Global occurrence of the bacteria with capability for extracellular reduction of iodate

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The γ-proteobacterium Shewanella oneidensis MR-1 reduces iodate to iodide extracellularly. Both dmsEFAB and mtrCAB gene clusters are involved in extracellular reduction of iodate by S. oneidensis MR-1. DmsEFAB reduces iodate to hypoiodous acid and hydrogen peroxide (H₂O₂). Subsequently, H₂O₂ is reduced by MtrCAB to facilitate DmsEFAB-mediated extracellular reduction of iodate. To investigate the distribution of bacteria with the capability for extracellular reduction of iodate, bacterial genomes were systematically searched for both dmsEFAB and mtrCAB gene clusters. The dmsEFAB and mtrCAB gene clusters were found in three Ferrimonas and 26 Shewanella species. Coexistence of both dmsEFAB and mtrCAB gene clusters in these bacteria suggests their potentials for extracellular reduction of iodate. Further analyses demonstrated that these bacteria were isolated from a variety of ecosystems, including the lakes, rivers, and subsurface rocks in East and Southeast Asia, North Africa, and North America. Importantly, most of the bacteria with both dmsEFAB and mtrCAB gene clusters were found in different marine environments, which ranged from the Arctic Ocean to Antarctic coastal marine environments as well as from the Atlantic Ocean to the Indian and Pacific Oceans. Widespread distribution of the bacteria with capability for extracellular reduction of iodate around the world suggests their significant importance in global biogeochemical cycling of iodine. The genetic organization of dmsEFAB and mtrCAB gene clusters also varied substantially. The identified mtrCAB gene clusters often contained additional genes for multiheme c-type cytochromes. The numbers of dmsEFAB gene cluster detected in a given bacterial genome ranged from one to six. In latter, duplications of dmsEFAB gene clusters occurred. These results suggest different paths for these bacteria to acquire their capability for extracellular reduction of iodate.

KEYWORDS
DmsEFAB, MtrCAB, extracellular reduction of iodate, Ferrimonas, Shewanella, global biogeochemical cycling of iodine
**Introduction**

Iodine (I) is a trace element of both health and environmental significance. As iodine is a crucial component of human thyroid hormones triiodothyronine and thyroxine, iodine deficiency disorders of humans (e.g., goiter and cretinism) are attributed to insufficient intake of iodine, such as drinking of the groundwater with low iodine level (Laurberg et al., 2001; Zimmermann, 2009; Zhang E. et al., 2013; Duan et al., 2016). Excessive intake of iodine, such as drinking of the groundwater with high iodine level, also results in thyroiditis and probably cancer (Laurberg et al., 2001; Andersen et al., 2009; Zhang E. et al., 2013; Duan et al., 2016). Thus, abnormal level of iodine in groundwater affects human health (Wen et al., 2013; Zhang E. et al., 2013; Duan et al., 2016). Furthermore, radioactive iodine-129 ($^{129}$I) is an important risk driver of the Hanford and Savannah River Sites in Washington and South Carolina States, respectively, United States, where $^{129}$I level in groundwater is higher than that for the drinking water standard (Otoška et al., 2011; Zhang S. et al., 2013; Kaplan et al., 2014). Finally, global cycling of iodine impacts air quality and climate (Carpenter et al., 2021).

In environment, iodate (IO$_3^-$) and iodide (I$^-$) are the two dominant species of inorganic iodine. For example, IO$_3^-$ is the major iodine species found in groundwater of the Hanford and Savannah River Sites (Otoška et al., 2011; Zhang S. et al., 2013), while the dominant species found in the groundwater in Datong and Taiyuan Basins, China and North China Plains is I$^-$ (Li et al., 2013; Tang et al., 2013; Zhang E. et al., 2013). Both IO$_3^-$ and I$^-$ are found in oceans where their combined concentration is 0.4–0.5µM (Chance et al., 2014).

Microorganisms play crucial roles in redox transformation of IO$_3^-$ and I$^-$ in environments. I$^-$-oxidizing microorganisms oxidize I$^-$ to molecular iodine (I$_2$), while IO$_3^-$-reducing microorganisms reduce IO$_3^-$ to I$^-$ (Iino et al., 2016; Yamazaki et al., 2020; Reyes-Umana et al., 2022). The enzymes involved in microbial oxidation of I$^-$ to I$_2$ include the extracellular multicopper oxidase LoxA (Suzuki et al., 2012; Shiroyama et al., 2015), while those involved in microbial reduction of IO$_3^-$ to I$^-$ include two different types of enzymes (Yamazaki et al., 2020; Guo et al., 2022; Reyes-Umana et al., 2022; Shin et al., 2022).

