsp. mycoides, strain PG1)	18	6	0	26	1	0
	Mycoplasma penetrans (strain HF-2)	24	7	0	17	0	0

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Transmembrane protein loops (cytoplasmic and extra-cytoplasmic)

Gram-negative Bacteria	Alpha proteobacteria	Species	Number of extra-cytoplasmic TM-protein loops				Number of cytoplasmic TM-protein loops			
			0 Cys	1 Cys	2 Cys	3 Cys	0 Cys	1 Cys	2 Cys	3 Cys
Aerobic		Acidiphilium cryptum (strain JF-5)	526	69	38	17	413	117	59	31
		Agrobacterium tumefaciens (strain C58 / ATCC 33970)	840	59	70	9	719	160	62	19
		Azorhizobium caulinodans (strain ATCC 43989 / DSM 5975 / ORS 571)	806	87	80	15	601	220	90	61
		Bartonella bacilliformis (strain ATCC 35685 / KC583)	154	21	8	2	106	57	18	3
		Bartonella henselae (strain Houston 1 / ATCC 49882)	189	34	17	5	140	59	27	14
		Bartonella quintana (strain Toulouse)	174	32	16	3	115	66	34	11
		Bartonella tribocorum CIP 105476	238	36	17	3	174	71	28	13
		Bradyrhizobium japonicum (strain USDA 110)	1211	144	126	29	916	311	154	83
		Brucella abortus (strain 2308)	450	43	24	6	358	109	37	12
		Brucella canis ATCC 23365	493	42	25	8	382	115	38	18
		Brucella melitensis (biovar 1, strain NCTC 10094 / ATCC 23456 / 16M)	488	48	30	10	373	123	44	17
		Brucella suis ATCC 23445	492	46	24	8	377	115	40	21
		Caulobacter crescentus (strain CB15 / ATCC 19089)	551	69	49	10	465	118	55	29
		Dinoroseobacter shibae DFL 12	609	34	55	10	502	131	40	18
		Ehrlichia canis (strain Jake)	128	37	13	6	61	35	42	18
		Ehrlichia chaffeensis (strain Arkansas)	114	29	20	5	64	36	33	12
		Ehrlichia ruminantium (strain Gardel)	145	37	27	9	77	50	37	23
		Erythrobacter litoralis (strain HTCC2594)	442	26	34	5	369	69	37	21
		Gluconacetobacter diazotrophicus PAI 5	472	91	34	16	334	159	66	31
		Gluconobacter oxydans (strain 621H)	334	68	32	15	254	104	47	33
		Granulobacter betshensis (strain ATCC BAA-1260 / CGDNIH1)	353	64	36	7	241	105	68	31
		Hyphomonas neptunium (strain ATCC 15444)	526	36	49	11	440	111	49	15
		Jannaschia sp. (strain CCS1)	719	51	69	10	630	134	48	18
		Magnetospirillum magneticum (strain AMB-1 / ATCC 700264)	620	79	65	19	442	142	99	62
		Maricaulis maris (strain MCS10)	567	27	46	3	475	96	43	15
		Mesorhizobium sp. (strain BNC1)	678	52	46	8	577	121	41	29
		Methylobacterium extorquens PA1	713	71	62	13	594	147	82	25
		Neorickettsia sennetsu (strain Miyayama)	102	30	17	3	54	30	37	13
		Nitrobacter hamburgensis (strain X14 / DSM 10229)	587	61	41	16	450	144	58	33
		Nitrobacter winogradskyi (strain Nb-255 / ATCC 25391)	406	26	39	6	318	102	32	16
		Novosphingobium aromaticivorans (strain DSM 12444)	479	44	40	8	382	105	45	17
		Orientia tsutsugamushi (strain Boryong)	101	41	20	3	67	56	23	17
		Paracoccus denitrificans (strain Pd 1222)	730	60	60	11	596	149	68	18
		Parvibaculum lavamentivorans (strain DS-1 / DSM 13023 / NCIMB 13966)	537	51	48	3	453	109	35	23
		Pelagibacter ubique (strain HTCC1062)	207	16	18	6	186	38	14	6
		Rhizobium etli (strain CFN 42 / ATCC 51251)	887	97	80	18	737	205	90	36
		Rhizobium leguminosarum bv. viciae (strain 3841)	1044	108	87	18	897	199	99	36
		Rhizobium loti (strain MAFF303099)	1012	97	94	17	809	205	121	47
		Rhizobium meliloti (strain 1021)	949	69	57	13	757	203	60	34
		Rhodobacter sphaeroides (strain ATCC 17023 / 2.4.1 / NCIB 8253 / DSM 158)	650	46	56	5	540	124	45	20
		Rhodopseudomonas palustris (strain ATCC BAA-98 / CGA009)	758	88	64	10	608	165	90	27
		Rhodospirillum rubrum (strain ATCC 11170 / NCIB 8255)	634	67	47	15	496	133	68	35
		Roseobacter denitrificans (strain OCh 114 / ATCC 33942)	665	45	49	8	507	160	53	23
		Silicibacter pomeroyi (strain ATCC 700808 / DSS-3 / DSM 15171)	647	52	66	19	532	148	63	19
		Silicibacter sp. (strain TM1040)	565	49	38	14	452	120	47	24
		Sinorhizobium medicae (strain WSM419)	935	77	70	11	777	204	65	31
		Sphingomonas wittichii (strain RW1 / DSM 6014 / JCM 10273)	640	59	67	17	514	146	83	33
		Sphingopyxis alaskensis	463	50	41	11	395	78	49	23
		Wolbachia pipientis wMel	153	31	9	6	79	47	37	13
		Wolbachia sp. (subsp. Brugia malayi, strain TRS)	110	25	12	3	64	36	26	8
		Xanthobacter sp.	705	76	54	9	569	146	78	25
		Zymomonas mobilis (strain CP4 / ZM4 / ATCC 31821)	236	38	26	2	174	71	30	12

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Beta Proteobacteria	Anaerobic	Anaplasma marginale (strain St. Maries)	107	48	24	14	57	45	42	28
		Anaplasma phagocytophilum (strain HZ)	95	35	24	8	51	30	33	21
		Brucella ovis (strain ATCC 25840 / 63/290 / NCTC 10512)	447	42	29	7	336	114	41	21
		Ochrobactrum anthropi (strain ATCC 49188 / DSM 6882 / NCTC 12168)	799	60	48	6	641	169	61	20
		Rickettsia akari (strain Hartford)	159	27	18	5	109	58	32	6
		Rickettsia bellii (strain OSU 85-389)	180	31	16	9	125	67	27	20
		Rickettsia canadensis (strain McKiel)	162	24	15	3	106	62	29	4
		Rickettsia conorii (strain ATCC VR-613 / Malish 7)	181	44	17	7	137	65	29	16
		Rickettsia felis (strain ATCC VR-1525 / URRWXC2)	201	34	18	9	145	72	29	14
		Rickettsia massiliae (strain Mtu5)	165	35	20	9	116	63	27	14
		Rickettsia prowazekii (strain Madrid E)	134	34	20	4	105	49	30	4
	Aerobic	Rickettsia rickettsii Iowa	169	37	13	4	121	63	28	11
		Rickettsia typhi (strain Wilmington / ATCC VR-144)	138	37	11	4	95	52	22	11
		Acidovorax avenae (subsp. citrulli, strain AAC00-1)	701	81	72	15	542	169	84	34
		Acidovorax sp. (strain JS42)	573	58	51	10	452	122	58	31
		Bordetella avium (strain 197N)	519	53	44	4	380	123	55	32
		Bordetella bronchiseptica (strain NCTC 13252 / RB50 / ATCC BAA-588)	688	57	44	9	509	159	80	27
		Bordetella parapertussis (strain NCTC 13253 / ATCC BAA-587 / 12822)	613	49	44	8	448	153	77	22
		Bordetella pertussis (strain ATCC BAA-589 / Tohama I / NCTC 13251)	485	33	28	3	353	106	59	16
		Bordetella petrii	685	81	56	8	521	186	77	28
		Burkholderia cenocepacia (strain AU 1054)	916	91	72	15	670	257	94	53
		Burkholderia cepacia (strain ATCC 53795 / AMMD)	878	113	85	22	656	266	89	47
		Burkholderia mallei (strain ATCC 23344)	651	81	56	17	443	193	91	32
		Burkholderia multivorans ATCC 17616	853	96	69	23	626	239	93	45
		Burkholderia pseudomallei (strain 668)	839	108	80	25	570	262	111	47
		Burkholderia thailandensis (strain E264 / ATCC 700388 / DSM 13276 / CIP 106301)	786	109	95	27	526	264	112	59
		Burkholderia vietnamiensis (strain R1808 / G4 / LMG 22486) / LMG 22486)	951	121	94	16	709	269	126	54
		Burkholderia xenovorans (strain LB400)	1055	144	80	25	777	284	133	64
		Chromobacterium violaceum (strain IFO 12614 / ATCC 12472 / DSM 30191 / NCIB 9131 / JCM 1249)	634	81	40	14	436	175	90	32
		Dechloromonas aromatica (strain RCB)	648	80	58	17	477	149	88	33
		Delftia acidovorans SPH-1	853	113	70	16	616	213	111	52
		Janthinobacterium sp. (strain Marseille)	590	69	39	16	448	143	64	21
		Methylobium petroleiphilum (strain PM1)	651	66	61	15	493	157	63	39
		Methylobacillus flagellatus (strain KT / ATCC 51484 / DSM 6875)	475	34	34	7	351	114	46	20
		Neisseria gonorrhoeae (strain ATCC 700825 / FA 1090)	245	13	14	6	158	66	28	15
		Neisseria meningitidis (serogroup C / , serovar 2a, strain ATCC 700532 / FAM18)	254	22	20	9	186	63	25	16
		Nitrosomonas europaea (strain IFO 14298 / ATCC 19718)	380	41	31	15	294	88	48	18
		Nitrosomonas eutropha (strain C71)	403	34	36	12	313	78	49	25
		Nitrospira multiformis (strain ATCC 25196 / NCIMB 11849)	425	46	43	7	344	85	45	27
		Polaromonas naphthalenivorans (strain CJ2)	634	67	47	15	504	141	64	21
		Polaromonas sp. (strain JS666 / ATCC BAA-500)	766	81	46	15	607	177	71	28
		Polynucleobacter sp. (strain QLW-P1DMWA-1)	325	36	21	6	264	67	34	14
		Ralstonia eutropha (strain ATCC 17699 / H16 / DSM 428 / Stanier 337)	840	104	73	22	633	215	104	40
		Ralstonia metallidurans (strain CH34 / ATCC 43123)	866	114	74	24	651	231	114	45
		Ralstonia solanacearum (strain GMI1000)	694	95	62	26	558	168	91	34
		Thiobacillus denitrificans (strain ATCC 25259)	475	37	25	7	354	85	55	18
		Verminephrobacter eiseniae (strain EF01-2)	597	93	59	16	428	202	74	31
Beta Proteobacteria	Anaerobic	Herminiimonas arsenicoxydans (strain ULPAs1)	519	50	28	13	368	131	53	31
		Rhodoferrax ferrireducens (strain DSM 15236 / ATCC BAA-621 / T118)	649	62	62	21	529	128	78	30

