Sulfur metabolisms in epsilon- and gamma-Proteobacteria in deep-sea hydrothermal fields

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In deep-sea hydrothermal systems, super hot and reduced vent fluids from the seafloor blend with cold and oxidized seawater. Very unique and dense ecosystems are formed within these environments. Many molecular ecological studies showed that chemosynthetic epsilon- and gamma-Proteobacteria are predominant primary producers in both free-living and symbiotic microbial communities in global deep-sea hydrothermal fields. Inorganic sulfur compounds are important substrates for the energy conservative metabolic pathways in these microorganisms. Recent genomic and metagenomic analyses and biochemical studies have contributed to the understanding of potential sulfur metabolic pathways for these chemoautotrophs. Epsilon-Proteobacteria use sulfur compounds for both electron-donors and -acceptors. On the other hand, gamma-Proteobacteria utilize two different sulfur-oxidizing pathways. It is hypothesized that differences between the metabolic pathways used by these two predominant proteobacterial phyla are associated with different ecophysiological strategies; extending the energetically feasible habitats with versatile energy metabolisms in the epsilon-Proteobacteria and optimizing energy production rate and yield for relatively narrow habitable zones in the gamma-Proteobacteria.

Keywords: deep-sea hydrothermal vents, energy metabolism, chemoautotroph

INTRODUCTION

The deep-sea hydrothermal system is one of the most extreme environments on Earth. It is characterized by darkness, high pressures, and steep physical and chemical gradients formed in mixing zones between hot hydrothermal vent fluid and cold deep-sea water. Very unique and dense communities of invertebrates are sustained in absence of a photosynthetic energy source (Dubilier et al., 2008). Almost all vent-endemic animals are strongly associated with the primary production of the endo- and/or epibenthonic chemoautotrophic microorganisms (Jeannin, 2000). With the discovery of deep-sea hydrothermal ecosystem in 1977, it had been proposed that hydrogen sulfide-oxidizing and oxygen-reducing chemoautotrophs potentially sustain the primary production in these unique ecosystems (Kvenvolden et al., 1979). However, anoxic hydrothermal fluids contain several reduced compounds such as H₂, CH₄, and reduced metal ions in addition to H₂S (Jannasch and Mottl, 1985). Recent cultivation studies have demonstrated that these chemicals are all used as energy sources for chemoautotrophs (Jannasch and Mottl, 1985; Nakagawa et al., 2005; Campbell et al., 2006; Emerson et al., 2007), indicating the great diversity of chemoautotrophic energy metabolic processes in the ecosystems. Hydrogen gas (H₂) is one of the most important energy sources, and the hydrogen-dependent ecosystems may represent analogs for the earliest biological communities on Earth (Takai et al., 2004, 2006b; Nealon et al., 2005).

Hydrogen sulfide or sulfide is primarily supplied via high temperatures of seawater–rock interactions in the seafloor hydrothermal reaction zones (Jannasch and Mottl, 1985). Thermodynamic modeling indicated that the hydrogen sulfide or sulfide abundantly contained in the hydrothermal fluids represented the dominating energy source in the mesophilic deep-sea vent chemoautotrophic ecosystems (McCollom and Shock, 1997). In addition, partially oxidized inorganic sulfur compounds such as polysulfide, elemental sulfur, and thiosulfate are generated in the in situ mixing zones, and serve as both electron donor and acceptor in a variety of energy metabolisms. The chemical and microbial oxidation and reduction reactions of sulfur compounds probably establish the overall complex sulfur metabolism network in the ecosystem. This article reviews the representative microbial components capable of utilizing the inorganic sulfur compounds as the energy sources in the deep-sea hydrothermal environments and highlights the biochemical and genetic components of their metabolisms. In addition, the possible segregation of different sulfur metabolic pathways and their host chemoautotrophic Proteobacteria associated with the physical and chemical transition in the mixing zones of the deep-sea hydrothermal environments is discussed.