IdrABP$_2$, P$_2$ of the dissimilatory IO$_3^-$-reducing bacterium *Pseudomonas* sp. strain SCT is the first enzyme identified for IO$_3^-$ reduction (Yamazaki et al., 2020). This enzyme consists of four subunits, in which IdrA is suggested to be the catalytic subunit, IdrB is the electron transfer subunit and IdrP$_1$ and IdrP$_2$ are the detoxification subunits. All of these subunits are believed to be localized in the periplasm. During IO$_3^-$ reduction, IdrB is proposed to receive electrons from cytochrome c (Cyt-c) in the periplasm and then transfers the electrons to IdrA. IdrA is suggested to use the electrons to reduce IO$_3^-$ to hypooxidous acid (HIO) and hydrogen peroxide (H$_2$O$_2$). The generated H$_2$O$_2$ is proposed to be reduced to H$_2$O by IdrP$_1$ and IdrP$_2$. Cyt-c may also supply electrons to IdrP$_1$ and IdrP$_2$. HIO is suggested to be further reduced to I$^-$ probably by Cl( (Yamazaki et al., 2020). Based on their polypeptide sequences, IdrA and IdrB are the homologs of dimethylsulfoxide (DMSO) reductase DmsA and DmsB, respectively. IdrP$_1$ and IdrP$_2$ are the c-type cytochromes (c-Cyt) that are peroxidases. The genes for IdrABP$_2$, P$_2$ are clustered together in the genome of *Pseudomonas* sp. strain SCT (Yamazaki et al., 2020).

The idrABP$_2$, P$_2$ gene cluster also exists in the genome of the dissimilatory IO$_3^-$-reducing bacterium *Denitromonas* sp. IR-12 (Reyes-Umana et al., 2022). Deletion of idrA gene impairs bacterial ability to grow with IO$_3^-$ as the sole terminal electron acceptor. The proposed functions of IdrABP$_2$, P$_2$ from *Denitromonas* sp. IR-12 in IO$_3^-$ reduction are the same to those proposed for the IdrABP$_2$, P$_2$ of *Pseudomonas* sp. strain SCT except that HIO is proposed to be disproportionate to IO$_3^-$ and I$^-$. The IO$_3^-$ is further reduced by IdrAB (Reyes-Umana et al., 2022). Furthermore, microorganisms with idrABP$_2$, P$_2$ gene cluster are widespread in oceans, suggesting their global significance in biogeochemical cycling of iodine (Reyes-Umana et al., 2022).

The dissimilatory metal-reducing bacterium *Shewanella oneidensis* MR-1 reduces IO$_3^-$ extracellularly via DmsEFAB and MrtCAB (Mok et al., 2018; Toporek et al., 2019; Guo et al., 2022; Shin et al., 2022). The DmsEFAB is the first enzyme demonstrated experimentally for reducing IO$_3^-$ to HIO and H$_2$O$_2$ (Guo et al., 2022). Correspondingly, MrtCAB is also confirmed experimentally to reduce H$_2$O$_2$ to H$_2$O for facilitating DmsEFAB-mediated IO$_3^-$ reduction. The Mrt extracellular electron transfer pathway is suggested to transfer electrons from the cytoplasmic membrane and across the periplasm to the DmsEF and MtrAB in the outer membrane. DmsEF and MtrAB transfer electrons across the outer membrane to DmsAB and MtrC, respectively. On bacterial surface, DmsAB and MtrC work collaboratively to reduce IO$_3^-$ (Guo et al., 2022).

Unlike *Pseudomonas* sp. strain SCT and *Denitromonas* sp. IR-12, *S. oneidensis* MR-1 is not an IO$_3^-$-respiring bacterium (Toporek et al., 2019). In *S. oneidensis* MR-1, DmsEFAB and MrtCAB are for extracellular respiration of DMSO and ferric iron [Fe(III)]-containing minerals, respectively (Beliaev and Saffarini, 1998; Beliaev et al., 2001; Gralnick et al., 2006; Bretschger et al., 2007; Coursolle and Gralnick, 2010). It is believed that their extracellular localization enables DmsEFAB and MtrCAB to reduce IO$_3^-$ collaboratively and extracellular reduction of IO$_3^-$ by *S. oneidensis* MR-1 is a fortunate function (Guo et al., 2022). The non-IO$_3^-$-respiring bacteria with the capability for extracellular reduction of IO$_3^-$ are also believed to impact the fate, transport, and global biogeochemical cycling of iodine (Guo et al., 2022). However, to what extent the bacteria with the capability for extracellular reduction of IO$_3^-$ distribute around the world has never been investigated before.