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Delta Proteobacteria	Aerobic	Bdellovibrio bacteriovorus (strain DSM 50701 / NCIB 9529 / HD100 / ATCC 15356)	465	60	59	9	387	113	58	22
		Geobacter uraniumreducens (strain Rf4)	484	99	46	26	367	133	75	37
		Lawsonia intracellularis (strain PHE/MN1-00)	190	50	17	9	156	66	25	15
		Myxococcus xanthus (strain DK 1622)	981	161	160	50	788	293	183	78
		Sorangium cellulosum (strain So ce56)	1011	274	226	94	890	334	256	115
	Anaerobic	Anaeromyxobacter dehalogenans (strain 2CP-C)	670	78	61	26	571	158	77	31
		Desulfococcus oleovorans Hxd3	387	90	44	18	289	130	60	35
		Desulfotalea psychrophila (strain LSv54 / DSM 12343)	468	124	56	25	309	164	98	46
		Desulfovibrio desulfuricans (strain G20)	410	102	44	18	275	137	67	31
		Desulfovibrio vulgaris (strain Hildenborough / ATCC 29579 / NCIMB 8303)	418	102	34	16	255	130	74	46
		Geobacter metallireducens (strain GS-15 / ATCC 53774 / DSM 7210)	431	80	34	26	315	132	67	33
		Geobacter sulfurreducens (strain ATCC 51573 / DSM 12127 / PCA)	459	81	42	21	342	135	75	36
		Pelobacter carbinolicus (strain DSM 2380 / Gra Bd 1)	377	93	41	23	243	142	77	27
		Pelobacter propionicus (strain DSM 2379)	440	96	49	22	267	142	71	54
		Syntrophobacter fumaroxidans (strain DSM 10017 / MPOB)	463	108	58	30	345	151	84	37
		Syntrophus aciditrophicus (strain SB)	342	83	26	17	256	113	58	15
		Arcobacter butzleri (strain RM4018)	398	40	24	4	316	73	50	20
		Campylobacter concisus (strain 13826)	306	33	23	7	215	83	39	13
		Campylobacter curvus (strain 525.92)	303	39	17	7	215	82	41	16
		Campylobacter fetus (subsp. fetus, strain 82-40)	280	28	17	2	198	69	36	13
		Campylobacter hominis (strain ATCC BAA-381 / LMG 19568 / NCTC 13146 / CH001A)	243	54	11	2	185	67	39	17
		Campylobacter jejuni (subsp. doylei, strain ATCC BAA-1458 / RM4099 / 269.97)	247	27	13	1	171	54	38	15
		Helicobacter acinonychis (strain Sheeba)	212	25	10	5	148	59	27	15
		Helicobacter hepaticus (strain 3B1 / ATCC 51449)	251	40	30	9	191	66	43	16
		Helicobacter pylori (strain 26695 / ATCC 700392)	230	24	12	7	163	59	32	9
		Nitratiruptor sp. (strain SB155-2)	292	23	29	5	222	68	31	18
		Sulfurimonas denitrificans (strain ATCC33889/DSM 1251)	326	35	31	9	267	50	47	22
		Sulfurovum sp. (strain NBC37-1)	405	35	30	7	336	57	30	23
Epsilon Proteobacteria	Aerobic	Wolinella succinogenes (strain DSMZ 1740)	377	23	21	7	303	67	30	19
		Acinetobacter baumannii (strain ATCC 17978)	525	52	23	6	396	131	43	19
		Acinetobacter sp. (strain ADP1)	548	43	30	6	402	129	52	22
		Actinobacillus pleuropneumoniae (serovar5b, strain L20)	331	26	15	3	251	76	30	9
		Actinobacillus succinogenes (strain ATCC 55618 / 130Z)	340	29	18	2	260	83	27	14
		Aeromonas hydrophila (subsp. hydrophila, strain ATCC 7966 / NCIB 9240)	709	79	63	16	452	184	112	54
		Aeromonas salmonicida (strain A449)	702	89	53	14	471	181	104	56
		Alcanivorax borkumensis (strain SK2 / ATCC 700651 / DSM 11573)	448	47	36	11	337	109	54	20
		Baumannia cicadellinicola coagulata (subsp. Homalodisca)	55	15	6	4	39	22	10	7
		Blochmannia floridanus	45	30	9	6	37	30	13	8
		Blochmannia pennsylvanicus (strain BPEN)	55	20	8	8	46	23	13	7
		Buchnera aphidicola (subsp. Acyrthosiphon pisum, strain Tokyo 1998)	47	15	3	2	35	17	10	4
		Buchnera aphidicola cedri (subsp. Cinara)	20	6	3	2	12	7	7	3
		Chromohalobacter salexigens (strain DSM 3043 / ATCC BAA-138 / NCIMB 13768)	611	49	30	6	444	139	72	20
		Citrobacter koseri (strain ATCC BAA-895 / CDC 4225-83 / SGSC4696)	722	76	60	10	490	211	85	44
Gamma Proteobacteria	Aerobic	Colwellia psychrerythraea	720	55	67	12	501	169	83	39
		Coxiella burnetii (strain Dugway 5J108-111)	285	32	18	11	207	61	41	21
		Dichelobacter nodosus (strain VCS1703A)	169	29	22	3	120	52	28	17
		Enterobacter sakazakii (strain ATCC BAA-894)	651	72	54	12	450	200	77	38
		Erwinia carotovora (subsp. atroseptica, strain ATCC BAA-672 / SCRI 1043)	729	47	39	16	490	179	98	32

