Sulfur Oxidation Genes in Diverse Deep-Sea Viruses

Karthik Anantharaman,[1] Melissa B. Duhaime,[2] John A. Breier,[3] Kathleen Wendt,[4] Brandy M. Toner,[4] Gregory J. Dick[1,2,5]*

[1]Department of Earth and Environmental Sciences, University of Michigan, Ann Arbor, MI, USA.
[2]Department of Ecology and Evolutionary Biology, University of Michigan, Ann Arbor, MI, USA. [3]Applied Ocean Physics and Engineering, Woods Hole Oceanographic Institution, Woods Hole, MA, USA.
[4]Department of Soil, Water and Climate, University of Minnesota–Twin Cities, St. Paul, MN, USA. [5]Center for Computational Medicine and Bioinformatics, University of Michigan, Ann Arbor, MI, USA.

*Corresponding author. E-mail: gdick@umich.edu

Viruses are the most abundant biological entities in the oceans and a pervasive cause of mortality of microorganisms that drive biogeochemical cycles. Although the ecological and evolutionary impacts of viruses on marine phototrophs are well-recognized, little is known about their impact on ubiquitous marine lithotrophs. Here we report 18 genome sequences of double-stranded DNA viruses that putatively infect widespread sulfur-oxidizing bacteria. Fifteen of these viral genomes contain auxiliary metabolic genes for the alpha and gamma subunits of reverse dissimilatory sulfite reductase (*rdsr*). This enzyme oxidizes elemental sulfur, which is abundant in the hydrothermal plumes studied here. Our findings implicate viruses as a key agent in the sulfur cycle and as a reservoir of genetic diversity for bacterial enzymes that underpin chemosynthesis in the deep oceans.

Chemolithoautotrophic bacteria are ubiquitous in the dark oceans (*1*), where they serve as a sink for CO_2 (*2*) through primary production that equals up to 53% of the particulate organic carbon exported from the photic zone (*3*). Uncultured sulfur-oxidizing bacteria of the SUP05 clade are among the most abundant and widespread marine chemolithoautotrophs, fixing carbon and oxidizing reduced sulfur species and hydrogen in diverse marine environments such as hydrothermal vent plumes (*4*), hydrothermal vent-associated animals (*5*, *6*), and oxygen minimum zones (*7*), where they underpin cryptic links between the sulfur and nitrogen cycles (*8*). Although viruses are abundant in these deep-sea ecosystems (*9*), little is known about viruses that infect lithotrophic primary producers.

We conducted shotgun metagenomic sequencing on samples from five different hydrothermal vent plumes and associated deep ocean waters at the Eastern Lau Spreading Center (Lau Basin) in the Western Pacific Ocean and one plume at Guaymas Basin in the Gulf of California (*10*, *11*) (table S1). De novo assembly of sequence reads and binning by tetranucleotide signatures (*12*) revealed discrete genomic "bins" (fig. S1). Five bins (henceforth Lau77, Lau85, Lau87, Lau218, and Lau220) contained 18 viral genome sequences of putative SUP05 viruses. Phylogeny of the viral large terminase gene (*terL*) (fig. S2) [which reflects phage DNA packaging mechanisms (*13*)], synteny with well-characterized phage of known taxonomy (fig. S3), and results of protein sequence similarity searches against public sequence databases (fig. S4) indicated that the five viruses belonged to three marine viral families of the orders *Caudovirales* (dsDNA viruses, no RNA stage), *Podoviridae*, *Siphoviridae*, and *Myoviridae* (table S2).

Fifteen of the 18 viral genomes (from four of the five SUP05 viral genomic bins) contained genes encoding the alpha (*rdsrA*) and gamma (*rdsrC*) subunits of the reverse-acting dissimilatory sulfite reductase (Rdsr) complex for elemental sulfur oxidation (Fig. 1). No other *rdsr* genes or other sulfur oxidation genes were present on the viral genomes. Analysis of bacterial genome bins recovered from Lau and Guaymas metagenomes revealed co-localized *rdsr* genes in the order *rdsrABE-FHCMKLJOPN* in the Gammaproteobacteria Lau10 (SUP05), Lau62 (EC-01-9C-26), and Lau60 (unclassified). The deltaproteobacterium Lau20 (Sar324) (*14*) possessed only *rdsrAB*. Regions flanking the bacterial *rdsr* genes showed no similarity to the viral genome sequences, suggesting that viral *rdsr* genes were derived from selective retention rather than recent homologous recombination with bacterial genomic DNA.

Phylogenetic analyses indicated that all viral *rdsrA* genes recovered affiliated with SUP05 Gammaproteobacteria (74-96% amino acid identity, fig. S5) and were distinct from *rdsrA* genes of other bacteria (Fig. 2). We identified two distinct groups of *rdsrA* sequences that each included both viral and bacterial sequences. All viral *rdsrA* genes fall into Group one except for Lau85, which contained two copies of *rdsrA* with one representative in each group. Bacterial representatives of Group one included the SUP05 GB-1 and GB-2 from Guaymas as well as *Bathymodiolus* mussel symbionts (*6*). Group two was populated by SUP05 from oxygen minimum zones (*7*) and symbionts of deep-sea clams (*5*). The tight phylogenetic clustering of *rdsrA* gene sequences of three distinct phage families with SUP05 bacteria in two separate lineages suggests that the phage *rdsrA* genes originated from SUP05 and were transferred to viruses. These observations are analogous to those of core photosynthesis genes in cyanobacterial phages and other microbe-derived auxiliary metabolic genes (*15*, *16*) (e.g., *psbA*, *psbD*, *mazG*) that are similar but not identical to known hosts, forming sub-clusters distinct from host proteins (*17*, *18*).