Like idrABP$_2$, P$_2$ gene cluster, the genes for DmsEFAB and MtrCAB are also clustered together, respectively (Gralnick et al., 2006; Fredrickson et al., 2008; Wang et al., 2008). In this investigation, we searched bacterial genomes for dmsEFAB and mtrCAB gene clusters and found that the bacteria with both dmsEFAB and mtrCAB gene clusters were widespread, showing global distribution...
of the bacteria possessing capability for extracellular reduction of IO$_3^−$. Worldwide occurrence of the bacteria with capability for extracellular reduction of IO$_3^−$ suggests the importance of these bacteria in global biogeochemical cycling of iodine.

Approach

Identification of $dmsEFAB$ and $mtrCAB$ homologs

$DmsE$ and $DmsF$ are homologous to $MtrA$ and $MtrB$, respectively (Gralnick et al., 2006). Thus, we searched microbial genomes for $DmsE/MtrA$ and $DmsF/MtrB$ homologs by the approach that was described before (Shi et al., 2012a, 2014; Zhong and Shi, 2018). The $DmsE$, $DmsF$, $MtrA$, and $MtrB$ of $S. oneidensis$ MR-1 served as templates for identifying microbial open reading frames (ORFs) whose protein sequences shared similarity to the templates by BLAST programs of the National Center for Biotechnology Information (NCBI) and of the Universal Protein Resource (UniProt; $E<0.01$; Altschul et al., 1990), in which scoring matrix = BLOSUM62, gapopen = 0, gapextend = 0 and databases = non-redundant protein sequences database (nr) and UniprotKB database. The in-house Perl scripts and a hidden Markov model-based PRED-TMBB software were used to verify identified homologs with the CX$_2$CH motifs and the trans-outer membrane motifs, respectively (Bagos et al., 2004; Shi et al., 2012b; Shi et al., 2014; Zhong and Shi, 2018). After verification, the identified homologs served as the templates for next round of genome search. The polypeptide sequences from the genes immediately upstream and downstream of the identified $dmsEF/mtrAB$ gene clusters were further compared with previously identified $DmsA$, $DmsB$, and $MtrC$. The identified $dmsEFAB$ gene clusters and the $mtrCAB$ gene clusters co-existed with $dmsEFAB$ gene clusters in the same bacterial genome were subjected to the additional analyses.

Phylogenetic reconstruction and identification of additional genes for c-Cyts

Clustal W (version 2.1) was used to align the polypeptide sequences identified. The parameters used were Gap Opening Penalty = 10; Gap Extension Penalty = 0.2; Protein matrix = BLOSUM series (Larkin et al., 2007). MEGA7 was used to analyze the aligned sequences of $DmsA$, $DmsB$, $DmsE/MtrA$, $DmsF/MtrB$, or $MtrC$ homologs. Phylogenetic trees were constructed with Maximum Likelihood at a confidence level determined by 1,000 bootstrap replications (Kumar et al., 2016). The results of phylogenetic reconstruction were displayed with Evolview v2 (He et al., 2016). The $genes$ for $c$-$Cyts$ on the upstream and downstream of the $mtrCAB$ gene clusters were also identified by the method described above (Shi et al., 2012b, 2014; Zhong and Shi, 2018). The map of distribution for the identified bacteria was constructed similarly to that described previously (Baker et al., 2022).

Results and discussion

Overviews

$DmsE/MtrA$ is a decaheme $c$-$Cyt$ that is inserted into the outer membrane porin protein $DmsF/MtrB$. The function of $DmsF/MtrB$ is to insulate $DmsE/MtrA$ from the outer membrane, which permits rapid electron transfer across the outer membrane by $DmsE/MtrA$ (Hartshorne et al., 2009; White et al., 2013; Edwards et al., 2020). Thus, the identified microorganisms that possessed $dmsEFAB$ and/or $mtrCAB$ gene clusters were all the Gram-negative bacteria. Supplementary Table S1 listed the bacteria identified with both $dmsEFAB$ and $mtrCAB$ gene clusters from this investigation. These included three $Ferrimonas$ and 26 $Shewanella$ species. It should be noted that in $S. oneidensis$ MR-1, $dmsEFAB$ and $mtrCAB$ gene clusters are for extracellular respiration of DMSO and Fe(III)-containing minerals, respectively (Beliaev and Saffarini, 1998; Beliaev et al., 2001; Gralnick et al., 2006; Breitschger et al., 2007; Coursolle et al., 2010). Involvement of $dmsEFAB$ and $mtrCAB$ gene clusters in extracellular reduction of IO$_3^−$ is fortuitous (Guo et al., 2022). Restrictive distribution of the bacteria with capability for extracellular reduction of IO$_3^−$ in $Ferrimonas$ and $Shewanella$ species may be attributed to this fortuitous function.