MICROBIAL COMMUNITIES IN DEEP-SEA HYDROTHERMAL FIELDS

Recent culture-independent analyses of the both symbiotic and free-living microbial communities in the various deep-sea hydrothermal environments have revealed a great phylogenetic diversity of Archaea and Bacteria (Takai et al., 2006a); these analyses have also indicated a predominance in biomass of members within the gamma-Proteobacteria and epsilon-Proteobacteria (Stewart et al., 2005; Suzuki et al., 2005; Urakawa et al., 2005; Campbell et al., 2006; Nakagawa and Takai, 2008; Table 1). Based
| Organism, isolation, or observation site | Genome accession number*1 | Growth temperature (°C) | Electron donor | Electron acceptor | Genes | Locus tag | Pathways |
|----------------------------------------|--------------------------|-------------------------|----------------|------------------|-------|-----------|----------|
| **EPSILON-PROTEOBACTERIA**             |                          |                         |                |                  |       |           |          |
| Sulfurovum sp. NBC37-1 Sediment, Mid-Okinawa Trough | AP009179 | 30–37 | H₂, S⁰, S₂O₅²⁻ | S⁰, NO₃⁻, O₂ | psrACB | SUN0510-0508 | Sulfur respiration | Nakagawa et al. (2005, 2007) |
|                                         |                          |                         |                |                  |       |           |          |
|                                         |                          |                         |                |                  |       | soxXYZAB | Sox system | Yamamoto et al. (2010) |
|                                         |                          |                         |                |                  |       | soxCDYZ  | Sox system    |                      |
|                                         |                          |                         |                |                  |       | soxJ      | Sox system (7) |                      |
|                                         |                          |                         |                |                  |       | sqr       | Sulfide oxidation |                      |
|                                         |                          |                         |                |                  |       | sorAB     | Sulfite oxidation |                      |
|                                         |                          |                         |                |                  |       |           |          |
| Nitratiruptor sp. SB155-2 Sediment, Mid-Okinawa Trough | AP009178 | 55 | H₂, S⁰, S₂O₅²⁻ | S⁰, NO₃⁻, O₂ | psrACB | NIS0573-0571 | Sulfur respiration | Nakagawa et al. (2005, 2007) |
|                                         |                          |                         |                |                  |       |           |          |
|                                         |                          |                         |                |                  |       | soxXYZAB | Sox system    |                      |
|                                         |                          |                         |                |                  |       | soxYZ     | Sox system    |                      |
|                                         |                          |                         |                |                  |       | soxJ      | Sox system (7) |                      |
|                                         |                          |                         |                |                  |       | sqr       | Sulfide oxidation |                      |
|                                         |                          |                         |                |                  |       |           |          |
| Nautilia profundicola AmH Alvinella tube, East Pacific Rise | CP001279 | 45 | H₂, formate | S⁰ | psrABC | NAMH1518-1520 | Sulfur respiration | Campbell et al. (2001, 2009) |
|                                         |                          |                         |                |                  |       |           |          |
| Sulfurimonas autotrophica DSM16294 Sediment, Mid-Okinawa Trough | CP002205 | 25 | H₂S, S⁰, S₂O₅²⁻ | O₂ | psrACB | Saut1622-1644 | Sulfur respiration | Inagaki et al. (2003) |
|                                         |                          |                         |                |                  |       |           |          |
|                                         |                          |                         |                |                  |       | soxXYZAB | Sox system    |                      |
|                                         |                          |                         |                |                  |       | soxCDYZ  | Sox system    |                      |
|                                         |                          |                         |                |                  |       | soxJ      | Sox system (7) |                      |
|                                         |                          |                         |                |                  |       | sqr       | Sulfide oxidation |                      |
|                                         |                          |                         |                |                  |       | sorAB     | Sulfite oxidation |                      |
|                                         |                          |                         |                |                  |       |           |          |
| Sulfurimonas denitrificans DSM1251 Tidal flat mud, Dutch Wadden Sea² | CP000153 | 25 | S₂O₅²⁻ | NO₃⁻ | psrACB | Suden0500-0498 | Sulfur respiration | Sievert et al. (2008) |
|                                         |                          |                         |                |                  |       |           |          |
|                                         |                          |                         |                |                  |       | soxXYZAB | Sox system    |                      |
|                                         |                          |                         |                |                  |       | soxCDYZ  | Sox system    |                      |
|                                         |                          |                         |                |                  |       | soxJ      | Sox system (7) |                      |
|                                         |                          |                         |                |                  |       | sqr       | Sulfide oxidation |                      |