The amino acid sequences deduced from the viral *rdsrA* and *rdsrC* genes indicated the capacity to serve as functional sulfur-oxidizing enzymes. Phage RdsrA contained all conserved sulfite reductase residues and secondary structure elements for α-helix and β-sheets (fig. S6). Similarly, a multiple alignment of RdsrC amino acid sequences indicated highly conserved residues across two distinct groups (fig. S7). We also identified other genes in the viral genomes with high sequence identity to SUP05, including iron-sulfur cluster proteins, cytochromes, and sulfur relay proteins (table S3). The existence of these additional SUP05-like genes on viral genomes supports their specificity to SUP05 bacteria and suggests a role for viral genes in supplementing host metabolism.

Elemental sulfur is a key and stable intermediate of bacterial oxidation of reduced sulfur compounds and constitutes a "bottleneck" in the sulfur cycle (*19*). Sulfur-oxidizing bacteria with an incomplete Sox pathway, such as SUP05, form intracellular or extracellular globules of elemental sulfur that serve as a store of electron donor for energy metabolism (Fig. 3) (*4*, *19–21*). The presence of *rdsrA* and *rdsrC* on viral genomes may offer selective advantages to the viruses by supplementing host pathways for oxidation of this sulfur during infection. First, enhanced expression of *rdsrA* could replenish proteins involved in a rate limiting reaction in the host, as previously demonstrated with cyanobacterial phage D1 proteins involved in photosynthesis (*22*). Second, phage *rdsrC* could maintain or increase high transcription levels to ensure efficient delivery of sulfur-substrate to the RdsrAB complex during infection. Thus, phage auxiliary metabolic genes that can supplement or

sustain sulfur oxidation metabolism in their hosts may ensure continued viral infection and replication.

A coupled bioenergetic-thermodynamic reaction path model (*23*) indicated that aerobic elemental sulfur oxidation potentially accounted for 80-93% of the total lithotrophic energy available in the ABE and Mariner hydrothermal plumes at Lau Basin at two temperatures that were representative of the plumes studied here (Fig. 4A and fig. S8). The model predicted a minor role for 19 other lithotrophic microbial metabolisms considered, including oxidation of other reduced sulfur species such as hydrogen sulfide, sulfite and thiosulfate. These results were consistent with a previous model of hydrothermal plumes that found elemental sulfur to be the most abundant source of chemosynthetic energy (*24*), and with our previous work (*4*, *25*, *26*), which was based on the same approach. Our model predicted greater relative energy yields for more energetically favored metabolisms such as sulfur oxidation because we assumed that metabolic reactions occurred in sequence, from most to least energetically favored, and enabled reactions to explicitly modify the product and reactant pools. Although there is some uncertainty regarding the speciation of sulfur present in the plume, elemental sulfur is a central intermediate in the oxidation of other reduced forms of sulfur used by SUP05 (Fig. 3). X-ray fluorescence maps showed that sulfur was abundant in the plumes (Fig. 4B and fig. S8), while microprobe X-ray diffraction showed elemental sulfur was widely present in particle aggregates with other crystalline phases, such as pyrite (Fig. 4C and fig. S8). Although we did not conclusively identify intracellular elemental sulfur, our results showed that elemental sulfur presented an abundant source of energy for SUP05 bacteria in hydrothermal plumes and deep ocean waters of Guaymas and Lau basins.

The abundance and diversity of viruses infecting SUP05 bacteria in hydrothermal plumes suggests that chemolithoautotrophs in the deep sea face viral predation pressures similar to photosynthetic microbes in the surface waters (*27*). The remarkable synteny and conservation of the four viruses studied here (95-99% genome nucleotide identity) across hydrothermal vent environments and ocean basins suggests that these viruses are ubiquitous in marine environments dominated by SUP05 bacteria. Analyses of the Pacific Ocean Virome dataset (*28*) (fig. S9), which notably contains viral communities from oxygen minimum zones dominated by SUP05 (*7*), revealed the presence of *rdsrA* and *rdsrC* genes (table S5), consistent with the prevalence of phage-encoded sulfur oxidation beyond hydrothermal plumes and in the wider pelagic oceans.

To date, deep-sea SUP05 has evaded growth in laboratory cultures, thus direct host-phage manipulations and validation of the underlying mechanisms of phage-influenced sulfur oxidation remain a challenge. Yet, this study demonstrates the sequence-based elucidation of microbial community dynamics through the discovery of phages that infect a widespread deep-sea bacterium. The existence of *rdsr* genes in viral genomes reveals a mechanism for horizontal transfer of genes associated with sulfur cycling (*29*) and implicates viruses in the evolutionary dynamics of a central step in the planetary cycling of sulfur.