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Escherichia coli (strain K12)	661	68	47	11	450	193	71	29
Francisella tularensis (subsp. tularensis, strain FSC 198)	272	35	14	8	185	87	28	15
Haemophilus ducreyi (strain ATCC 700724 / 35000HP)	236	21	12	6	191	54	14	10
Haemophilus influenzae (strain ATCC 51907 / DSM 11121 / KW20 / Rd)	259	23	14	2	191	69	20	10
Haemophilus somnus (strain 129Pt)	267	31	18	5	201	70	22	17
Hahella chejuensis (strain KCTC 2396)	901	86	80	17	625	225	123	61
Halorhodospira halophila (strain DSM 244 / SL1)	386	21	19	6	301	68	38	16
Idiomarina loihiensis (strain L2-TR / DSM 15497 / ATCC BAA-735)	443	24	32	5	334	86	47	15
Klebsiella pneumoniae (subsp. pneumoniae, strain ATCC 700721 / MGH 78578)	798	85	47	12	554	213	93	36
Legionella pneumophila (strain Paris)	459	87	43	18	315	142	67	41
Marinobacter aquaeolei (strain DSM 11845 / ATCC 700491 / VT8) / VT8)	670	61	45	13	488	144	76	44
Marinomonas sp. MWYL1	788	59	39	13	553	206	76	35
Methylococcus capsulatus (strain Bath / NCIMB 11132)	441	69	40	11	305	111	66	40
Nitrosococcus oceani (strain ATCC 19707 / NCIMB 11848)	495	49	45	19	389	124	51	22
Pasteurella multocida (strain Pm70)	353	32	19	3	253	87	35	17
Photobacterium profundum (strain SS9)	884	84	76	16	548	262	124	56
Photorhabdus luminescens laumondii (strain TT01)	542	64	42	14	353	156	74	34
Pseudoalteromonas atlantica (strain T6c / BAA-1087)	688	79	56	11	508	169	95	39
Pseudoalteromonas haloplanktis (strain TAC 125)	566	42	36	8	380	136	61	34
Pseudomonas aeruginosa (strain LMG 12228 / ATCC 15692 / PRS 101 / 1C / PAO1)	869	88	63	20	610	234	105	48
Pseudomonas entomophila (strain L48)	779	70	52	21	529	205	93	31
Pseudomonas fluorescens (strain Pf-5 / ATCC BAA-477)	957	94	66	24	632	270	108	60
Pseudomonas mendocina (strain ymp)	737	75	40	14	519	188	77	34
Pseudomonas putida (strain F1 / ATCC 700007)	819	72	58	15	548	208	100	41
Pseudomonas stutzeri (strain A1501)	703	69	47	14	478	186	90	44
Pseudomonas syringae (pathovar phaseolicola, strain 1448A / Race 6)	776	82	55	20	552	205	95	43
Psychrobacter arcticum (strain 273-4)	333	25	21	12	273	73	27	11
Psychrobacter cryohalolentis (strain K5)	425	26	29	15	344	88	41	12
Psychrobacter sp. (strain PRwf-1)	417	29	15	5	314	90	36	15
Saccharophagus degradans (strain 2-40 / ATCC 43961 / DSM 17024)	630	80	59	14	451	161	89	46
Salmonella arizonae (strain ATCC BAA-731 / CDC346-86 / RSK2980)	659	63	48	13	409	213	94	37
Salmonella choleraesuis (strain SC-B67)	693	66	46	16	443	217	96	34
Salmonella paratyphi A (strain SARB42 / ATCC 9150)	643	58	44	12	414	186	93	29
Salmonella paratyphi B (strain ATCC BAA-1250 / SPB7)	720	64	47	16	465	221	94	32
Salmonella typhi (strain ATCC 700931 / Ty2)	669	64	51	15	445	195	93	34
Salmonella typhimurium (strain ATCC 700720 / SGSC1412 / LT2)	704	66	52	14	460	212	100	31
Serratia proteamaculans (strain 568)	791	63	40	18	536	217	92	36
Shewanella amazonensis (strain ATCC BAA-1098 / SB2B)	605	57	52	11	401	150	83	44
Shewanella baltica (strain OS155 / ATCC BAA-1091)	716	69	63	14	477	179	94	52
Shewanella denitrificans (strain OS217 / ATCC BAA-1090 / DSM 15013)	592	50	63	9	388	142	87	41
Shewanella frigidimarina (strain NCIMB 400)	695	57	55	9	466	163	100	35
Shewanella loihica (strain BAA-1088 / PV-4)	671	63	49	13	457	148	102	43
Shewanella oneidensis (strain MR-1)	640	59	67	13	436	161	94	39
Shewanella pealeana ATCC 700345	696	69	52	13	464	186	96	44
Shewanella putrefaciens (strain CN-32/ATCC BAA-453)	665	66	49	8	436	161	84	47
Shewanella sediminis HAW-EB3	729	81	57	25	490	180	97	60
Shewanella sp. (strain ANA-3)	759	64	67	9	505	185	111	54
Shigella boydii (serovar 4, strain Sb227)	567	48	33	9	374	158	68	24
Shigella dysenteriae (serovar 1, strain Sd97 / Sd197)	529	57	36	7	354	164	64	17
Shigella flexneri (serovar 2a, strain 2457T/ATCC 700930)	564	59	38	6	387	168	64	24
Shigella sonnei (strain Ss046)	596	56	39	11	398	165	72	29
Sodalis glossinidius (strain morsitans)	312	43	21	7	199	92	47	24
Vesicomyxosocius okutanii (subsp. Calyptogena okutanii, strain HA)	128	8	10	1	106	26	5	2
Vibrio cholerae (strain ATCC 39541 / O395)	637	52	52	7	429	140	85	45

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Gram-positive Bacteria	Actinobacteria	Aerobic	Vibrio fischeri (strain ATCC 700601 / ES114)	651	52	55	13	438	173	80	36			
			Vibrio harveyi (strain ATCC BAA-1116 / BB120)	775	69	59	15	537	200	96	49			
			Vibrio parahaemolyticus (serovar O3:K6, strain RIMD 2210633)	786	63	61	8	543	182	93	60			
			Vibrio vulnificus (strain CMCP6)	764	55	50	15	531	161	97	48			
			Wigglesworthia glossinidia brevipalpis	77	23	4	3	48	32	15	6			
			Xanthomonas axonopodis citri (strain 306)	637	75	73	17	469	170	83	46			
			Xanthomonas campestris (strain ATCC 33913)	623	71	66	20	465	165	78	45			
			Xanthomonas oryzae (pathovar oryzae, MAFF 311018)	529	66	43	24	392	138	69	38			
			Xylella fastidiosa (strain 9a5c)	284	44	32	13	196	92	54	20			
			Xylella fastidiosa (strain Temecula1 / ATCC 700964)	260	38	27	9	165	84	41	19			
			Yersinia enterocolitica (serovar O:8 / strain 8081)	657	69	36	17	451	175	91	40			
			Yersinia pestis (biovar Antiqua Antiqua, strain Antiqua)	658	54	47	15	432	187	87	41			
			Yersinia pseudotuberculosis (serovar I, strain IP32953)	646	57	48	12	436	183	77	44			
			Alkalilimnicola ehrlichei (strain MLHE-1)	492	30	32	5	352	111	53	23			
			Carsonella ruddii (strain PV)	9	4	0	3	5	4	4	1			
			Mannheimia succiniciproducens (strain MBEL55E)	340	32	24	4	251	96	27	17			
			Psychromonas ingrahamii (strain 37)	600	48	39	10	427	126	79	34			
			Ruthia magnifica magnifica (subsp. Calyptogena)	132	9	11	1	111	31	7	6			
			Thiomicrospira crunogena (strain XCL-2)	391	25	22	3	297	81	26	15			
	Gram-negative Bacteria	Actinobacteria	Aerobic	Acidothermus cellulolyticus (strain ATCC 43068 / 11B)	360	39	50	8	304	82	46	12		
				Arthrobacter aureus (strain TC1)	806	62	67	10	711	140	63	29		
				Clavibacter michiganensis (subsp. michiganensis, strain NCPPB 382)	614	39	59	3	598	90	29	11		
				Corynebacterium diphtheriae (biovar gravis, strain NCTC 13129 / ATCC 700971)	394	41	50	9	340	102	47	11		
				Corynebacterium efficiens (strain DSM 44549 / NBRC 100395 / JCM 11189 / YS-314 / AJ 12310)	547	39	57	14	453	118	65	23		
				Corynebacterium glutamicum (strain R)	573	31	53	8	504	107	41	9		
				Corynebacterium jeikeium (strain K411)	374	31	42	4	361	60	35	6		
				Frankia alni (strain ACN14a)	767	136	103	47	682	212	93	59		
				Kineococcus radiotolerans SRS30216	763	66	95	11	678	150	59	26		
				Leifsonia xyli xyli (strain CTCB07)	339	27	32	10	313	72	17	9		
				Mycobacterium avium (strain 104)	524	82	74	18	493	106	60	29		
				Mycobacterium bovis (strain ATCC BAA-935)	490	81	66	17	382	137	76	34		
				Mycobacterium gilvum PYR-GCK	696	90	78	12	622	133	81	39		
				Mycobacterium leprae (strain TN)	198	37	40	14	165	80	21	22		
				Mycobacterium paratuberculosis (strain ATCC BAA-968 / K-10)	510	80	73	20	474	107	58	25		
				Mycobacterium smegmatis (strain ATCC 700084 / mc(2 / mc(2)155)	885	80	87	16	791	174	71	25		
				Mycobacterium tuberculosis (strain ATCC 25177 / H37Ra)	497	80	72	16	379	139	76	33		
				Mycobacterium ulcerans (strain Agy99)	510	83	65	17	408	141	80	31		
				Mycobacterium vanbaalenii (strain DSM 7251 / PYR-1)	800	104	86	21	708	161	84	43		
Gram-negative Bacteria	Actinobacteria	Aerobic	Nocardia farcinica (strain IFM 10152)	774	113	112	23	709	179	85	36			
			Nocardioideis sp. (strain BAA-499 / JS614)	737	54	73	15	666	135	59	26			
			Renibacterium salmoninarum (strain ATCC 33209)	531	63	52	11	491	117	47	12			
			Rhodococcus sp. (strain RHA1)	1043	119	110	26	889	239	105	56			
			Rubrobacter xylanophilus (strain DSM 9941 / NBRC 16129)	524	36	21	5	434	90	34	20			
			Saccharopolyspora erythraea (strain NRRL 23338)	905	66	109	23	783	205	87	39			
			Salinispora arenicola CNS-205	723	65	98	15	642	152	76	32			
			Salinispora tropica CNB-440	711	75	96	21	631	145	83	30			
			Streptomyces avermitilis (strain DSM 46492 / NRRL 8165 / NCIMB 12804 / ATCC 31267 / JCM 5070)	1066	135	148	43	956	249	127	55			
			Streptomyces coelicolor (strain ATCC BAA-471 / M145 / A3(2))	1118	114	157	42	977	266	129	49			
			Thermobifida fusca (strain YX)	538	64	68	12	457	122	64	26			
			Tropheryma whipplei (strain Twist)	131	16	29	2	86	52	24	11			
			Gram-negative Bacteria	Actinobacteria	Anaerobic	Bifidobacterium adolescentis (strain ATCC 15703 / DSM 20083)	247	60	54	13	216	87	40	21
						Bifidobacterium longum (strain NCC 2705)	238	70	39	14	215	87	41	20
						Propionibacterium acnes (strain DSM 16379 / KPA171202)	394	57	50	10	297	127	58	20

