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Supplementary Materials for

Global prevalence and distribution of genes and microorganisms involved in mercury methylation

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This PDF file includes:

Other Supplementary Material for this manuscript includes the following:
(available at advances.sciencemag.org/cgi/content/full/1/9/e1500675/DC1)

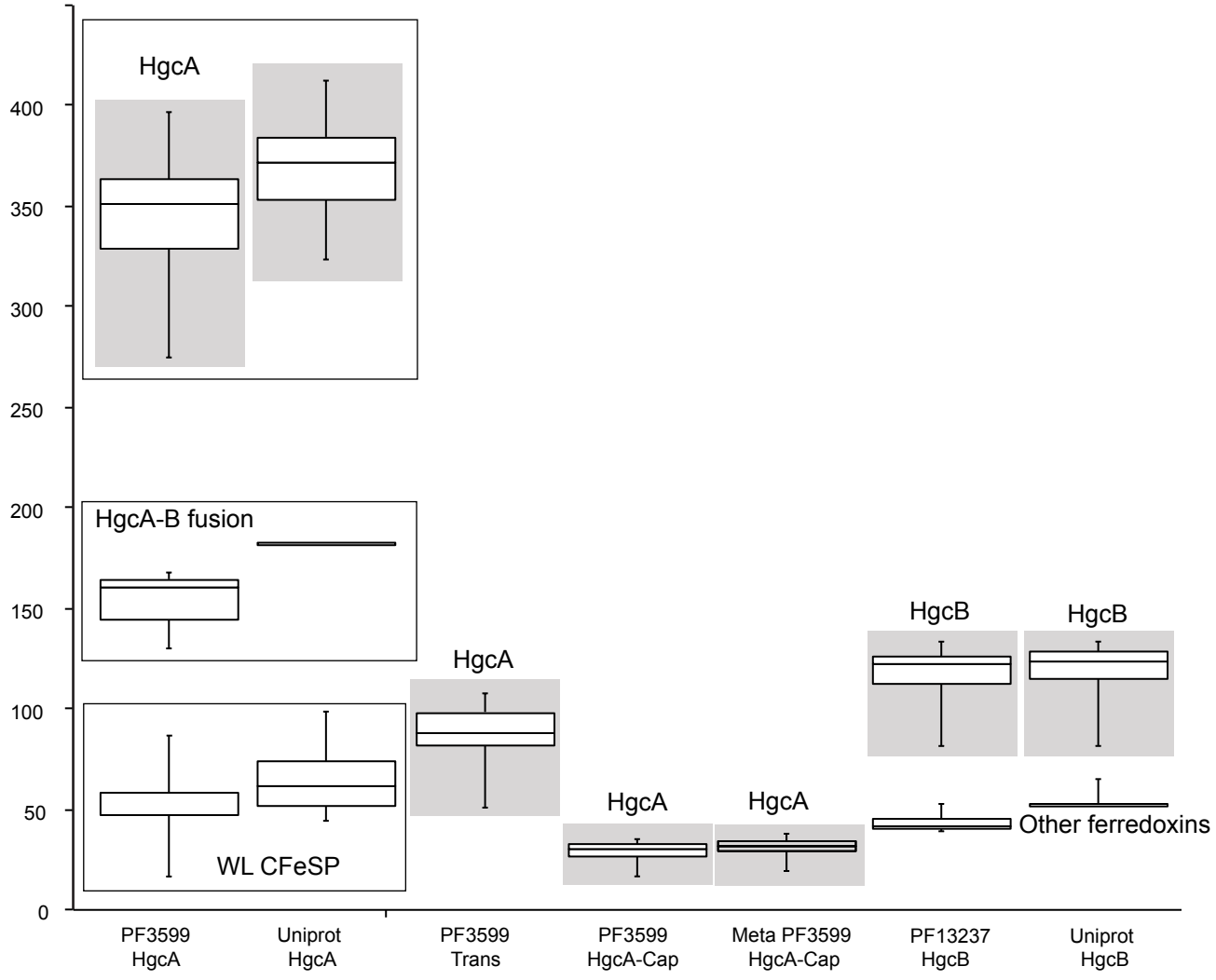


Figure S1. Whisker plot distribution of genomic and metagenomic PFam3599 protein hits to various Hidden Markov profiles (HgcA, HgcA Transmembrane domain, HgcA cap helix domain and HgcB) based on hmmsearch. The Uniprot database was also used as a reference. HgcA and B proteins are grouped in the shaded boxes and have distinct high scores (Y-axis). The other members of the superfamilies (HgcA-B fusion, Wood Lungdahl CFeSP and ferredoxins) have lower scores.