Possession of both $dmsEFAB$ and $mtrCAB$ gene clusters suggests that these bacteria are capable of reducing IO$_3^−$ extracellularly. This is consistent with the previous observations that in addition to $S. oneidensis$ MR-1, other $Shewanella$ species, such as $S. putrefaciens$, reduced IO$_3^−$ (Councell et al., 1997; Farrenkorf et al., 1997; Mok et al., 2018; Toporek et al., 2019; Guo et al., 2022; Shi et al., 2022).

Global distribution

The original habitats for this group of bacteria with both $dmsEFAB$ and $mtrCAB$ gene clusters varied substantially (Figure 1; Supplementary Table S1). Some of them were isolated from the sediments of the lakes located in China, India, and the United States (Myers and Nealson, 1988; Li et al., 2014; Rathour et al., 2021); city drainage in Vietnam (Diao et al., 2022); subsurface rock in United States (Fredrickson et al., 1998), river water in Tunisia and rainbow trout in South Korea (Figure 1; Supplementary Table S1). Notably, >82% of the identified bacteria with $dmsEFAB$ and $mtrCAB$ gene clusters were isolated from a variety of marine environments around the world. These included the costal sediments in Mallorca, Spain; Xiamen, China, and Nova Scotia, Canada (Rossello-Mora et al., 1995; Zhao et al., 2005; Huang et al., 2010); sediments in Ross Sea, South China Sea, and Arctic Ocean.
Ivanova et al., 2003; Hwang et al., 2019; Li et al., 2021); a cold seep field in South China Sea (Figure 1; Supplementary Table S1) and deep-sea sediments in Southwest Indian Ocean and West Pacific Ocean (Wang et al., 2004, 2021; Yu et al., 2021). Some were also found in seawater in East Sea, Korea; North Sea, United Kingdom; Troitsa Bay, Russia; Alboran Sea and Black Sea (Makemson et al., 1997; Reid and Gordon, 1999; Venkateswaran et al., 1999; Kim et al., 2017; Bae et al., 2021) as well as sea ice floe close to Point Barrow, Alaska, United States (Figure 1; Supplementary Table S1); the gastric cavity of galaxy coral in the coastal area near Hainan Island, China (Tang et al., 2020), and the intestines of sea animals in Japan (Satomi et al., 2003). Thus, bacteria with both dmsEFAB and mtrCAB gene clusters occur globally. Distribution of the bacteria with dmsEFAB and mtrCAB gene clusters is also comparable to the distribution of bacteria with porin-cytochrome genes (Baker et al., 2022). Widespread occurrence of these bacteria around the world, especially their distribution in a variety of marine environments, suggests significant importance of the bacteria with capability for extracellular reduction of IO$_3^-$ in global biogeochemical cycling of iodine.

### DmsA and DmsB homologs

A total of 52 DmsA homologs were identified from the bacteria with dmsEFAB and mtrCAB gene clusters (Figure 2A; Supplementary Table S2). For comparison, IdrA of Pseudomonas sp. strain SCT and Denitromonas sp. IR-12 were included for phylogenetic analyses (Yamazaki et al., 2020; Reyes-Umana et al., 2022). Like DmsA and SO$_4^{3-}$ of S. oneidensis MR-1, all the identified DmsA homologs possessed the twin-arginine sequence at their N-termini for their secretion to the periplasm via the twin-arginine protein secretion system (Supplementary Figure S1; Gralnick et al., 2006). The identified DmsA homologs were 35–98% identical to DmsA of S. oneidensis MR-1. Notably, SO$_4^{3-}$ of S. oneidensis MR-1 was 36% identical to DmsA of S. oneidensis MR-1 (Supplementary Table S2). SO$_4^{3-}$ is not involved in extracellular reduction of iodate by S. oneidensis MR-1 (Guo et al., 2022). The identified DmsA homologs were distantly related to IdrA of Pseudomonas sp. strain SCT and Denitromonas sp. IR-12 (Figure 2A; Supplementary Table S2).