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Firmicutes	Aerobic	Acholeplasma laidlawii (strain PG-8A)	293	12	1	0	257	32	6	5
		Bacillus amyloliquefaciens (strain FZB42)	656	50	17	3	506	139	39	27
		Bacillus anthracis (strain Ames, isolate Porton)	939	59	23	6	742	181	54	27
		Bacillus cereus (strain ATCC 10987)	984	67	32	8	760	201	70	32
		Bacillus clausii (strain KSM-K16)	752	44	24	4	595	144	42	22
		Bacillus halodurans (strain C-125 / ATCC BAA-125 / JCM 9153 / FERM 7344 / DSM 18197)	726	40	16	2	585	127	39	15
		Bacillus licheniformis (strain DSM 13 / ATCC 14580, sub_strain Goettingen)	767	46	24	9	591	155	48	24
		Bacillus pumilus (strain SAFR-032)	656	40	12	6	516	118	39	21
		Bacillus subtilis (strain 168)	730	59	22	5	574	142	51	27
		Bacillus thuringiensis (strain Al Hakam)	863	74	24	3	615	211	83	29
		Bacillus weihenstephanensis KBAB4	931	65	26	3	715	185	60	31
		Desulfotobacterium hafniense (strain Y51)	785	119	39	20	580	191	99	49
		Enterococcus faecalis (strain V583 / ATCC 700802)	563	41	11	3	452	113	28	17
		Geobacillus kaustophilus (strain HTA426)	546	48	25	7	443	111	43	21
		Geobacillus thermodenitrificans (strain NG80-2)	610	65	21	6	480	126	58	18
		Lactobacillus acidophilus (strain NCFM)	329	40	13	0	279	72	20	6
		Lactobacillus brevis (strain ATCC 367 / JCM 1170)	424	24	2	0	373	56	10	3
		Lactobacillus casei (strain ATCC 334)	527	36	5	3	480	56	24	6
		Lactobacillus delbrueckii (subsp. bulgaricus, strain ATCC BAA-365)	259	36	4	2	212	61	19	6
		Lactobacillus gasserii (strain ATCC 33323 / DSM 20243)	317	43	10	0	272	63	19	7
		Lactobacillus helveticus (strain DPC 4571)	253	29	2	0	222	41	14	2
		Lactobacillus johnsonii (strain NCC 533)	368	44	6	4	316	69	22	8
		Lactobacillus plantarum (strain WCFS1 / ATCC BAA-793 / NCIMB 8826)	574	46	8	3	481	99	31	9
		Lactobacillus reuteri (strain ATCC 23272 / DSM 20016 / F275)	308	23	5	5	267	50	14	4
		Lactobacillus sakei (subsp. sakei, strain 23K)	25	5	0	0	28	2	0	0
		Lactobacillus salivarius (subsp. salivarius, strain UCC118)	337	30	3	1	283	55	17	3
		Lactococcus lactis (subsp. lactis, strain IL1403)	409	27	1	2	346	58	23	8
		Leuconostoc mesenteroides (subsp. mesenteroides, strain ATCC 8293 / NCDO 523)	390	13	7	2	373	26	11	2
		Listeria innocua (serovar 6a, strain CLIP 11262)	514	39	11	0	451	72	25	10
		Listeria monocytogenes (serovar 1/2a, strain EGD-e / ATCC BAA-679)	511	42	12	0	451	74	29	8
		Listeria welshimeri (serovar 6b, strain ATCC 35897 / DSM 20650 / SLCC5334)	508	42	8	1	447	68	28	8
		Mesoplasma florum (strain ATCC 33453 / NCTC 11704 / L1 / NBRC 100688)	122	15	3	0	95	25	6	10
		Mycoplasma agalactiae	107	21	3	0	94	22	10	3
		Mycoplasma capricolum (subsp. capricolum, strain California kid / ATCC 27343 / NCTC 10154)	138	13	4	1	99	34	10	4
		Mycoplasma gallisepticum (strain R(low))	125	12	2	1	93	28	9	6
		Mycoplasma genitalium (strain ATCC 33530 / NCTC 10195 / G-37)	74	8	3	1	48	25	6	5
		Mycoplasma hyopneumoniae (strain J / ATCC 25934 / NCTC 10110)	110	11	4	1	97	17	9	3
		Mycoplasma mobile (strain 163K / NCTC 11711 / ATCC 43663)	113	3	1	1	95	16	6	1
		Mycoplasma pneumoniae (strain M129 / ATCC 29342)	97	13	2	2	63	30	11	7
		Mycoplasma pulmonis (strain UAB CTIP)	138	9	1	1	115	26	4	3
		Mycoplasma synoviae (strain 53)	106	5	3	0	90	18	5	0
		Oceanobacillus iheyensis (strain KCTC 3954 / DSM 14371 / JCM 11309 / HTE831)	690	29	15	3	592	94	29	12
		Oenococcus oeni (strain BAA-331 / PSU-1)	317	20	4	1	290	38	9	3
		Onion yellows phytoplasma (strain OY-M)	45	35	10	5	55	27	7	6
		Pediococcus pentosaceus (strain ATCC25745 / 183-1w)	319	17	4	0	282	40	10	1
		Pelotomaculum thermopropionicum (strain DSM 13744 / JCM 10971 / SI)	310	67	22	8	224	97	44	20
		Staphylococcus aureus (strain Mu50 / ATCC 700699)	479	32	11	3	405	78	22	13
		Staphylococcus epidermidis (strain ATCC 12228)	424	38	5	2	351	81	22	3
		Staphylococcus haemolyticus (strain JCSC1435)	465	24	3	1	380	72	27	5
		Staphylococcus saprophyticus (subsp. saprophyticus, strain ATCC 15305 / DSM 20229)	433	18	4	1	362	62	19	4

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Anaerobic	Streptococcus agalactiae Ia (serovar Ia, strain A909 / CDC SS700 / ATCC 27591)	378	34	4	4	314	71	17	8
	Streptococcus agalactiae III (serovar III, strain NEM316)	396	30	4	5	327	79	21	5
	Streptococcus agalactiae V (serovar V, strain 2603 V/R / ATCC BAA-611)	369	27	6	7	303	73	17	8
	Streptococcus gordonii (strain Challis / ATCC 35105 / CH1 / DL1 / V288)	421	22	7	1	326	87	18	11
	Streptococcus mutans (serovar c, strain ATCC 700610 / UA159)	355	23	6	3	286	78	14	4
	Streptococcus pneumoniae (serovar 2, strain D39 / NCTC 7466)	357	30	4	3	300	62	21	8
	Streptococcus pneumoniae (strain ATCC BAA-255 / R6)	353	35	5	4	294	65	26	7
	Streptococcus pneumoniae (strain TIGR4 / ATCC BAA-334)	343	31	4	5	282	61	30	6
	Streptococcus pyogenes M1 (serovar M1, strain MGAS5005 / ATCC BAA-947)	295	27	8	3	241	59	19	6
	Streptococcus sanguinis (strain SK36)	459	29	7	2	377	76	27	9
	Streptococcus suis (strain 05ZYH33)	376	41	7	4	308	81	25	13
	Streptococcus thermophilus (strain ATCC BAA-491 / LMD-9)	277	22	7	0	238	38	14	8
	Symbiobacterium thermophilum (strain IAM 14863 / T)	577	62	24	11	484	108	46	20
	Ureaplasma parvum (serovar 3, strain ATCC 700970)	94	9	5	1	76	20	8	3
	Alkaliphilus metalliredigens (strain QYMF)	775	68	11	9	569	166	62	27
	Alkaliphilus oremlandii (strain OhILAs)	518	54	15	6	390	111	44	30
	Caldicellulosiruptor saccharolyticus (strain ATCC 43494 / DSM 8903)	343	70	14	10	262	91	42	14
	Carboxydotherrmus hydrogenoformans (strain Z-2901 / DSM 6008)	353	32	17	5	314	53	17	10
	Clostridium acetobutylicum (strain DSM 792 / JCM 1419 / LMG 5710 / ATCC 824 / VKM B-1787)	527	111	39	28	399	175	68	35
	Clostridium beijerinckii (strain ATCC 51743 / NCIMB 8052)	683	135	42	11	491	208	75	47
	Clostridium botulinum (strain ATCC 19397 / Type A)	512	88	33	12	336	181	66	33
	Clostridium difficile (strain 630)	516	122	31	13	343	179	75	36
	Clostridium kluyveri (strain ATCC 8527 / DSM 555 / NCIMB 10680)	475	116	30	17	350	151	51	36
	Clostridium novyi (strain NT)	305	77	23	15	206	103	58	27
	Clostridium perfringens (strain ATCC 13124 / NCTC 8237 / Type A)	501	72	38	14	351	158	56	30
	Clostridium phytofermentans (strain ATCC 700394 / DSM 18823 / ISDg)	580	141	46	31	393	204	83	53
	Clostridium tetani (strain Massachusetts / E88)	368	89	35	16	277	141	52	19
	Clostridium thermocellum (strain ATCC 27405 / DSM 1237)	380	97	39	19	280	113	63	34
	Desulfotomaculum reducens (strain MI-1)	463	68	18	10	325	121	53	22
	Moorella thermoacetica (strain ATCC 39073)	383	51	13	4	287	90	32	16
	Mycoplasma mycoides SC (subsp. mycoides, strain PG1)	143	19	3	1	110	32	10	7
	Mycoplasma penetrans (strain HF-2)	138	40	12	2	113	42	20	7