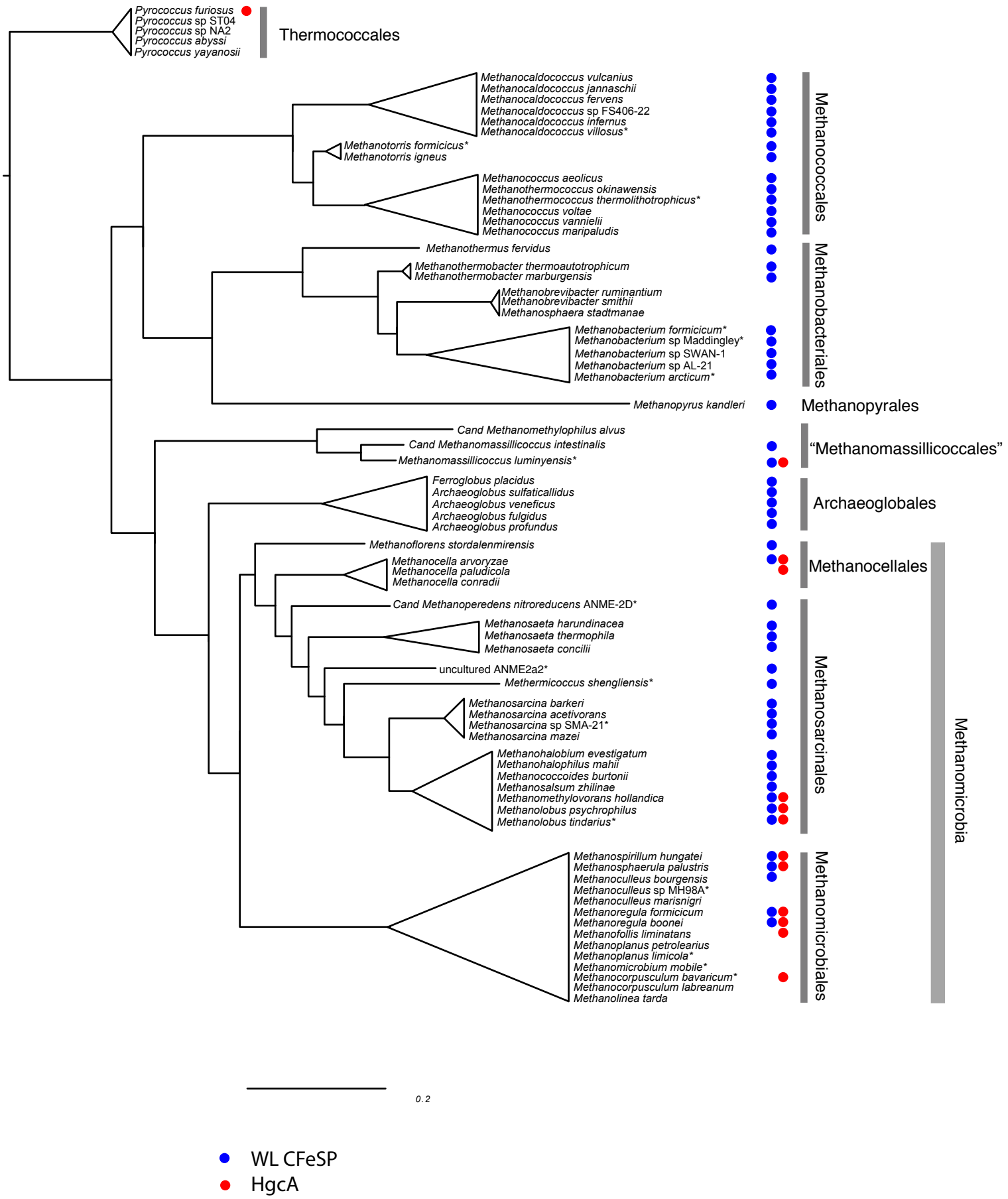


Figure S2. Distribution of HgcA and Wood-Lungdahl CFeSP encoding genes in methanogenic Archaea, relative to their phylogenetic classification based on SSU rRNA gene. The Pyrococcus genus (Thermococcales, Euryarchaeota) was used as outgroup as one of its species encodes an hgcA-B fusion gene.