*1 Genome accession numbers and growth temperatures are from the authors' research. *2 Sulfur metabolism in hydrothermal epsilon- and gamma-Proteobacteria.
### Sulfur metabolism in hydrothermal epsilon- and gamma-Proteobacteria

| Organism | Accession | pH | DO | Redox | Gene(s) | System/Reduction | Reference |
|----------|-----------|----|----|-------|---------|------------------|-----------|
| *Alvinella episymbiont East Pacific Rise* | AAUQ | 10-65 | ND | ND | *psrAB* | ND | Sulfur respiration | Grzymski et al. (2008) |
| | | | | | *soxXYZABCD* | ND | Sox system |
| | | | | | *soxJ* | ND | Sox system (?) |

#### GAMMA-PROTEOBACTERIA

| Organism | Accession | pH | DO | Redox | Gene(s) | System/Reduction | Reference |
|----------|-----------|----|----|-------|---------|------------------|-----------|
| *Thiomicrospira crunogena* XCL-2 Galapagos Rift vent | CP000109 | 33 | H₂, H₂S, S⁰, O₂ | SiO₂ | *soxXYZA* | Tcr0604-0601 | Sox system | Scott et al. (2006) |
| | | | | | *soxB* | Tcr1549 | Sox system |
| | | | | | *soxCD* | Tcr0156-0157 | Sox system |
| | | | | | *sqr* | Tcr0619 | Sulfide oxidation |
| | | | | | *dsrAB* | COSY0795-0794 | Reverse sulfate reduction |
| | | | | | *aprAB* | COSY0092-0091 | Reverse sulfate reduction |
| | | | | | *sat* | COSY0089 | Reverse sulfate reduction |
| *CoSym Vesicomyosocius symbiont* | AP009247 | ND | ND | ND | *soxXYZA* | COSY0733-0730 | Sox system | Kuwahara et al. (2007) |
| | | | | | *soxB* | COSY 0161 | Sox system |
| | | | | | *soxJ* | COSY0750 | Sox system (?) |
| | | | | | *sqr* | COSY0963 | Sulfide oxidation |
| | | | | | *dsrAB* | COSY0795-0794 | Reverse sulfate reduction |
| | | | | | *aprAB* | COSY0092-0091 | Reverse sulfate reduction |
| | | | | | *sat* | COSY0089 | Reverse sulfate reduction |
| *Ruthia magnifica Calyptogena symbiont* | CP000488 | ND | ND | ND | *soxXYZA* | Rmag0808-0805 | Sox system | Newton et al. (2007) |
| | | | | | *soxB* | Rmag0156 | Sox system |
| | | | | | *soxJ* | Rmag0824 | Sox system (?) |
| | | | | | *sqr* | Rmag1053 | Sulfide oxidation |
| | | | | | *dsrAB* | Rmag0870-0869 | Reverse sulfate reduction |
| | | | | | *aprAB* | Rmag0088-0087 | Reverse sulfate reduction |
| | | | | | *sat* | Rmag0085 | Reverse sulfate reduction |
| *Endoriftia persephone Riftia symbiont* | AASF | ND | ND | ND | *soxXAB*⁴ | ND | Sox system | Robidart et al. (2008) |
| | | | | | *dsrAB* | ND | Reverse sulfate reduction |
| | | | | | *aprAB* | ND | Reverse sulfate reduction |
| | | | | | *sat* | ND | Reverse sulfate reduction |