References and Notes
1. B. K. Swan, M. Martinez-Garcia, C. M. Preston, A. Sczyrba, T. Woyke, D. Lamy, T. Reinthaler, N. J. Poulton, E. D. Masland, M. L. Gomez, M. E. Sieracki, E. F. DeLong, G. J. Herndl, R. Stepanauskas, Potential for chemolithoautotrophy among ubiquitous bacteria lineages in the dark ocean. *Science* **333**, 1296–1300 (2011).doi:10.1126/science.1203690
2. J. Arístegui, J. M. Gasol, C. M. Duarte, G. J. Herndl, Microbial oceanography of the dark ocean's pelagic realm. *Limnol. Oceanogr.* **54**, 1501–1529 (2009). doi:10.4319/lo.2009.54.5.1501
3. T. Reinthaler, H. M. van Aken, G. J. Herndl, Major contribution of autotrophy to microbial carbon cycling in the deep North Atlantic's interior. *Deep-Sea Res. II* **57**, 1572–1580 (2010). doi:10.1016/j.dsr2.2010.02.023
4. K. Anantharaman, J. A. Breier, C. S. Sheik, G. J. Dick, Evidence for hydrogen oxidation and metabolic plasticity in widespread deep-sea sulfur-oxidizing

bacteria. *Proc. Natl. Acad. Sci. U.S.A.* **110**, 330–335 (2013). Medline doi:10.1073/pnas.1215340110
5. I. L. Newton, T. Woyke, T. A. Auchtung, G. F. Dilly, R. J. Dutton, M. C. Fisher, K. M. Fontanez, E. Lau, F. J. Stewart, P. M. Richardson, K. W. Barry, E. Saunders, J. C. Detter, D. Wu, J. A. Eisen, C. M. Cavanaugh, The *Calyptogena magnifica* chemoautotrophic symbiont genome. *Science* **315**, 998–1000 (2007).doi:10.1126/science.1138438
6. J. M. Petersen, F. U. Zielinski, T. Pape, R. Seifert, C. Moraru, R. Amann, S. Hourdez, P. R. Girguis, S. D. Wankel, V. Barbe, E. Pelletier, D. Fink, C. Borowski, W. Bach, N. Dubilier, Hydrogen is an energy source for hydrothermal vent symbioses. *Nature* **476**, 176–180 (2011). Medline doi:10.1038/nature10325
7. D. A. Walsh, E. Zaikova, C. G. Howes, Y. C. Song, J. J. Wright, S. G. Tringe, P. D. Tortell, S. J. Hallam, Metagenome of a versatile chemolithoautotroph from expanding oceanic dead zones. *Science* **326**, 578–582 (2009).doi:10.1126/science.1175309
8. D. E. Canfield, F. J. Stewart, B. Thamdrup, L. De Brabandere, T. Dalsgaard, E. F. Delong, N. P. Revsbech, O. Ulloa, A cryptic sulfur cycle in oxygen-minimum-zone waters off the Chilean coast. *Science* **330**, 1375–1378 (2010).doi:10.1126/science.1196889
9. S. Hara, I. Koike, K. Terauchi, H. Kamiya, E. Tanoue, Abundance of viruses in deep oceanic waters. *Mar. Ecol. Prog. Ser.* **145**, 269–277 (1996). doi:10.3354/meps145269
10. G. J. Dick, B. M. Tebo, Microbial diversity and biogeochemistry of the Guaymas Basin deep-sea hydrothermal plume. *Environ. Microbiol.* **12**, 1334–1347 (2010). Medline doi:10.1111/j.1462-2920.2010.02177.x
11. R. A. Lesniewski, S. Jain, K. Anantharaman, P. D. Schloss, G. J. Dick, The metatranscriptome of a deep-sea hydrothermal plume is dominated by water column methanotrophs and lithotrophs. *ISME J.* **6**, 2257–2268 (2012). Medline doi:10.1038/ismej.2012.63
12. G. J. Dick, A. F. Andersson, B. J. Baker, S. L. Simmons, B. C. Thomas, A. P. Yelton, J. F. Banfield, Community-wide analysis of microbial genome sequence signatures. *Genome Biol.* **10**, R85 (2009). Medline doi:10.1186/gb-2009-10-8-r85
13. S. R. Casjens, E. B. Gilcrease, D. A. Winn-Stapley, P. Schicklmaier, H. Schmieger, M. L. Pedulla, M. E. Ford, J. M. Houtz, G. F. Hatfull, R. W. Hendrix, The generalized transducing *Salmonella* bacteriophage ES18: Complete genome sequence and DNA packaging strategy. *J. Bacteriol.* **187**, 1091–1104 (2005). Medline doi:10.1128/JB.187.3.1091-1104.2005
14. C. S. Sheik, S. Jain, G. J. Dick, Metabolic flexibility of enigmatic SAR324 revealed through metagenomics and metatranscriptomics. *Environ. Microbiol.* **16**, 304–317 (2014). Medline doi:10.1111/1462-2920.12165
15. M. Breitbart, L. R. Thompson, C. A. Suttle, M. B. Sullivan, Exploring the vast diversity of marine viruses. *Oceanography* **20**, 135–139 (2007). doi:10.5670/oceanog.2007.58
16. M. Breitbart, Marine viruses: Truth or dare. *Annu. Rev. Mar. Sci.* **4**, 425–448 (2012).doi:10.1146/annurev-marine-120709-142805 Medline
17. J. C. Ignacio-Espinoza, M. B. Sullivan, Phylogenomics of T4 cyanophages: Lateral gene transfer in the "core" and origins of host genes. *Environ. Microbiol.* **14**, 2113–2126 (2012). Medline doi:10.1111/j.1462-2920.2012.02704.x
18. D. Lindell, M. B. Sullivan, Z. I. Johnson, A. C. Tolonen, F. Rohwer, S. W. Chisholm, Transfer of photosynthesis genes to and from *Prochlorococcus* viruses. *Proc. Natl. Acad. Sci. U.S.A.* **101**, 11013–11018 (2004). Medline doi:10.1073/pnas.0401526101
19. W. Ghosh, B. Dam, Biochemistry and molecular biology of lithotrophic sulfur oxidation by taxonomically and ecologically diverse bacteria and archaea. *FEMS Microbiol. Rev.* **33**, 999–1043 (2009). Medline doi:10.1111/j.1574-6976.2009.00187.x
20. D. Hensen, D. Sperling, H. G. Trüper, D. C. Brune, C. Dahl, Thiosulphate oxidation in the phototrophic sulphur bacterium *Allochromatium vinosum*. *Mol. Microbiol.* **62**, 794–810 (2006). Medline doi:10.1111/j.1365-2958.2006.05408.x
21. K. T. Marshall, R. M. Morris, Isolation of an aerobic sulfur oxidizer from the SUP05/Arctic96BD-19 clade. *ISME J.* **7**, 452–455 (2013). Medline doi:10.1038/ismej.2012.78
22. D. Lindell, J. D. Jaffe, Z. I. Johnson, G. M. Church, S. W. Chisholm, Photosynthesis genes in marine viruses yield proteins during host infection. *Nature* **438**, 86–89 (2005). Medline doi:10.1038/nature04111