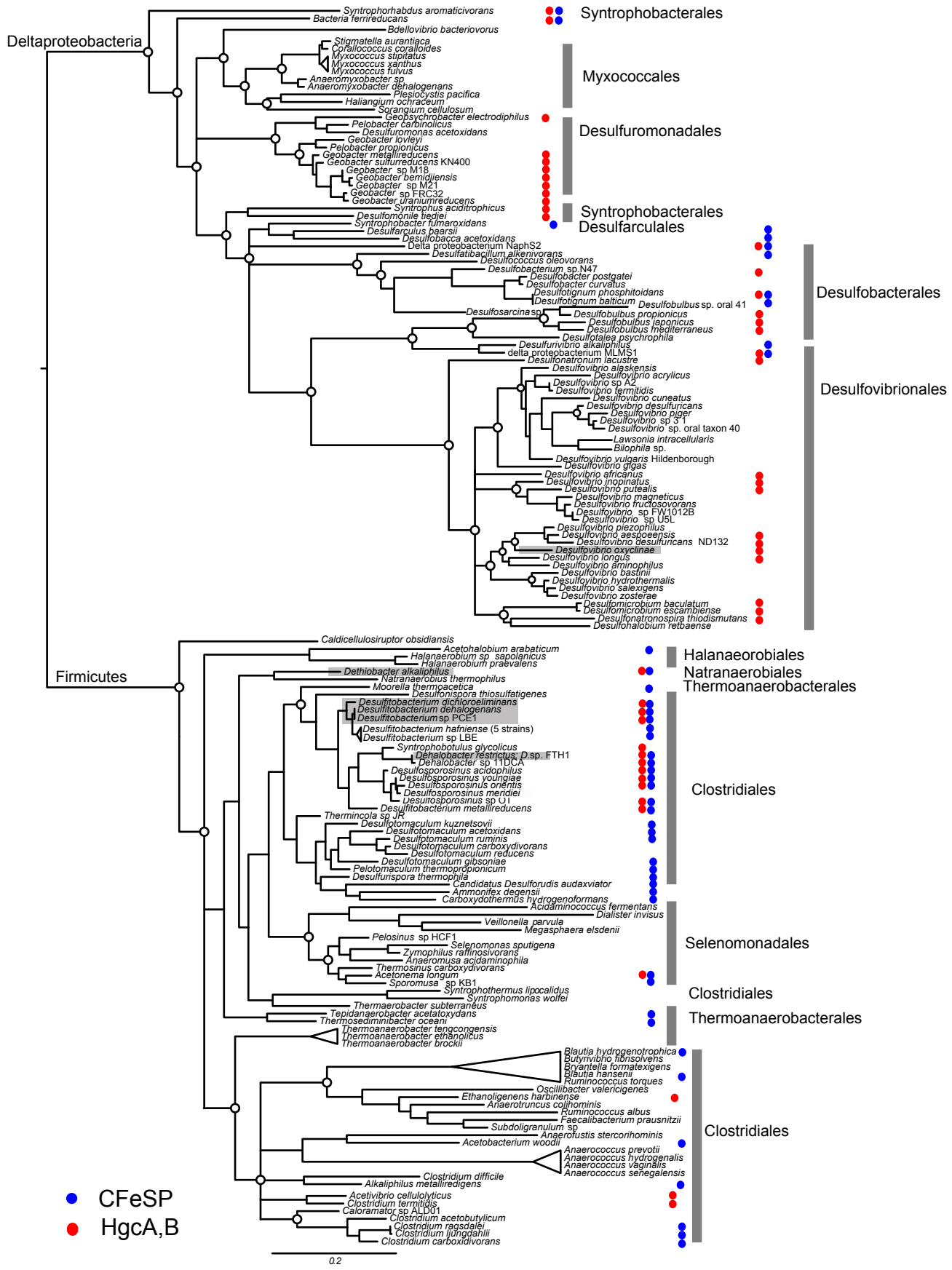


Figure S3. Distribution of HgcA, HgcB and WL CFeSP in genomes of Deltaproteobacteria and Firmicutes relative to other sequenced members of those taxa. The RNA polymerase subunits B-B' sequences were used to construct a maximum likelihood phylogenetic tree (circles at major nodes denote bootstrap supports >50, nodes <50 were collapsed).