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Cytoplasmic proteins

				Number of cytoplasmic proteins				
				0 Cys	1 Cys	2 Cys	3 Cys	
Gram-negative Bacteria	Alpha proteobacteria	Aerobic	Species	Trend				
			Acidiphilium cryptum (strain JF-5)	Semi-normal	277	342	347	297
			Agrobacterium tumefaciens (strain C58 / ATCC 33970)	Semi-normal	476	577	551	416
			Azorhizobium caulinodans (strain ATCC 43989 / DSM 5975 / ORS 571)	Semi-normal	393	460	467	375
			Bartonella bacilliformis (strain ATCC 35685 / KC583)	Semi-normal	89	109	101	92
			Bartonella henselae (strain Houston 1 / ATCC 49882)	Semi-normal	95	126	113	117
			Bartonella quintana (strain Toulouse)	Semi-normal	61	101	92	97
			Bartonella tribocorum CIP 105476	Semi-normal	112	154	121	139
			Bradyrhizobium japonicum (strain USDA 110)	Semi-normal	580	710	778	572
			Brucella abortus (strain 2308)	Semi-normal	268	335	297	254
			Brucella canis ATCC 23365	Semi-normal	259	338	312	258
			Brucella melitensis (biovar 1, strain NCTC 10094 / ATCC 23456 / 16M)	Semi-normal	323	357	345	277
			Brucella suis ATCC 23445	Semi-normal	268	329	312	255
			Caulobacter crescentus (strain CB15 / ATCC 19089)	Semi-normal	359	440	378	279
			Dinoroseobacter shibae DFL 12	Semi-normal	280	336	331	307
			Ehrlichia canis (strain Jake)	Semi-normal	25	64	66	79
			Ehrlichia chaffeensis (strain Arkansas)	Semi-normal	22	62	68	63
			Ehrlichia ruminantium (strain Gardel)	Semi-normal	26	64	63	56
			Erythrobacter litoralis (strain HTCC2594)	Semi-normal	281	304	290	229
			Gluconacetobacter diazotrophicus PAI 5	Semi-normal	256	297	317	270
			Gluconobacter oxydans (strain 621H)	Semi-normal	218	223	241	194
			Granulobacter betshdensis (strain ATCC BAA-1260 / CGDNIH1)	Semi-normal	188	225	241	196
			Hyphomonas neptunium (strain ATCC 15444)	Semi-normal	290	349	342	313
			Jannaschia sp. (strain CCS1)	Semi-normal	332	416	431	375
			Magnetospirillum magneticum (strain AMB-1 / ATCC 700264)	Semi-normal	335	388	365	313
			Maricaulis maris (strain MCS10)	Semi-normal	248	272	295	230
			Mesorhizobium sp. (strain BNC1)	Semi-normal	416	474	444	380
			Methylobacterium extorquens PA1	Exclusionary	464	462	450	344
			Neorickettsia sennetsu (strain Miyayama)	Semi-normal	18	55	60	51
			Nitrobacter hamburgensis (strain X14 / DSM 10229)	Semi-normal	406	420	402	308
			Nitrobacter winogradskyi (strain Nb-255 / ATCC 25391)	Semi-normal	331	348	292	221
			Novosphingobium aromaticivorans (strain DSM 12444)	Semi-normal	269	332	308	264
			Orientia tsutsugamushi (strain Boryong)	Semi-normal	37	42	66	57
			Paracoccus denitrificans (strain Pd 1222)	Semi-normal	434	493	466	402
			Parvibaculum lavamentivorans (strain DS-1 / DSM 13023 / NCIMB 13966)	Semi-normal	331	396	374	311
			Pelagibacter ubique (strain HTCC1062)	Semi-normal	112	154	127	120
			Rhizobium etli (strain CFN 42 / ATCC 51251)	Semi-normal	544	611	633	501
			Rhizobium leguminosarum bv. viciae (strain 3841)	Semi-normal	657	729	724	549
			Rhizobium loti (strain MAFF303099)	Semi-normal	628	794	664	561
			Rhizobium meliloti (strain 1021)	Semi-normal	507	582	578	504
			Rhodobacter sphaeroides (strain ATCC 17023 / 2.4.1 / NCIB 8253 / DSM 158)	Semi-normal	340	408	413	300
			Rhodopseudomonas palustris (strain ATCC BAA-98 / CGA009)	Semi-normal	390	464	457	357
			Rhodospirillum rubrum (strain ATCC 11170 / NCIB 8255)	Semi-normal	325	360	345	292
			Roseobacter denitrificans (strain OCh 114 / ATCC 33942)	Semi-normal	286	377	328	336
			Silicibacter pomeroyi (strain ATCC 700808 / DSS-3 / DSM 15171)	Semi-normal	297	411	413	348
			Silicibacter sp. (strain TM1040)	Semi-normal	336	373	404	365
			Sinorhizobium medicae (strain WSM419)	Semi-normal	490	606	547	512
			Sphingomonas wittichii (strain RW1 / DSM 6014 / JCM 10273)	Semi-normal	482	545	505	419
			Sphingopyxis alaskensis	Semi-normal	308	346	316	232
			Wolbachia pipientis wMel	Semi-normal	69	70	96	84
			Wolbachia sp. (subsp. Brugia malayi, strain TRS)	Semi-normal	46	65	75	57
			Xanthobacter sp.	Semi-normal	421	510	419	351
Zymomonas mobilis (strain CP4 / ZM4 / ATCC 31821)	Exclusionary	201	195	188	148			

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Beta Proteobacteria	Anaerobic	Anaplasma marginale (strain St. Maries)	Semi-normal	9	39	55	53
		Anaplasma phagocytophilum (strain HZ)	Semi-normal	17	44	53	65
		Brucella ovis (strain ATCC 25840 / 63/290 / NCTC 10512)	Semi-normal	244	308	282	252
		Ochrobactrum anthropi (strain ATCC 49188 / DSM 6882 / NCTC 12168)	Semi-normal	463	547	457	361
		Rickettsia akari (strain Hartford)	Semi-normal	89	106	109	73
		Rickettsia bellii (strain OSU 85-389)	Semi-normal	126	180	133	116
		Rickettsia canadensis (strain McKiel)	Semi-normal	62	72	91	79
		Rickettsia conorii (strain ATCC VR-613 / Malish 7)	Semi-normal	150	152	129	90
		Rickettsia felis (strain ATCC VR-1525 / URRWXCal2)	Semi-normal	142	165	145	119
		Rickettsia massiliae (strain Mtu5)	Semi-normal	83	95	96	71
		Rickettsia prowazekii (strain Madrid E)	Semi-normal	52	78	74	72
		Rickettsia rickettsii Iowa	Exclusionary	125	118	111	82
	Aerobic	Rickettsia typhi (strain Wilmington / ATCC VR-144)	Semi-normal	52	67	81	55
		Acidovorax avenae (subsp. citrulli, strain AAC00-1)	Semi-normal	313	389	388	356
		Acidovorax sp. (strain JS42)	Semi-normal	298	301	328	307
		Bordetella avium (strain 197N)	Semi-normal	262	291	325	263
		Bordetella bronchiseptica (strain NCTC 13252 / RB50 / ATCC BAA-588)	Semi-normal	325	362	450	346
		Bordetella parapertussis (strain NCTC 13253 / ATCC BAA-587 / 12822)	Semi-normal	258	307	375	291
		Bordetella pertussis (strain ATCC BAA-589 / Tohama I / NCTC 13251)	Semi-normal	245	261	291	253
		Bordetella petrii	Semi-normal	322	372	429	368
		Burkholderia cenocepacia (strain AU 1054)	Semi-normal	447	562	561	502
		Burkholderia cepacia (strain ATCC 53795 / AMMD)	Semi-normal	464	555	564	505
		Burkholderia mallei (strain ATCC 23344)	Semi-normal	304	390	449	377
		Burkholderia multivorans ATCC 17616	Semi-normal	424	503	511	473
		Burkholderia pseudomallei (strain 668)	Semi-normal	385	501	532	479
		Burkholderia thailandensis (strain E264 / ATCC 700388 / DSM 13276 / CIP 106301)	Semi-normal	328	413	474	420
		Burkholderia vietnamiensis (strain R1808 / G4 / LMG 22486) / LMG 22486)	Semi-normal	535	694	652	582
		Burkholderia xenovorans (strain LB400)	Semi-normal	534	716	754	680
		Chromobacterium violaceum (strain IFO 12614 / ATCC 12472 / DSM 30191 / NCIB 9131 / JCM 1249)	Semi-normal	280	362	385	371
		Dechloromonas aromatica (strain RCB)	Semi-normal	250	293	337	288
		Delftia acidovorans SPH-1	Semi-normal	391	469	507	437
		Janthinobacterium sp. (strain Marseille)	Semi-normal	261	347	371	257
		Methylobium petroleiphilum (strain PM1)	Semi-normal	304	397	383	330
		Methylobacillus flagellatus (strain KT / ATCC 51484 / DSM 6875)	Semi-normal	235	249	240	207
		Neisseria gonorrhoeae (strain ATCC 700825 / FA 1090)	Semi-normal	136	161	190	161
		Neisseria meningitidis (serogroup C / , serovar 2a, strain ATCC 700532 / FAM18)	Semi-normal	130	174	185	159
		Nitrosomonas europaea (strain IFO 14298 / ATCC 19718)	Semi-normal	185	207	189	222
		Nitrosomonas eutropha (strain C71)	Semi-normal	175	203	211	199
		Nitrospira multiformis (strain ATCC 25196 / NCIMB 11849)	Exclusionary	230	226	212	215
		Polaromonas naphthalenivorans (strain CJ2)	Semi-normal	335	398	438	369
		Polaromonas sp. (strain JS666 / ATCC BAA-500)	Semi-normal	364	479	502	443
		Polynucleobacter sp. (strain QLW-P1DMWA-1)	Semi-normal	162	185	188	184
		Ralstonia eutropha (strain ATCC 17699 / H16 / DSM 428 / Stanier 337)	Semi-normal	390	510	535	498
		Ralstonia metallidurans (strain CH34 / ATCC 43123)	Semi-normal	383	494	550	477
		Ralstonia solanacearum (strain GMI1000)	Semi-normal	342	452	391	386
		Thiobacillus denitrificans (strain ATCC 25259)	Semi-normal	210	261	262	228
		Verminephrobacter eiseniae (strain EF01-2)	Semi-normal	221	329	329	368
Beta Proteobacteria	Anaerobic	Herminiimonas arsenicoxydans (strain ULPAs1)	Semi-normal	253	295	291	248
	Anaerobic	Rhodoferrax ferrireducens (strain DSM 15236 / ATCC BAA-621 / T118)	Semi-normal	269	342	357	359