(Continued)
| Organism, isolation, or observation site | Genome accession number*1 | Growth temperature (°C) | Electron donor | Electron acceptor | Genes | Locus tag | Pathways |
|----------------------------------------|---------------------------|-------------------------|----------------|------------------|-------|-----------|----------|
| *Beggiatoa* spp. Sediment, Baltic Sea  | ABBY                      | ND                      | ND             | ND               | soxXYZAB | ND        | Sox system         |
|                                        | 00000000,                 |                         |                |                  | fccAB    | ND        | Sulfide oxidation |
|                                        | ABBBZ                     |                         |                |                  | sqr      | ND        | Sulfide oxidation |
|                                        | 00000000                  |                         |                |                  | dsrAB    | ND        | Reverse sulfate reduction |
|                                        |                           |                         |                |                  | aprAB    | ND        | Reverse sulfate reduction |
|                                        |                           |                         |                |                  | sat      | ND        | Reverse sulfate reduction |
|                                        |                           |                         |                |                  | dmsABC   | ND        | DMSO respiration  |
|                                        |                           |                         |                |                  | phsABC   | ND        | Thiosulfate respiration |

ND no data, psr, polysulfide reductase; sox, Sox multi enzyme system; sqr, sulfide:quinone oxidoreductase; fcc, flavocytochrome c; dsr, dissimilatory sulfite reductase; apr, adenosine 5′-phosphosulfate reductase; sat, sulfate adenylyltransferase; dms, dimethyl sulfide reductase; phs, thiosulfate reductase.

*1 Deposited in DDBJ/EMBL/GenBank databases.
*2 It is not a hydrothermal field.
*3 For which the metagenome sequence is published.
*4 SoxXAB were detected, but not SoxYZ.
*5 Sequence assemblies were incomplete.
on the culture-dependent characterization and genetic components analyses, these *Proteobacteria* are found to be chemoheterotrophs strongly associated with utilization of inorganic sulfur compounds (Durand et al., 1993; Inagaki et al., 2003, 2004; Takai et al., 2003b, 2006a,c). Therefore, the sulfur-related energy metabolisms in these *Proteobacteria* are important traits for understanding ecology and biogeochemistry in deep-sea hydrothermal vent ecosystems.

**MICROBIAL SULFUR METABOLIC PATHWAYS FOR ENERGY CONSERVATION**

Inorganic sulfur compounds are used as both electron donor and acceptor by microorganisms. Sulfur-oxidation pathways have been well studied biochemically in anaerobic phototrophs (e.g., *Chlorobium* and *Allochromatium*), facultatively chemooautotrophic *Proteobacteria* (e.g., *Acidithiobacillus* and *Paracoccus*) and *Sulfolobales* (e.g., *Sulfolobus* and *Acidianus*; Friedrich, 1998; Kletzin et al., 2004; Friedrich et al., 2005; Frigaard and Dahl, 2009). Bacterial sulfur-oxidation pathways under the neutral pH are generally classified into two different types: (1) reverse sulfate reduction, and (2) Sox multienzyme system. Reverse sulfate reduction is reversal of sulfate reduction pathway, in which sulfide is oxidized to sulfate via sulfite. This pathway uses the same families of enzymes as of the sulfate reduction pathway, such as dissimilatory sulfate reductase (Dsr), adenylylphosphosulfate reductase (Apr), and sulfate adenylyltransferase (Sat; Kappler and Dahl, 2001). Sox multienzyme complex catalyzes oxidation of inorganic sulfur compounds. Four protein components, SoxY, SoxX, SoxB, and SoxCD are required for complete oxidation of sulfide and thiosulfate to sulfate (Complete Sox pathway; Friedrich et al., 2001). However, the sulfur oxidizers that possess the *soxYXAB* genes but not *soxCD* have often been reported (Friedrich et al., 2005). It is considered that SoxCD acts as sulfur dehydrogenase. Sox system without SoxCD is able to oxidize sulfite and sulfone group (-SO₂⁻) in thiosulfate, but not sulfide, elemental sulfur, and sulfane sulfur (-S⁻) in thiosulfate (Friedrich et al., 2001). In addition to these two sulfur-oxidation pathways, several enzymes have been proposed to oxidize inorganic sulfur compounds, including sulfide:quinone oxidoreductase (Sqr), flavocytochrome c (Fcc), and sulfite oxidase (SO), though the exact physiological functions are still debated. Sqr is a single-subunit flavoprotein that catalyzes oxidation of sulfide to elemental sulfur (Shahak and Hauska, 2008; Frigaard and Dahl, 2009). Recent protein structural study divided Sqr family into six groups (Marcia et al., 2010). These different types of Sqr probably have different mechanisms and physiological roles (Chan et al., 2009; Gregersen et al., 2011; Holkenbrink et al., 2011). Fcc also catalyzes oxidation of sulfide to elemental sulfur, but electrons are transferred to the level of cytochrome c (Oh-oka and Blankenship, 2004). Fcc consists of a flavoprotein subunit, FccB, and a c-type cytochrome subunit, FccA (Kusai and Yamanaka, 1973; Reinartz et al., 1998). The *soxE* genes conserved in the *sox* gene clusters are homologous genes of the *fcbAB* genes (Friedrich et al., 2008). The *soxF* gene is a homologous gene of *fcbB*/*soxF*, that does not cluster with a homolog of *fcaA*/*soxE* (Gregersen et al., 2011). SO is a molybdtenum enzyme and catalyzes the direct oxidation of sulfite to sulfate (Kappler and Dahl, 2001). Acidiphilic sulfur-oxidizing bacteria and archaea use different sulfur-oxidation pathways that will not be further described here (see Ghosh and Dam, 2009).