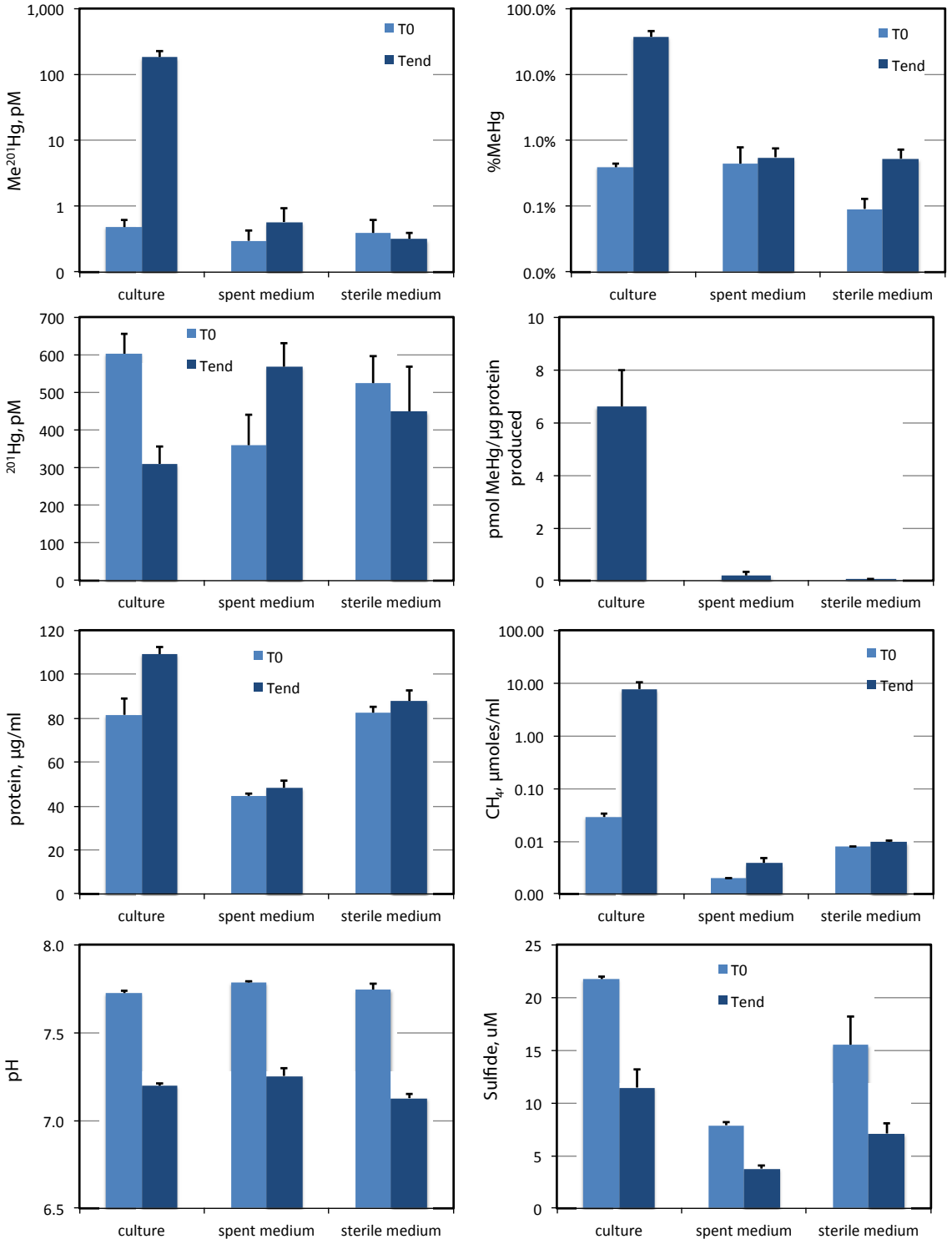


Figure S4. Batch growth mercury methylation assays for Methanomassiliicoccus luminyensis B10 at 37°C. Assays were performed with enriched ^{201}Hg added at 1 nM. All assays and controls were performed in triplicate. Data are shown for bottles at the time of inoculation, and at the end of batch growth (here 142 h). Top left, unfiltered Me^{201}Hg formed from added ^{201}Hg; top right, Me^{201}Hg expressed as a fraction of ^{201}Hg in culture