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Delta Proteobacteria	Aerobic	Bdellovibrio bacteriovorus (strain DSM 50701 / NCIB 9529 / HD100 / ATCC 15356)	Semi-normal	227	260	302	227
		Geobacter uraniumreducens (strain Rf4)	Semi-normal	264	319	308	262
		Lawsonia intracellularis (strain PHE/MN1-00)	Remaining	94	77	106	105
		Myxococcus xanthus (strain DK 1622)	Semi-normal	537	634	643	487
		Sorangium cellulosum (strain So ce56)	Semi-normal	552	701	708	627
	Anaerobic	Anaeromyxobacter dehalogenans (strain 2CP-C)	Semi-normal	399	408	382	261
		Desulfococcus oleovorans Hxd3	Semi-normal	172	242	238	212
		Desulfotalea psychrophila (strain LSV54 / DSM 12343)	Semi-normal	158	191	232	213
		Desulfovibrio desulfuricans (strain G20)	Semi-normal	165	202	193	213
		Desulfovibrio vulgaris (strain Hildenborough / ATCC 29579 / NCIMB 8303)	Semi-normal	161	194	233	212
		Geobacter metallireducens (strain GS-15 / ATCC 53774 / DSM 7210)	Semi-normal	193	244	268	240
		Geobacter sulfurreducens (strain ATCC 51573 / DSM 12127 / PCA)	Semi-normal	192	254	250	221
		Pelobacter carbinolicus (strain DSM 2380 / Gra Bd 1)	Semi-normal	137	186	214	240
		Pelobacter propionicus (strain DSM 2379)	Semi-normal	196	243	237	273
		Syntrophobacter fumaroxidans (strain DSM 10017 / MPOB)	Semi-normal	196	265	316	301
		Syntrophus aciditrophicus (strain SB)	Semi-normal	169	250	245	239
		Arcobacter butzleri (strain RM4018)	Semi-normal	173	237	212	184
		Campylobacter concisus (strain 13826)	Remaining	161	161	154	135
		Campylobacter curvus (strain 525.92)	Remaining	131	131	136	133
	Aerobic	Campylobacter fetus (subsp. fetus, strain 82-40)	Semi-normal	106	134	135	132
		Campylobacter hominis (strain ATCC BAA-381 / LMG 19568 / NCTC 13146 / CH001A)	Semi-normal	84	133	142	111
		Campylobacter jejuni (subsp. doylei, strain ATCC BAA-1458 / RM4099 / 269.97)	Semi-normal	112	119	134	104
		Helicobacter acinonychis (strain Sheeba)	Remaining	121	109	138	109
		Helicobacter hepaticus (strain 3B1 / ATCC 51449)	Semi-normal	100	121	135	120
		Helicobacter pylori (strain 26695 / ATCC 700392)	Semi-normal	107	108	133	101
		Nitratiruptor sp. (strain SB155-2)	Semi-normal	184	211	190	157
		Sulfurimonas denitrificans (strain ATCC33889/DSM 1251)	Exclusionary	221	207	203	185
		Sulfurovum sp. (strain NBC37-1)	Semi-normal	209	224	225	177
	Anaerobic	Wolinella succinogenes (strain DSMZ 1740)	Semi-normal	161	176	198	157
Epsilon Proteobacteria	Aerobic	Acinetobacter baumannii (strain ATCC 17978)	Exclusionary	530	468	432	346
		Acinetobacter sp. (strain ADP1)	Semi-normal	246	305	309	247
		Actinobacillus pleuropneumoniae (serovar5b, strain L20)	Semi-normal	154	186	167	187
		Actinobacillus succinogenes (strain ATCC 55618 / 130Z)	Semi-normal	136	203	165	183
		Aeromonas hydrophila (subsp. hydrophila, strain ATCC 7966 / NCIB 9240)	Semi-normal	208	265	330	295
		Aeromonas salmonicida (strain A449)	Semi-normal	253	285	369	326
		Alcanivorax borkumensis (strain SK2 / ATCC 700651 / DSM 11573)	Semi-normal	160	207	248	235
		Baumannia cicadellinicola coagulata (subsp. Homalodisca)	Semi-normal	41	56	48	61
		Blochmannia floridanus	Semi-normal	18	48	43	59
		Blochmannia pennsylvanicus (strain BPEN)	Semi-normal	25	45	52	57
		Buchnera aphidicola (subsp. Acyrthosiphon pisum, strain Tokyo 1998)	Semi-normal	54	60	50	65
		Buchnera aphidicola cedri (subsp. Cinara)	Semi-normal	31	34	40	42
		Chromohalobacter salexigens (strain DSM 3043 / ATCC BAA-138 / NCIMB 13768)	Semi-normal	202	287	331	283
		Citrobacter koseri (strain ATCC BAA-895 / CDC 4225-83 / SGSC4696)	Semi-normal	268	325	346	335
		Colwellia psychrerythraea	Semi-normal	216	317	330	305
		Coxiella burnetii (strain Dugway 5J108-111)	Semi-normal	134	168	153	175
		Dichelobacter nodosus (strain VCS1703A)	Semi-normal	85	90	103	99
		Enterobacter sakazakii (strain ATCC BAA-894)	Semi-normal	286	356	319	325
		Erwinia carotovora (subsp. atroseptica, strain ATCC BAA-672 / SCRI 1043)	Semi-normal	293	341	374	321
	Anaerobic						
Gamma Proteobacteria	Aerobic						