23. Materials and methods are available as supplementary materials on *Science* Online.

24. T. M. McCollom, Geochemical constraints on primary productivity in submarine hydrothermal vent plumes. *Deep-Sea Res. I* **47**, 85–101 (2000). doi:10.1016/S0967-0637(99)00048-5

25. J. A. Breier, B. M. Toner, S. C. Fakra, M. A. Marcus, S. N. White, A. M. Thurnherr, C. R. German, Sulfur, sulfides, oxides and organic matter aggregated in submarine hydrothermal plumes at 9°50′N East Pacific Rise. *Geochim. Cosmochim. Acta* **88**, 216–236 (2012). doi:10.1016/j.gca.2012.04.003

26. M. Li, B. M. Toner, B. J. Baker, J. A. Breier, C. S. Sheik, G. J. Dick, Microbial iron uptake as a mechanism for dispersing iron from deep-sea hydrothermal vents. *Nat. Commun.* **5**, 3192 (2014). Medline doi:10.1038/ncomms4192

27. S. Avrani, O. Wurtzel, I. Sharon, R. Sorek, D. Lindell, Genomic island variability facilitates *Prochlorococcus*-virus coexistence. *Nature* **474**, 604–608 (2011). Medline doi:10.1038/nature10172

28. B. L. Hurwitz, M. B. Sullivan, The Pacific Ocean virome (POV): A marine viral metagenomic dataset and associated protein clusters for quantitative viral ecology. *PLOS ONE* **8**, e57355 (2013). Medline doi:10.1371/journal.pone.0057355

29. M. Klein, M. Friedrich, A. J. Roger, P. Hugenholtz, S. Fishbain, H. Abicht, L. L. Blackall, D. A. Stahl, M. Wagner, Multiple lateral transfers of dissimilatory sulfite reductase genes between major lineages of sulfate-reducing prokaryotes. *J. Bacteriol.* **183**, 6028–6035 (2001). Medline doi:10.1128/JB.183.20.6028-6035.2001

30. J. A. Breier, C. G. Rauch, K. McCartney, B. M. Toner, S. C. Fakra, S. N. White, C. R. German, A suspended-particle rosette multi-sampler for discrete biogeochemical sampling in low-particle-density waters. *Deep-Sea Res. I* **56**, 1579–1589 (2009). doi:10.1016/j.dsr.2009.04.005

31. Y. Peng, H. C. M. Leung, S. M. Yiu, F. Y. L. Chin, IDBA-UD: A de novo assembler for single-cell and metagenomic sequencing data with highly uneven depth. *Bioinformatics* **28**, 1420–1428 (2012). Medline doi:10.1093/bioinformatics/bts174

32. D. R. Zerbino, E. Birney, Velvet: Algorithms for de novo short read assembly using de Bruijn graphs. *Genome Res.* **18**, 821–829 (2008). Medline doi:10.1101/gr.074492.107

33. T. Namiki, T. Hachiya, H. Tanaka, Y. Sakakibara, MetaVelvet: An extension of Velvet assembler to de novo metagenome assembly from short sequence reads. *Nucleic Acids Res.* **40**, e155 (2012). Medline doi:10.1093/nar/gks678

34. R. Schmieder, Y. W. Lim, R. Edwards, Identification and removal of ribosomal RNA sequences from metatranscriptomes. *Bioinformatics* **28**, 433–435 (2012). Medline doi:10.1093/bioinformatics/btr669

35. V. M. Markowitz, N. N. Ivanova, E. Szeto, K. Palaniappan, K. Chu, D. Dalevi, I. M. Chen, Y. Grechkin, I. Dubchak, I. Anderson, A. Lykidis, K. Mavromatis, P. Hugenholtz, N. C. Kyrpides, IMG/M: A data management and analysis system for metagenomes. *Nucleic Acids Res.* **36** (suppl. 1), D534–D538 (2008). Medline doi:10.1093/nar/gkm869

36. D. Hyatt, G. L. Chen, P. F. Locascio, M. L. Land, F. W. Larimer, L. J. Hauser, Prodigal: Prokaryotic gene recognition and translation initiation site identification. *BMC Bioinformatics* **11**, 119 (2010). Medline doi:10.1186/1471-2105-11-119

37. S. F. Altschul, W. Gish, W. Miller, E. W. Myers, D. J. Lipman, Basic local alignment search tool. *J. Mol. Biol.* **215**, 403–410 (1990). Medline doi:10.1016/S0022-2836(05)80360-2