medium (note log scale); top middle left, unfiltered ^{201}Hg; top middle right, $Me^{201}Hg$ normalized to protein produced; bottom middle left, total protein (medium contains significant protein); bottom middle right, headspace methane normalized to culture volume; bottom left, culture pH; bottom right, sulfide (which was set at 20 μM in medium).

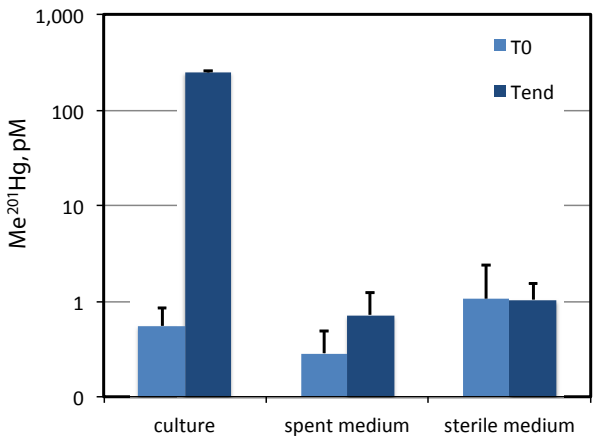
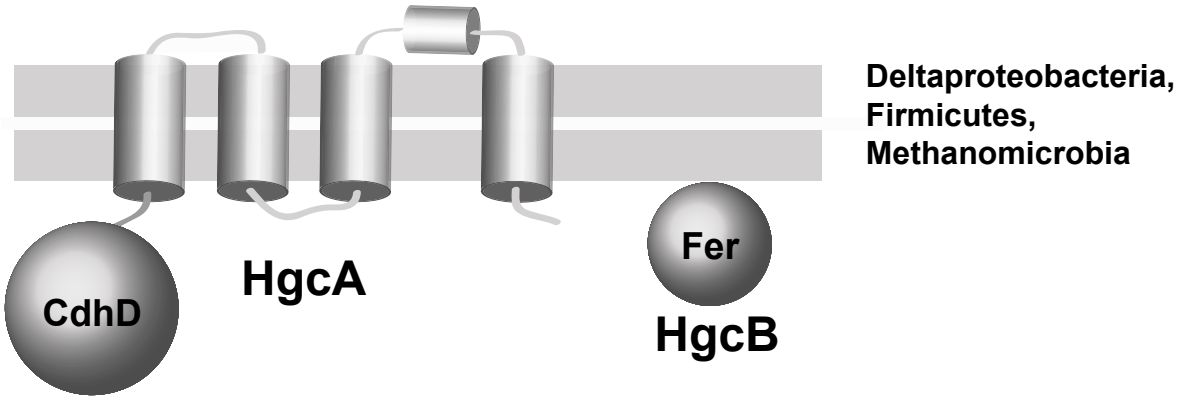
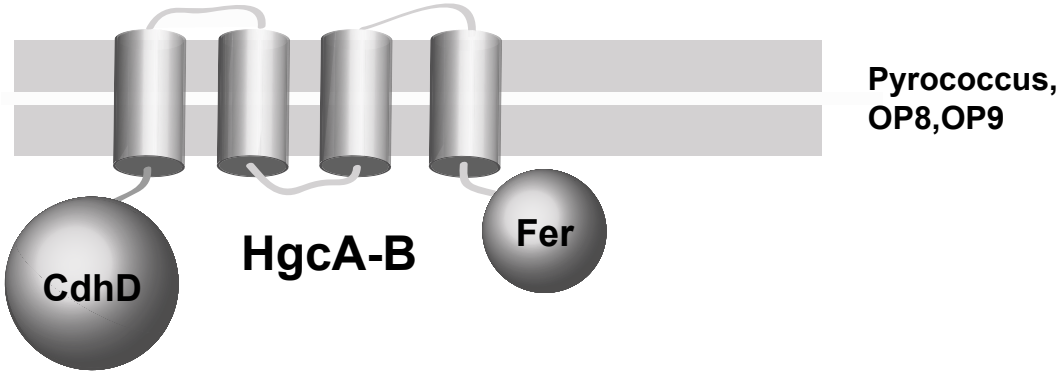