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	Escherichia coli (strain K12)	Semi-normal	247	295	312	338
	Francisella tularensis (subsp. tularensis, strain FSC 198)	Semi-normal	85	134	156	113
	Haemophilus ducreyi (strain ATCC 700724 / 35000HP)	Semi-normal	128	151	135	131
	Haemophilus influenzae (strain ATCC 51907 / DSM 11121 / KW20 / Rd)	Semi-normal	137	150	169	147
	Haemophilus somnus (strain 129Pt)	Semi-normal	127	184	158	160
	Hahella chejuensis (strain KCTC 2396)	Semi-normal	387	467	513	509
	Halorhodospira halophila (strain DSM 244 / SL1)	Semi-normal	170	202	233	212
	Idiomarina loihiensis (strain L2-TR / DSM 15497 / ATCC BAA-735)	Semi-normal	212	237	221	210
	Klebsiella pneumoniae (subsp. pneumoniae, strain ATCC 700721 / MGH 78578)	Semi-normal	282	384	454	403
	Legionella pneumophila (strain Paris)	Semi-normal	169	202	196	201
	Marinobacter aquaeolei (strain DSM 11845 / ATCC 700491 / VT8) / VT8)	Semi-normal	303	321	389	304
	Marinomonas sp. MWYL1	Semi-normal	246	354	390	395
	Methylococcus capsulatus (strain Bath / NCIMB 11132)	Semi-normal	185	223	243	226
	Nitrosococcus oceani (strain ATCC 19707 / NCIMB 11848)	Semi-normal	202	249	237	235
	Pasteurella multocida (strain Pm70)	Semi-normal	121	148	166	166
	Photobacterium profundum (strain SS9)	Semi-normal	300	368	422	375
	Photorhabdus luminescens laumondii (strain TT01)	Semi-normal	337	367	363	310
	Pseudoalteromonas atlantica (strain T6c / BAA-1087)	Semi-normal	235	320	347	297
	Pseudoalteromonas haloplanktis (strain TAC 125)	Semi-normal	211	246	269	289
	Pseudomonas aeruginosa (strain LMG 12228 / ATCC 15692 / PRS 101 / 1C / PAO1)	Semi-normal	324	438	479	388
	Pseudomonas entomophila (strain L48)	Semi-normal	293	388	423	392
	Pseudomonas fluorescens (strain Pf-5 / ATCC BAA-477)	Semi-normal	326	451	502	444
	Pseudomonas mendocina (strain ymp)	Semi-normal	269	337	361	374
	Pseudomonas putida (strain F1 / ATCC 700007)	Semi-normal	299	357	410	385
	Pseudomonas stutzeri (strain A1501)	Semi-normal	216	282	346	293
	Pseudomonas syringae (pathovar phaseolicola, strain 1448A / Race 6)	Semi-normal	292	416	425	420
	Psychrobacter arcticum (strain 273-4)	Semi-normal	179	218	204	181
	Psychrobacter cryohalolentis (strain K5)	Semi-normal	173	221	232	223
	Psychrobacter sp. (strain PRwf-1)	Semi-normal	185	242	227	182
	Saccharophagus degradans (strain 2-40 / ATCC 43961 / DSM 17024)	Semi-normal	204	278	305	258
	Salmonella arizonae (strain ATCC BAA-731 / CDC346-86 / RSK2980)	Semi-normal	243	353	334	309
	Salmonella choleraesuis (strain SC-B67)	Semi-normal	249	350	354	340
	Salmonella paratyphi A (strain SARB42 / ATCC 9150)	Semi-normal	239	295	295	309
	Salmonella paratyphi B (strain ATCC BAA-1250 / SPB7)	Semi-normal	257	344	359	326
	Salmonella typhi (strain ATCC 700931 / Ty2)	Semi-normal	244	324	319	312
	Salmonella typhimurium (strain ATCC 700720 / SGSC1412 / LT2)	Semi-normal	251	338	340	337
	Serratia proteamaculans (strain 568)	Semi-normal	314	398	413	342
	Shewanella amazonensis (strain ATCC BAA-1098 / SB2B)	Semi-normal	187	239	279	252
	Shewanella baltica (strain OS155 / ATCC BAA-1091)	Semi-normal	252	324	304	319
	Shewanella denitrificans (strain OS217 / ATCC BAA-1090 / DSM 15013)	Semi-normal	232	285	315	269
	Shewanella frigidimarina (strain NCIMB 400)	Semi-normal	234	293	292	279
	Shewanella loihica (strain BAA-1088 / PV-4)	Semi-normal	198	256	278	274
	Shewanella oneidensis (strain MR-1)	Semi-normal	282	308	320	309
	Shewanella pealeana ATCC 700345	Semi-normal	211	256	325	285
	Shewanella putrefaciens (strain CN-32/ATCC BAA-453)	Semi-normal	223	289	302	290
	Shewanella sediminis HAW-EB3	Semi-normal	239	284	308	318
	Shewanella sp. (strain ANA-3)	Semi-normal	250	315	320	312
	Shigella boydii (serovar 4, strain Sb227)	Semi-normal	238	292	317	275
	Shigella dysenteriae (serovar 1, strain Sd97 / Sd197)	Semi-normal	248	291	294	276
	Shigella flexneri (serovar2a, strain 2457T/ATCC 700930)	Semi-normal	242	291	299	293
	Shigella sonnei (strain Ss046)	Semi-normal	263	279	305	289
	Sodalis glossinidius (strain morsitans)	Semi-normal	177	215	253	219
	Vesicomysococcus okutanii (subsp. Calyptogena okutanii, strain HA)	Semi-normal	75	88	84	104
	Vibrio cholerae (strain ATCC 39541 / O395)	Semi-normal	217	257	303	263

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Gram-positive Bacteria	Actinobacteria	Anaerobic	<i>Vibrio fischeri</i> (strain ATCC 700601 / ES114)	Semi-normal	225	272	308	276
			<i>Vibrio harveyi</i> (strain ATCC BAA-1116 / BB120)	Semi-normal	321	329	397	398
			<i>Vibrio parahaemolyticus</i> (serovar O3:K6, strain RIMD 2210633)	Semi-normal	268	306	348	343
			<i>Vibrio vulnificus</i> (strain CMCP6)	Remaining	318	316	364	372
			<i>Wigglesworthia glossinidia brevipalpis</i>	Semi-normal	42	50	50	57
			<i>Xanthomonas axonopodis citri</i> (strain 306)	Semi-normal	373	381	379	304
			<i>Xanthomonas campestris</i> (strain ATCC 33913)	Remaining	367	357	359	279
			<i>Xanthomonas oryzae</i> (pathovar <i>oryzae</i> , MAFF 311018)	Semi-normal	304	338	341	292
			<i>Xylella fastidiosa</i> (strain 9a5c)	Semi-normal	159	220	200	186
			<i>Xylella fastidiosa</i> (strain Temecula1 / ATCC 700964)	Semi-normal	129	172	181	145
			<i>Yersinia enterocolitica</i> (serovar O:8 / strain 8081)	Semi-normal	278	344	359	289
			<i>Yersinia pestis</i> (biovar Antiqua Antiqua, strain Antiqua)	Semi-normal	262	341	336	277
			<i>Yersinia pseudotuberculosis</i> (serovar I, strain IP32953)	Semi-normal	267	328	333	278
			<i>Alkalilimnicola ehrlichei</i> (strain MLHE-1)	Semi-normal	202	235	255	227
			<i>Carsonella ruddii</i> (strain PV)	Semi-normal	4	19	27	20
			<i>Mannheimia succiniciproducens</i> (strain MBEL55E)	Semi-normal	145	200	194	163
			<i>Psychromonas ingrahamii</i> (strain 37)	Semi-normal	200	238	282	270
			<i>Ruthia magnifica magnifica</i> (subsp. <i>Calyptogena</i>)	Semi-normal	71	91	97	111
			<i>Thiomicrospira crunogena</i> (strain XCL-2)	Remaining	211	204	226	190
	Actinobacteria	Aerobic	<i>Acidothermus cellulolyticus</i> (strain ATCC 43068 / 11B)	Semi-normal	225	232	218	194
			<i>Arthrobacter aureus</i> (strain TC1)	Semi-normal	583	640	480	349
			<i>Clavibacter michiganensis</i> (subsp. <i>michiganensis</i> , strain NCPPB 382)	Exclusionary	603	491	284	161
			<i>Corynebacterium diphtheriae</i> (biovar <i>gravis</i> , strain NCTC 13129 / ATCC 700971)	Semi-normal	213	264	236	188
			<i>Corynebacterium efficiens</i> (strain DSM 44549 / NBRC 100395 / JCM 11189 / YS-314 / AJ 12310)	Exclusionary	355	345	330	233
			<i>Corynebacterium glutamicum</i> (strain R)	Semi-normal	425	451	335	238
			<i>Corynebacterium jeikeium</i> (strain K411)	Semi-normal	234	269	215	168
			<i>Frankia alni</i> (strain ACN14a)	Semi-normal	746	785	695	505
			<i>Kineococcus radiotolerans</i> SRS30216	Semi-normal	584	636	492	354
			<i>Leifsonia xyli xyli</i> (strain CTCB07)	Exclusionary	390	315	199	121
			<i>Mycobacterium avium</i> (strain 104)	Semi-normal	527	538	496	399
			<i>Mycobacterium bovis</i> (strain ATCC BAA-935)	Semi-normal	354	395	401	349
			<i>Mycobacterium gilvum</i> PYR-GCK	Semi-normal	521	613	585	438
			<i>Mycobacterium leprae</i> (strain TN)	Semi-normal	145	165	170	168
			<i>Mycobacterium paratuberculosis</i> (strain ATCC BAA-968 / K-10)	Semi-normal	443	510	471	348
			<i>Mycobacterium smegmatis</i> (strain ATCC 700084 / mc(2 / mc(2)155)	Semi-normal	719	774	673	560
			<i>Mycobacterium tuberculosis</i> (strain ATCC 25177 / H37Ra)	Semi-normal	366	395	414	361
			<i>Mycobacterium ulcerans</i> (strain Agy99)	Semi-normal	373	467	388	369
			<i>Mycobacterium vanbaalenii</i> (strain DSM 7251 / PYR-1)	Semi-normal	595	673	636	482
			<i>Nocardia farcinica</i> (strain IFM 10152)	Semi-normal	777	788	603	477
			<i>Nocardioideis</i> sp. (strain BAA-499 / JS614)	Semi-normal	545	619	520	412
			<i>Renibacterium salmoninarum</i> (strain ATCC 33209)	Semi-normal	485	498	409	214
			<i>Rhodococcus</i> sp. (strain RHA1)	Semi-normal	864	1004	983	727
			<i>Rubrobacter xylanophilus</i> (strain DSM 9941 / NBRC 16129)	Exclusionary	345	332	327	260
			<i>Saccharopolyspora erythraea</i> (strain NRRL 23338)	Semi-normal	753	792	724	556
			<i>Salinispora arenicola</i> CNS-205	Semi-normal	462	563	493	425
			<i>Salinispora tropica</i> CNB-440	Semi-normal	427	550	517	409
			<i>Streptomyces avermitilis</i> (strain DSM 46492 / NRRL 8165 / NCIMB 12804 / ATCC 31267 / JCM 5070)	Semi-normal	752	840	736	580
			<i>Streptomyces coelicolor</i> (strain ATCC BAA-471 / M145 / A3(2))	Semi-normal	816	896	785	572
			<i>Thermobifida fusca</i> (strain YX)	Semi-normal	334	360	324	264
			<i>Tropheryma whipplei</i> (strain Twist)	Semi-normal	43	51	45	66
Gram-negative Bacteria	Actinobacteria	Anaerobic	<i>Bifidobacterium adolescentis</i> (strain ATCC 15703 / DSM 20083)	Semi-normal	141	153	168	111
			<i>Bifidobacterium longum</i> (strain NCC 2705)	Exclusionary	177	176	167	161
			<i>Propionibacterium acnes</i> (strain DSM 16379 / KPA171202)	Semi-normal	153	199	203	178