38. E. Quevillon, V. Silventoinen, S. Pillai, N. Harte, N. Mulder, R. Apweiler, R. Lopez, InterProScan: Protein domains identifier. *Nucleic Acids Res.* **33** (suppl. 2), W116–W120 (2005). Medline doi:10.1093/nar/gki442

39. Y. Zhao, B. Temperton, J. C. Thrash, M. S. Schwalbach, K. L. Vergin, Z. C. Landry, M. Ellisman, T. Deerinck, M. B. Sullivan, S. J. Giovannoni, Abundant SAR11 viruses in the ocean. *Nature* **494**, 357–360 (2013). Medline doi:10.1038/nature11921

40. M. B. Sullivan, K. H. Huang, J. C. Ignacio-Espinoza, A. M. Berlin, L. Kelly, P. R. Weigele, A. S. DeFrancesco, S. E. Kern, L. R. Thompson, S. Young, C. Yandava, R. Fu, B. Krastins, M. Chase, D. Sarracino, M. S. Osburne, M. R. Henn, S. W. Chisholm, Genomic analysis of oceanic cyanobacterial myoviruses compared with T4-like myoviruses from diverse hosts and environments. *Environ. Microbiol.* **12**, 3035–3056 (2010). Medline doi:10.1111/j.1462-2920.2010.02280.x

41. E. Šimoliūnas, L. Kaliniene, L. Truncaitė, A. Zajančkauskaitė, J. Staniulis, A. Kaupinis, M. Ger, M. Valius, R. Meškys, *Klebsiella* phage vB_KleM-RaK2—A giant singleton virus of the family *Myoviridae*. *PLOS ONE* **8**, e60717 (2013). Medline doi:10.1371/journal.pone.0060717

42. M. Krzywinski, J. Schein, I. Birol, J. Connors, R. Gascoyne, D. Horsman, S. J. Jones, M. A. Marra, Circos: An information aesthetic for comparative genomics. *Genome Res.* **19**, 1639–1645 (2009). Medline doi:10.1101/gr.092759.109

43. R. C. Edgar, MUSCLE: Multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Res.* **32**, 1792–1797 (2004). Medline doi:10.1093/nar/gkh340

44. R. R. Karkhoff-Schweizer, D. P. Huber, G. Voordouw, Conservation of the genes for dissimilatory sulfite reductase from *Desulfovibrio vulgaris* and *Archaeoglobus fulgidus* allows their detection by PCR. *Appl. Environ. Microbiol.* **61**, 290–296 (1995). Medline

45. T. Weissgerber, R. Zigann, D. Bruce, Y. J. Chang, J. C. Detter, C. Han, L. Hauser, C. D. Jeffries, M. Land, A. C. Munk, R. Tapia, C. Dahl, Complete genome sequence of *Allochromatium vinosum* DSM 180^T. *Stand. Genomic Sci.* **5**, 311–330 (2011). Medline doi:10.4056/sigs.2335270

46. C. Dahl, N. M. Kredich, R. Deutzmann, H. G. Trüper, Dissimilatory sulphite reductase from *Archaeoglobus fulgidus*: Physico-chemical properties of the enzyme and cloning, sequencing and analysis of the reductase genes. *J. Gen. Microbiol.* **139**, 1817–1828 (1993). Medline doi:10.1099/00221287-139-8-1817

47. A. Dhillon, S. Goswami, M. Riley, A. Teske, M. Sogin, Domain evolution and functional diversification of sulfite reductases. *Astrobiology* **5**, 18–29 (2005). Medline doi:10.1089/ast.2005.5.18

48. T. F. Oliveira, C. Vonrhein, P. M. Matias, S. S. Venceslau, I. A. Pereira, M. Archer, The crystal structure of *Desulfovibrio vulgaris* dissimilatory sulfite reductase bound to DsrC provides novel insights into the mechanism of sulfate respiration. *J. Biol. Chem.* **283**, 34141–34149 (2008). Medline doi:10.1074/jbc.M805643200

49. A. Stamatakis, RAxML-VI-HPC: Maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics* **22**, 2688–2690 (2006). Medline doi:10.1093/bioinformatics/btl446

50. M. B. Duhaime, A. Wichels, J. Waldmann, H. Teeling, F. O. Glöckner, Ecogenomics and genome landscapes of marine Pseudoalteromonas phage H105/1. *ISME J.* **5**, 107–121 (2011). Medline doi:10.1038/ismej.2010.94

51. M. J. Mottl, J. S. Seewald, C. G. Wheat, M. K. Tivey, P. J. Michael, G. Proskurowski, T. M. McCollom, E. Reeves, J. Sharkey, C.-F. You, L.-H. Chan, T. Pichler, Chemistry of hot springs along the Eastern Lau Spreading Center. *Geochim. Cosmochim. Acta* **75**, 1013–1038 (2011). doi:10.1016/j.gca.2010.12.008

52. T. S. Bowers, K. L. Von Damm, J. M. Edmond, Chemical evolution of mid-ocean ridge hot springs. *Geochim. Cosmochim. Acta* **49**, 2239–2252 (1985). doi:10.1016/0016-7037(85)90224-8

53. D. R. Janecky, W. E. Seyfried Jr., Formation of massive sulfide deposits on oceanic ridge crests: Incremental reaction models for mixing between hydrothermal solutions and seawater. *Geochim. Cosmochim. Acta* **48**, 2723–2738 (1984). doi:10.1016/0016-7037(84)90319-3

54. J. Seewald, T. McCollom, G. Proskurowski, E. Reeves, M. Mottl, J. Sharkey, C. G. Wheat, M. Tivey, Aqueous volatiles in Lau Basin hydrothermal fluids. in *AGU Fall Meeting Abstracts*, vol. 1, p. 0478 (2005).