Figure S5. Repeat methylation assay for M. luminyensis, same conditions as Fig SX. Note that log scale was used to better show control concentrations. T_{end} was at 100 h.





CdhD

cap helix

D. desulfuricans ND132 – SPVLVTANYKLTFDTLRERLTSIDAWLLVVDTRGINVWCAAGKGLFT–
Pyrococcus furiosus – SPVFLTGNYCVTVERVRRVLEGIDCYLLVANSRDINVWCSAAGGHFT–
OP8_SCGC_AAA252_F08 – SPVFLTCNYHLTVQRVKWALKGKDAYLLVANSRGINVWCAASGGHFT–
OP9_SCGC_AAA252_M02 – SPVFLTCNFHLTVERVKKALRGIDCYLLIANSKGINVWCAATGGHFT–

D. desulfuricans ND132 – HRELILPQLAATGVAAREVERICGFKVLWGPIRARDLPAFLRNGNKA–
Pyrococcus furiosus – HRNVILPQLAAVGIEARKVREKTGWNVIWGPVYARDIPEFLRNGYKK–
OP8_SCGC_AAA252_F08 – HRKVILPQLAAAGIEARVVHKKSGWRIIWGPVNIKDIQKFLGNRLNK–
OP9_SCGC_AAA252_M02 – HRKITFPQLAASGIEAKTIKNKTGWEIIWGPVYAKDIPLFITNNFKK–

Fer

4Fe-4S 4Fe-4S

D. desulfuricans ND132 – LDTDKCVGCGSCVDVCPHRILAV--RERKTTILDFDACMECGACARNCPV–
Pyrococcus furiosus – LDGERCTGCGVCVDVCPRACYEVDGENHTVMMPRADKCVQCGACIVQCPF–
OP8_SCGC_AAA252 – IDTEKCKGSAFCIDVCPRNCFAVDKSGRTVSIYRAGRCVGCGACIIQCPF–
OP9_SCGC_AAA252 – LDREKCKGIGFCEQVCPRNCFKIDKSRQITTMPGSARCIQCGACIIQCPL–

D. desulfuricans ND132 –EAITVTPGTG--CAAYLVSVWLHRLTGRKIDAACC
Pyrococcus furiosus –EALRFEAPDGRAIPPEIVRRFKLNLMGKRLVRVDEGRV
OP8_SCGC_AAA252_F08 –DAIYFESAEGDRIDPATLREFKLNILGKRSIKVNNR
OP9_SCGC_AAA252_M02 –NALYFKNPQGDIIPPEIIRKFKLNLMGKRREEKNYGIKK

Figure S6. Schematic representation of the domain architecture of HgcA and HgcB and the fusion HgcA-B. The sequence alignment shows conserved regions of the proteins between a representative of the HgcA and HgcB (D. desulfuricans) and three fused HgcA-B