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Firmicutes	Aerobic	Acholeplasma laidlawii (strain PG-8A)	Exclusionary	324	225	161	76
		Bacillus amyloliquefaciens (strain FZB42)	Semi-normal	399	412	370	283
		Bacillus anthracis (strain Ames, isolate Porton)	Semi-normal	491	529	472	375
		Bacillus cereus (strain ATCC 10987)	Semi-normal	466	559	517	399
		Bacillus clausii (strain KSM-K16)	Semi-normal	440	467	436	330
		Bacillus halodurans (strain C-125 / ATCC BAA-125 / JCM 9153 / FERM 7344 / DSM 18197)	Semi-normal	427	473	403	336
		Bacillus licheniformis (strain DSM 13 / ATCC 14580, sub_strain Goettingen)	Semi-normal	465	481	416	331
		Bacillus pumilus (strain SAFR-032)	Semi-normal	425	439	408	279
		Bacillus subtilis (strain 168)	Semi-normal	458	467	406	328
		Bacillus thuringiensis (strain AI Hakam)	Semi-normal	436	523	496	392
		Bacillus weihenstephanensis KBAB4	Semi-normal	457	539	486	363
		Desulfotobacterium hafniense (strain Y51)	Semi-normal	333	429	432	350
		Enterococcus faecalis (strain V583 / ATCC 700802)	Exclusionary	490	403	303	193
		Geobacillus kaustophilus (strain HTA426)	Exclusionary	355	348	317	297
		Geobacillus thermodenitrificans (strain NG80-2)	Semi-normal	347	365	354	292
		Lactobacillus acidophilus (strain NCFM)	Exclusionary	329	257	173	122
		Lactobacillus brevis (strain ATCC 367 / JCM 1170)	Exclusionary	544	304	172	121
		Lactobacillus casei (strain ATCC 334)	Exclusionary	775	310	180	105
		Lactobacillus delbrueckii (subsp. bulgaricus, strain ATCC BAA-365)	Exclusionary	254	202	165	115
		Lactobacillus gasseri (strain ATCC 33323 / DSM 20243)	Exclusionary	330	249	189	102
		Lactobacillus helveticus (strain DPC 4571)	Exclusionary	282	237	151	107
		Lactobacillus johnsonii (strain NCC 533)	Exclusionary	362	258	191	111
		Lactobacillus plantarum (strain WCFS1 / ATCC BAA-793 / NCIMB 8826)	Exclusionary	612	393	300	173
		Lactobacillus reuteri (strain ATCC 23272 / DSM 20016 / F275)	Exclusionary	404	264	195	110
		Lactobacillus sakei (subsp. sakei, strain 23K)	Exclusionary	505	321	229	130
		Lactobacillus salivarius (subsp. salivarius, strain UCC118)	Exclusionary	423	269	206	143
		Lactococcus lactis (subsp. lactis, strain IL1403)	Exclusionary	543	314	199	113
		Leuconostoc mesenteroides (subsp. mesenteroides, strain ATCC 8293 / NCDO 523)	Exclusionary	793	200	80	62
		Listeria innocua (serovar 6a, strain CLIP 11262)	Exclusionary	522	417	363	214
		Listeria monocytogenes (serovar 1/2a, strain EGD-e / ATCC BAA-679)	Exclusionary	482	387	347	211
		Listeria welshimeri (serovar 6b, strain ATCC 35897 / DSM 20650 / SLCC5334)	Exclusionary	461	365	344	193
		Mesoplasma florum (strain ATCC 33453 / NCTC 11704 / L1 / NBRC 100688)	Exclusionary	103	92	75	64
		Mycoplasma agalactiae	Semi-normal	94	98	69	59
		Mycoplasma capricolum (subsp. capricolum, strain California kid / ATCC 27343 / NCTC 10154)	Remaining	115	76	79	57
		Mycoplasma gallisepticum (strain R(low))	Remaining	88	72	82	56
		Mycoplasma genitalium (strain ATCC 33530 / NCTC 10195 / G-37)	Remaining	50	44	44	56
		Mycoplasma hyopneumoniae (strain J / ATCC 25934 / NCTC 10110)	Exclusionary	102	91	74	47
		Mycoplasma mobile (strain 163K / NCTC 11711 / ATCC 43663)	Exclusionary	170	122	54	34
		Mycoplasma pneumoniae (strain M129 / ATCC 29342)	Semi-normal	55	61	66	56
		Mycoplasma pulmonis (strain UAB CTIP)	Exclusionary	145	132	90	34
		Mycoplasma synoviae (strain 53)	Remaining	110	87	96	46
		Oceanobacillus iheyensis (strain KCTC 3954 / DSM 14371 / JCM 11309 / HTE831)	Exclusionary	474	454	385	265
		Oenococcus oeni (strain BAA-331 / PSU-1)	Exclusionary	539	224	110	56
		Onion yellows phytoplasma (strain OY-M)	Semi-normal	50	58	45	43
		Pediococcus pentosaceus (strain ATCC25745 / 183-1w)	Exclusionary	493	235	154	86
		Pelotomaculum thermopropionicum (strain DSM 13744 / JCM 10971 / SI)	Semi-normal	222	251	249	230
		Staphylococcus aureus (strain Mu50 / ATCC 700699)	Exclusionary	380	371	285	201
		Staphylococcus epidermidis (strain ATCC 12228)	Exclusionary	328	319	252	174
		Staphylococcus haemolyticus (strain JCSC1435)	Exclusionary	419	366	261	200
		Staphylococcus saprophyticus (subsp. saprophyticus, strain ATCC 15305 / DSM 20229)	Exclusionary	396	349	266	202

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			Streptococcus agalactiae Ia (serovar Ia, strain A909 / CDC SS700 / ATCC 27591)	Exclusionary	392	280	186	139
			Streptococcus agalactiae III (serovar III, strain NEM316)	Exclusionary	392	265	187	137
			Streptococcus agalactiae V (serovar V, strain 2603 V/R / ATCC BAA-611)	Exclusionary	386	267	190	134
			Streptococcus gordonii (strain Challis / ATCC 35105 / CH1 / DL1 / V288)	Exclusionary	392	253	204	149
			Streptococcus mutans (serovar c, strain ATCC 700610 / UA159)	Exclusionary	393	248	188	137
			Streptococcus pneumoniae (serovar 2, strain D39 / NCTC 7466)	Exclusionary	361	262	194	137
			Streptococcus pneumoniae (strain ATCC BAA-255 / R6)	Exclusionary	362	254	204	130
			Streptococcus pneumoniae (strain TIGR4 / ATCC BAA-334)	Exclusionary	358	245	190	130
			Streptococcus pyogenes M1 (serovar M1, strain MGAS5005 / ATCC BAA-947)	Exclusionary	348	227	153	129
			Streptococcus sanguinis (strain SK36)	Exclusionary	419	299	225	167
			Streptococcus suis (strain 05ZYH33)	Exclusionary	392	277	224	133
			Streptococcus thermophilus (strain ATCC BAA-491 / LMD-9)	Exclusionary	335	243	152	106
			Symbiobacterium thermophilum (strain IAM 14863 / T)	Semi-normal	305	314	320	261
			Ureaplasma parvum (serovar 3, strain ATCC 700970)	Exclusionary	87	73	56	55
			Alkaliphilus metalliredigens (strain QYMF)	Remaining	390	387	424	326
			Alkaliphilus oremlandii (strain OhILAs)	Semi-normal	260	264	261	225
			Caldicellulosiruptor saccharolyticus (strain ATCC 43494 / DSM 8903)	Remaining	201	200	223	181
			Carboxydotherrus hydrogenoformans (strain Z-2901 / DSM 6008)	Exclusionary	337	309	267	175
			Clostridium acetobutylicum (strain DSM 792 / JCM 1419 / LMG 5710 / ATCC 824 / VKM B-1787)	Semi-normal	231	297	329	285
			Clostridium beijerinckii (strain ATCC 51743 / NCIMB 8052)	Semi-normal	277	369	456	411
			Clostridium botulinum (strain ATCC 19397 / Type A)	Semi-normal	196	249	284	269
			Clostridium difficile (strain 630)	Semi-normal	198	275	317	275
			Clostridium kluyveri (strain ATCC 8527 / DSM 555 / NCIMB 10680)	Semi-normal	253	302	301	250
			Clostridium novyi (strain NT)	Semi-normal	146	190	176	170
			Clostridium perfringens (strain ATCC 13124 / NCTC 8237 / Type A)	Semi-normal	144	244	240	193
			Clostridium phytofermentans (strain ATCC 700394 / DSM 18823 / ISDg)	Semi-normal	178	247	312	281
			Clostridium tetani (strain Massachusetts / E88)	Semi-normal	136	194	220	195
			Clostridium thermocellum (strain ATCC 27405 / DSM 1237)	Semi-normal	213	281	234	219
			Desulfotomaculum reducens (strain MI-1)	Semi-normal	284	301	283	241
			Moorella thermoacetica (strain ATCC 39073)	Semi-normal	257	259	226	185
			Mycoplasma mycoides SC (subsp. mycoides, strain PG1)	Exclusionary	132	90	88	59
			Mycoplasma penetrans (strain HF-2)	Semi-normal	71	86	82	97