55. J. A. Brandes, N. Z. Boctor, G. D. Cody, B. A. Cooper, R. M. Hazen, H. S. Yoder Jr., Abiotic nitrogen reduction on the early Earth. *Nature* **395**, 365–367 (1998). Medline doi:10.1038/26450

56. L. D. Talley, Ed., *Hydrographic Atlas of the World Ocean Circulation Experiment (WOCE.)*, (International WOCE Project Office, Southampton, UK, 2007).

57. R. Conrad, W. Seiler, Methane and hydrogen in seawater (Atlantic Ocean). *Deep-Sea Res. I* **35**, 1903–1917 (1988). doi:10.1016/0198-0149(88)90116-1

58. R. F. Weiss, H. Craig, Precise shipboard determination of dissolved nitrogen, oxygen, argon, and total inorganic carbon by gas chromatography. *Deep-Sea Res. Oceanogr. Abstr.* **20**, 291–303 (1973). doi:10.1016/0011-7471(73)90054-5

59. J. W. Johnson, E. H. Oelkers, H. C. Helgeson, SUPCRT92: A software package for calculating the standard molal thermodynamic properties of minerals, gases, aqueous species, and reactions from 1 to 5000 bar and 0 to 1000°C. *Comput. Geosci.* **18**, 899–947 (1992). doi:10.1016/0098-

3004(92)90029-Q

60. H. C. Helgeson, J. M. Delaney, H. W. Nesbitt, D. K. Bird, *Am. J. Sci.* **278-A**, 1 (1978).

61. T. M. McCollom, E. L. Shock, Geochemical constraints on chemolithoautotrophic metabolism by microorganisms in seafloor hydrothermal systems. *Geochim. Cosmochim. Acta* **61**, 4375–4391 (1997). Medline doi:10.1016/S0016-7037(97)00241-X

62. P. J. Saccocia, W. E. Seyfried Jr., The solubility of chlorite solid solutions in 3.2 wt% NaCl fluids from 300–400°C, 500 bars. *Geochim. Cosmochim. Acta* **58**, 567–585 (1994). doi:10.1016/0016-7037(94)90489-8

63. E. L. Shock, H. C. Helgeson, Calculation of the thermodynamic and transport properties of aqueous species at high pressures and temperatures: Correlation algorithms for ionic species and equation of state predictions to 5 kb and 1000°C. *Geochim. Cosmochim. Acta* **52**, 2009–2036 (1988). doi:10.1016/0016-7037(88)90181-0

64. E. L. Shock, H. C. Helgeson, D. A. Sverjensky, Calculation of the thermodynamic and transport properties of aqueous species at high pressures and temperatures: Standard partial molal properties of inorganic neutral species. *Geochim. Cosmochim. Acta* **53**, 2157–2183 (1989). doi:10.1016/0016-7037(89)90341-4

65. E. L. Shock, D. C. Sassani, M. Willis, D. A. Sverjensky, Inorganic species in geologic fluids: Correlations among standard molal thermodynamic properties of aqueous ions and hydroxide complexes. *Geochim. Cosmochim. Acta* **61**, 907–950 (1997). Medline doi:10.1016/S0016-7037(96)00339-0

66. D. A. Sverjensky, E. L. Shock, H. C. Helgeson, Prediction of the thermodynamic properties of aqueous metal complexes to 1000°C and 5 kb. *Geochim. Cosmochim. Acta* **61**, 1359–1412 (1997). Medline doi:10.1016/S0016-7037(97)00009-4

67. R. A. Robie, B. S. Hemingway, J. R. Fisher, in Thermodynamic Properties of Minerals and Related Substances at 298.15 K and 1 Bar (10 Pascals) Pressure and at Higher Temperatures. Bulletin 1452 (U.S. Geological Survey, Reston, VA, 1979), pp. 456.

68. D. D. Wagman *et al.*, The NBS tables of chemical thermodynamic properties: Selected values for inorganic and C1 and C2 organic substances in SI units. *J. Phys. Chem. Ref. Data* **11** (suppl. 2), 392 (1982).

69. H. C. Helgeson, Thermodynamics of hydrothermal systems at elevated temperatures and pressures. *Am. J. Sci.* **267**, 729–804 (1969). doi:10.2475/ajs.267.7.729

70. H. C. Helgeson, D. H. Kirkham, Theoretical prediction of the thermodynamic behavior of aqueous electrolytes at high pressures and temperatures; II, Debye-Huckel parameters for activity coefficients and relative partial molal properties. *Am. J. Sci.* **274**, 1199–1261 (1974). doi:10.2475/ajs.274.10.1199

71. C. M. Bethke, *Geochemical and Biogeochemical Reaction Modeling* (Cambridge Univ. Press, Cambridge, ed. 2, 2007).

72. J. S. Cleverley, E. N. Bastrakov, K2GWB: Utility for generating thermodynamic data files for The Geochemist's Workbench® at 0–1000°C and 1–5000bar from UT2K and the UNITHERM database. *Comput. Geosci.* **31**, 756–767 (2005). doi:10.1016/j.cageo.2005.01.007