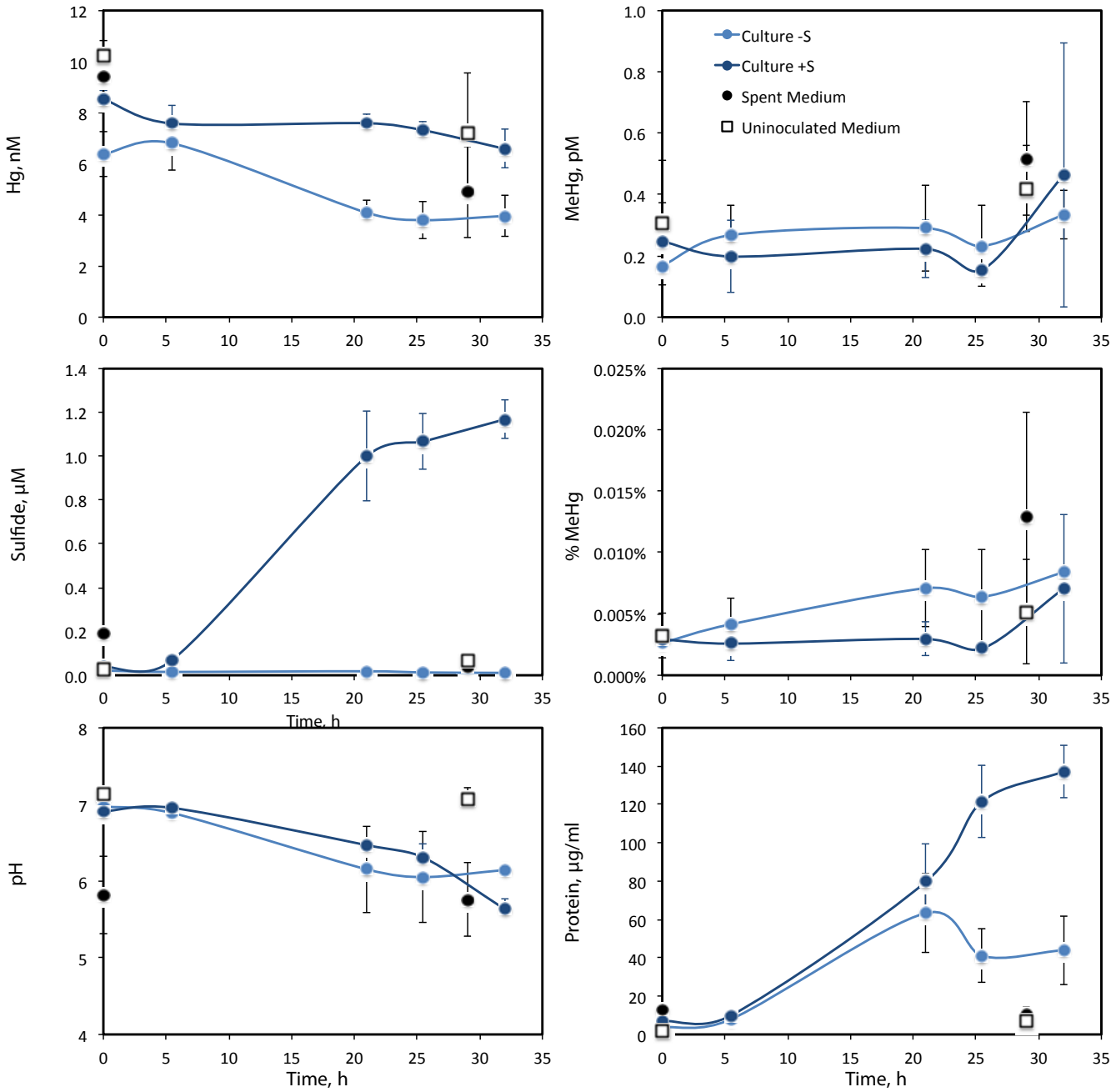


Figure S7. Batch growth mercury methylation assays for Pyrococcus furiosus at 95°C. Assays were performed with enriched ^{201}Hg added at 10 nM. All assays and controls were performed in triplicate. Assays are shown for Medium 377 with and without S^0. Data for uninoculated and spent medium controls are shown as the averages for all six control bottles (3 with and 3 without S^0). Top left, unfiltered excess ^{201}Hg concentration in the medium during growth (and in controls held for the same time); top right, unfiltered $Me^{201}Hg$ formed from added ^{201}Hg; middle left, sulfide produced; middle right, $Me^{201}Hg$ expressed as a fraction of ^{201}Hg in culture medium; bottom left, culture pH; bottom right, protein.