Sulfur-reduction pathways can be classified into two types based on the substrates: (1) sulfur-reduction (sulfur respiration), and (2) sulfate reduction (sulfate respiration). Sulfur-reduction pathways have been intensively studied in gastrointestinal epsilon-*Proteobacterium,* *Wolinella succinogenes* (Wolin et al., 1961; Hedrich et al., 1999). Sulfur-reduction is catalyzed by polysulfide reductase ( Psr), in which the actual substrate of the reaction is polysulfide but not elemental sulfur (Pfenng and Biebel, 1986). Sulfate reducing bacteria are found in several different phylogenetic lines (e.g., delta-*Proteobacteria,* *Clostridia,* *Thermodesulfo bacterium,* and *Nitrospira*; Muyzer and Stams, 2008). Sat, Apr, and Dsr catalyze the reduction of sulfate to sulfide.

**SULFUR METABOLISM IN EPSILON-*PROTEOBACTERIA* IN DEEP-SEA HYDROTHERMAL FIELDS**

The most well known epsilon-*Proteobacteria* are gastrointestinal pathogens such as *Helicobacter* and *Campylobacter*. They are heterotrophic and do not have inorganic sulfur metabolic pathways. The genus *Wolinella* isolated from cattle rumen can reduce elemental sulfur to hydrogen sulfide (Wolin et al., 1961).

There are many reports that chemoautotrophic epsilon-*Proteobacteria* are predominant in sulfidic environments such as deep-sea hydrothermal and subsurface environments (Takai et al., 2004; Campbell et al., 2006; Nakagawa and Takai, 2008). Recently, whole genome sequences of five strains of marine epsilon-*Proteobacteria*, i.e., *Sulfurovum* sp. NBC37-1, *Nitratiruptor* sp. SB155-2, *Nautilia profundicola* AmH, *Sulfurimonas autotrophica* DSM16294 isolated from deep-sea hydrothermal fields, and *S. denitrificans* DSM1251 isolated from a coastal wetland, have been determined (Nakagawa et al., 2007; Sievert et al., 2008; Campbell et al., 2009; Table 1). Moreover, the metabolism of epibiont epsilon-*Proteobacteria* in a polychaete worm was analyzed by metagenomic analysis (Grzymski et al., 2008). In addition, several biochemical studies on sulfur metabolic pathways in deep-sea epsilon-*Proteobacteria* have also been reported (Takai et al., 2005; Yamamoto et al., 2010).