73. S. E. Drummond, thesis, Pennsylvania State University (1981).

74. M. A. Marcus, A. A. MacDowell, R. Celestre, A. Manceau, T. Miller, H. A. Padmore, R. E. Sublett, Beamline 10.3.2 at ALS: A hard x-ray microprobe for environmental and materials sciences. *J. Synchrotron Radiat.* **11**, 239–247 (2004). Medline doi:10.1107/S0909049504005837

75. A. P. Hammersley, S. O. Svensson, M. Hanfland, A. N. Fitch, D. Hausermann, Two-dimensional detector software: From real detector to idealised image or two-theta scan. *High Press. Res.* **14**, 235–248 (1996). doi:10.1080/08957959608201408

76. V. Sangal, P. C. Fineran, P. A. Hoskisson, Novel configurations of type I and II CRISPR-Cas systems in *Corynebacterium diphtheriae*. *Microbiology* **159**, 2118–2126 (2013). Medline doi:10.1099/mic.0.070235-0

77. B. L. Hurwitz, S. J. Hallam, M. B. Sullivan, Metabolic reprogramming by viruses in the sunlit and dark ocean. *Genome Biol.* **14**, R123 (2013). Medline doi:10.1186/gb-2013-14-11-r123

78. K. L. Von Damm, J. M. Edmond, C. I. Measures, B. Grant, Chemistry of submarine hydrothermal solutions at Guaymas Basin, Gulf of California. *Geochim. Cosmochim. Acta* **49**, 2221–2237 (1985). doi:10.1016/0016-7037(85)90223-6

Supplementary Materials

www.sciencemag.org/cgi/content/full/science.1252229/DC1
Materials and Methods
Supplementary Text
Figs. S1 to S10
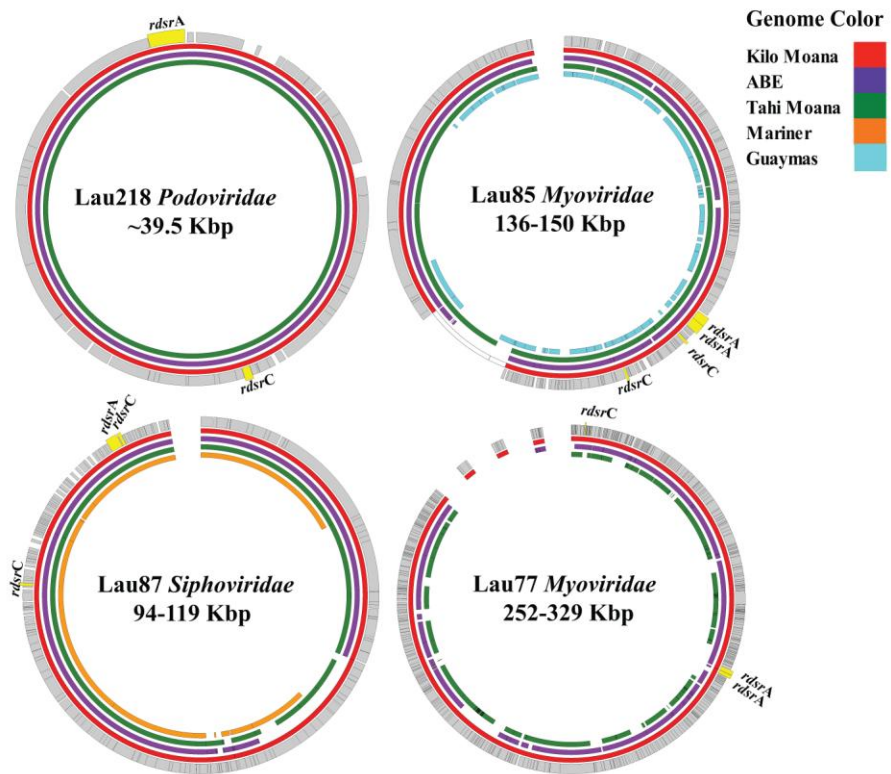Tables S1 to S6
References (*30–78*)
Data S1