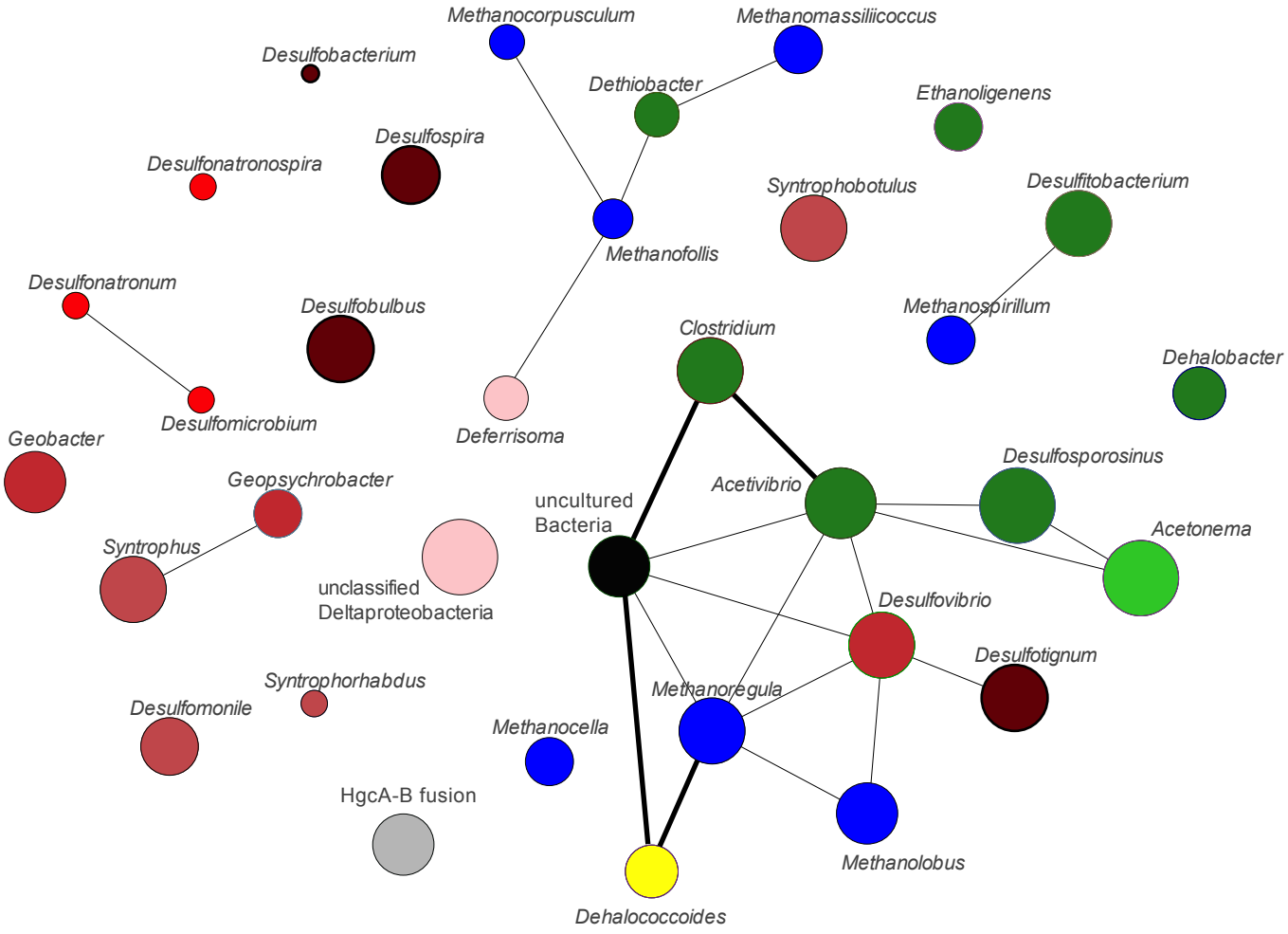


Figure S8. Megan-based co-occurence profiles of closest genera-assigned HgcAs based on all metagenomes. (thin lines indicate co-occurence at 70% probability, thick lines at 80% probability).
Colors are based on the taxonomic assignments, Figure 3.