Genetic and enzymatic components characterized by the genomic and biochemical analyses of deep-sea epsilon-*Proteobacteria* have facilitated considerable progress in determining the possible sulfur metabolism pathways of the deep-sea chemoautotrophic epsilon-*Proteobacteria* (Table 1). The deep-sea epsilon-*Proteobacteria* characterized so far possess (1) a hydrogen-oxidizing sulfur respiration pathway using hydrogenase and polysulfide reductase (Psr). And the epsilon-*Proteobacteria* except for *N. profundicola* AmH, possess (2) a sulfur compounds-oxidizing oxygen/nitrate-respiration pathway using the Sox multienzyme system. Both of the Sox systems coupled with and without SoxCD have been found. The *sox* genes are not organized in a single region, but at least in two separated regions of the *soxXYZAB* and *sox(CD)XY* genes. One or more *soxF* gene was conserved in each sox gene cluster. *Sulfurovum* sp., *Nitratiruptor* sp., and *S. autotrophica* have several *sqr* genes. Two sets and one set of the *sorAB* genes coding SO were observed in *Sulfurovum* sp. and *S. autotrophica*, respectively. No gene for sulfur-oxidation pathways was found in *N. profundicola*. In addition to the sulfur metabolic pathways...
pathways, many deep-sea epsilon-\textit{Proteobacteria} are capable of chemoaotrophic growth without sulfur compounds using a third metabolic pathway: (3) a hydrogen-oxidizing oxygen/nitrate-reduction pathway (Figure 1). The bacterial members that can use sulfur compounds as both electron acceptors and donors are identified only in the phyla of epsilon-\textit{Proteobacteria} and \textit{Aquificae}, both of which predominantly inhabit in deep-sea hydrothermal environments. This may provide an important clue into how the versatile energy metabolic pathways are associated with the energetic advantages adapted to the dynamic and transient environmental conditions in the mixing zones of the hydrothermal vent environments.