Fig. 1. Gene content of 15 phage genomes from three viral families retrieved from Lau and Guaymas basins. Colored nested circles represent syntenous viral genomes/contiguous genomic fragments from locations indicated in the legend. Gray boxes on outermost circle indicate predicted genes from the Kilo Moana viruses. *rdsrA* and *rdsrC* genes are highlighted in yellow.
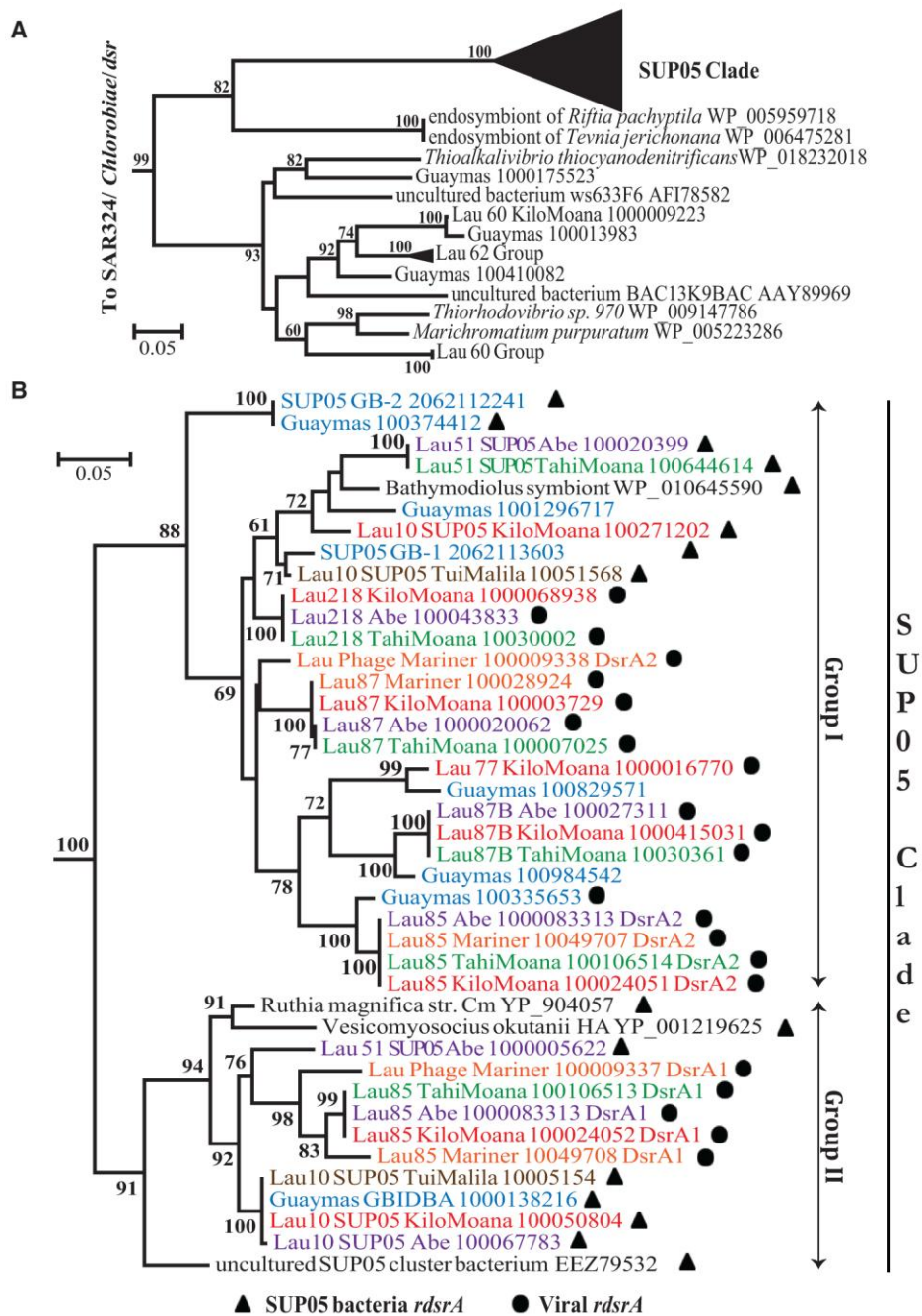
Fig. 2. Phylogeny of *rdsrA* genes from Lau and Guaymas basins inferred by Maximum Likelihood. (**A**) Phylogenetic tree showing the SUP05 *rdsrA* gene clade in relation to closely related sequences. (**B**) Detailed view of the SUP05 *rdsrA* clade. Group one and Group two sub-clades are shown on the right. Sequences are colored by geographical origin; Blue – Guaymas Basin; Red – Kilo Moana (Lau Basin); Green – Tahi Moana (Lau Basin); Purple – ABE (Lau Basin); Brown – Tui Malila (Lau Basin); Orange – Mariner (Lau Basin).
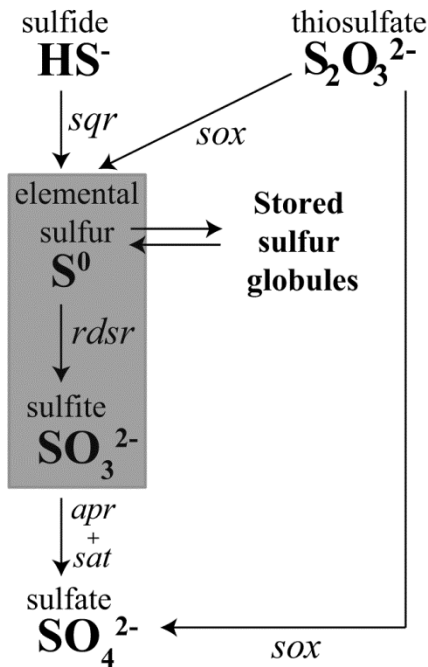
Fig. 3. Schematic of the sulfur oxidation pathway in SUP05 bacteria. Gray box indicates the reaction impacted by SUP05 viruses. Key genes are shown in italics: *sqr* (sulfide quinone reductase), *sox* (sulfur oxidation), *rdsr* (reverse dissimilatory sulfite reductase), *apr* (adenosine 5′-phosphosulfate reductase), and *sat* (sulfate adenylyltransferase).

Fig. 4. Bioenergetics and occurrence of sulfur in the ABE hydrothermal plume. (**A**) Modeled free energies of catabolic reactions as a percentage of total available free energy in the ABE hydrothermal plume at 2.5° and 5°C. Total available free energy in the plume is normalized per kilogram plume fluid (p.f.) and per kilogram vent fluid (v.f.). (**B**) Distribution of iron (displayed in red) and sulfur (displayed in green) in particles collected at 0.5 m above the ABE vent. Locations where elemental sulfur was detected by micro-probe X-ray diffraction measurements are indicated as spots 0, 1, 2, and 19. (**C**) Radially integrated diffractograms with elemental sulfur peaks annotated (*) at 22.0, 25.7, and 34.1 degrees 2-theta.