**SULFUR METABOLISM IN GAMMA-PROTEOBACTERIA IN DEEP-SEA HYDROTHERMAL FIELDS**

Gamma-proteobacterial symbionts with invertebrates have never been isolated from deep-sea hydrothermal vents. Furthermore only a couple of free-living sulfur-oxidizing gamma-\textit{Proteobacteria} have been isolated from deep-sea hydrothermal fields (Kuever et al., 2002; Takai et al., 2006c, 2009). Unculturable gamma-proteobacterial sulfur-oxidizing endosymbionts are classified into three groups based on their host invertebrates: symbionts of (1) \textit{Bathymodiolus} mussels and \textit{Calypgodigena} clams, (2) \textit{Alviniconcha} gastropods, and (3) gastropods (e.g., \textit{Ifremeria}) and tube-worms (Nakagawa and Takai, 2008). In recent years, two genome sequences of sulfur-oxidizing symbionts in \textit{Calypgodigena} have been published (Kuwahara et al., 2007; Newton et al., 2007; Table 1). Moreover, the metabolisms of symbiont in tubeworm were analyzed by metagenomic analysis (Robidart et al., 2008). In addition, a whole genome sequence of free-living sulfur-oxidizing gamma-\textit{Proteobacterium, Thiomicrospira crunogena} XCL-2 was also published (Scott et al., 2006). Putative metabolic pathways in \textit{Beggiatoa} sp. from marine sediments had also analyzed by using incomplete genome sequence (Mußmann et al., 2007). From these genome-based genetic characteristics, we can develop a preliminary outline of representative sulfur-related energy metabolisms in the deep-sea hydrothermal gamma-\textit{Proteobacteria}, which possess two different sulfur-oxidization pathways: (1) the reverse sulfate reduction using Dsr, Apr, and Sat, and (2) the Sox multienzyme system without SoxCD (Figure 1). \textit{T. crunogena} XCL-2, however, is an exception, in which no gene for the reverse sulfate reduction pathway was found. Moreover, this organism has the soxCD genes separated from other soc genes. In contrast, functional genes amplification studies suggested that hydrothermal gamma-\textit{Proteobacteria} use the reverse sulfate reduction pathway, but not the Sox pathway (Hügler et al., 2010). These results indicate that either of the two distinct sulfur-oxidation pathways is dispensable. Both of these pathways require O_{2} as a terminal electron acceptor in most cases. This fact indicates that the relatively reductive environment (\textit{O}_{2}-depleted condition) is inhibitory for the growth of deep-sea chemoaotrophic gamma-\textit{Proteobacteria} due to the limitation of primary electron acceptor despite an increasing abundance of electron-donors (reduced sulfur compounds). Thus, it is predicted that the metabolically habitable spaces of deep-sea chemoaotrotrophic gamma-\textit{Proteobacteria} strictly requiring co-existence of reduced sulfur compounds and \textit{O}_{2} are much more limited than those of deep-sea epsilon-\textit{Proteobacteria} in the mixing zones of hydrothermal environments. Conversely, however, it seems likely that the genomic installation and the biochemical operation of two different sulfur-oxidizing pathways in the deep-sea chemoaotrotrophic gamma-\textit{Proteobacteria} are kinetically advantageous if both the reduced sulfur compounds and \textit{O}_{2} are steadily supplied into the habitats. Particularly for the gamma-proteobacterial sulfur-oxidizing symbionts, the host invertebrates would prepare the metabolically suitable habitats in and around the bodies in the mixing zones of deep-sea hydrothermal environments. (Arp and Childress, 1983; Zal et al., 1998; Numoto et al., 2005). This may be one of the possible rationales for the domination of deep-sea gamma-proteobacterial sulfur-oxidizing chemoaotrophs in symbiotic communities in deep-sea hydrothermal environments. All deep-sea sulfur-oxidizing gamma-\textit{Proteobacteria} possess one \textit{sqr} gene in the genome, except for the metagenome of \textit{ Riftia} symbiont. Therefore, \textit{Sqr} probably has an important role for the sulfur-oxidation in the organisms. It is interesting that genes for sulfur compound (thiosulfate and dimethyl sulfoxide) respiration were found in genome of \textit{Beggiatoa} sp. (Mußmann et al., 2007). This is in accordance with previous results in \textit{Beggiatoa alba} that can reduce stored elemental sulfur to overcome short-term anoxic conditions (Nelson and Castenholz, 1981; Schmidt et al., 1987).

**CONCLUDING REMARKS**

In this review, we described the sulfur-related energy metabolisms of epsilon- and gamma-\textit{Proteobacteria} dominating the microbial
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communities in the global deep-sea hydrothermal environments. Most of the metabolic characteristics summarized in this article are based on the reconstruction of genetic components in the genomes determined for only a few representative species. Further genetic and biochemical variability associated with the sulfur-related energy metabolisms will be clarified in many species of deep-sea hydrothermal vent epsilon- and gamma-Proteobacteria in the future. Surely, the investigation of other inorganic sulfur metabolizing groups of microbial components such as sulfate-reducers (delta-Proteobacteria, Thermodesulfovibrio, and Deferribacteres) and sulfur-reducing thermophiles (Aquificae, Thermococcales, and Desulfovicrococcales) will be important for understanding the biogeochemical processes and energy- and elemental fluxes of sulfur compounds in the deep-sea hydrothermal environments (Reysenbach et al., 2000, 2002; Teske et al., 2002; Takai et al., 2003a). Recently, genetic information has been rapidly accumulating and has been providing the important genetic potential for understanding the complex sulfur-related metabolic networks in deep-sea hydrothermal vent ecosystems. In contrast, enzymatic and biochemical data are highly insufficient to clarify the in vivo and in situ kinetics, operation and control of metabolic pathways. These will be an important focus for future studies.

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www.frontiersin.org September 2011 | Volume 2 | Article 192 | 7
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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Received: 03 March 2011; accepted: 30 August 2011; published online: 20 September 2011.

Citation: Yamamoto M and Takai K (2011) Sulfur metabolism in epsilon- and gamma-Proteobacteria in deep-sea hydrothermal fields. Front. Microbiol. 2:192. doi: 10.3389/fmicb.2011.00192

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