The twin-arginine sequence was also detected in the DmsB homologs identified (Supplementary Figure S2; Gralnick et al., 2006). These DmsB homologs were 52–99% identical to DmsB of S. oneidensis MR-1. Among them, SO$_4^{3-}$ of S. oneidensis MR-1 was 61% identical to DmsB of S. oneidensis MR-1 (Supplementary Table S3). The identified DmsB homologs were 11–24% identical to IdrB of Pseudomonas sp. strain SCT and Denitromonas sp. IR-12 (Figure 2B; Supplementary Table S3).

### DmsE/MtrA/MtrD and DmsF/MtrB/MtrE homologs

In S. oneidensis MR-1, MtrDEF are homologous to MtrABC, respectively, and mtrDEF genes are clustered together and are part of the mtrABC operon. In S. oneidensis MR-1, DmsE and MtrA are homologous to MtrC and DmsA, respectively, and dmsEFAB genes are clustered together. DmsF and MtrD are homologous to MtrB and DmsB, respectively, and mtrCAB genes are clustered together.
of mtrCAB gene cluster (Fredrickson et al., 2008). However, the mtrDEF gene cluster is not involved in extracellular reduction of IO$_3^-$ by S. oneidensis MR-1 (Guo et al., 2022). Ninety-seven DmsE/MtrA/MtrD and DmsF/MtrB/MtrE homologs were identified, respectively, from the bacteria with dmsEFAB and mtrCAB gene cluster (Figures 2C,D; Supplementary Tables S4, S5). Identified DmsE/MtrA/MtrD homologs were 47%–98% identical to DmsE/MtrA/MtrD of S. oneidensis MR-1 (Supplementary Table S4), respectively; while the identified DmsF/MtrB/MtrE homologs were 24%–96% identical to DmsF/MtrB/MtrE of S. oneidensis MR-1, respectively (Supplementary Table S5). These DmsE/MtrA/MtrD and DmsF/MtrB/MtrE homologs were 32%–43% and 14%–23% identical to MtoA/PioA and MtoB/PioB of the Fe(II)-oxidizing bacteria Sideroxydans lithotrophicus ES-1 and Rhodopseudomonas palustris TIE-1 (Figures 2C,D; Supplementary Tables S4, S5). Previous results showed that MtoA/PioA and MtoB/PioB of S. lithotrophicus ES-1 were homologous to MtrA and MtrB of S. oneidensis MR-1, respectively (Jiao and Newman, 2007; Shi et al., 2012b; Liu et al., 2013). Purified MtoA of S. lithotrophicus ES-1 was capable of oxidizing Fe(II), including the solid phase Fe(II) (Liu et al., 2012, 2013). PioA and PioB were also involved in extracellular oxidation of Fe(II) by R. palustris TIE-1 (Jiao and Newman, 2007). Given that they are more homologous to DmsE/MtrA and DmsF/MtrB of S. oneidensis MR-1 than to MtoA/PioA and MtoB/PioB of S. lithotrophicus ES-1 and R. palustris TIE-1, the identified DmsE/MtrA/MtrD and DmsF/MtrB/MtrE homologs are most likely to mediate electron transfer from inside cells to outside cells.

**MtrC/MtrF homologs**

Forty-five MtrC/MtrF homologs were identified from the bacteria with dmsEFAB and mtrCAB gene clusters (Figure 2E; Supplementary Figure S3; Supplementary Table S6). These homologs were 29%–71% identical to MtrC of S. oneidensis MR-1 (Supplementary Table S6). MtrC of S. oneidensis MR-1 is a lipoprotein with 10 c-type hemes and is located on the bacterial surface (Shi et al., 2006, 2008; Lower et al., 2009; Edwards et al., 2020). It contains a lipid-binding site in its N-terminus and replacement of this site renders MtrC unable to bind to the outer membrane (Edwards et al., 2015). Similar to MtrC of S. oneidensis MR-1, all identified homologs possessed this lipid-binding site in their N-termini (Supplementary Figure S3). Thus, these MtrC/MtrF homologs are most likely on the bacterial cell surface. All the identified MtrC/MtrF homologs also possessed 10 c-type heme-binding sites (Supplementary Figure S3). Similar to MtrC and other c-Cyts, these MtrC homologs should possess intrinsic peroxidase activity to degrade the H$_2$O$_2$ formed from extracellular reduction of IO$_3^-$ (Thomas et al., 1976; Shi et al., 2006; Guo et al., 2022).
Genetic organization

Further investigation revealed that the numbers of dmsEFAB gene cluster and genes associated with mtrCAB gene cluster varied among the genomes of identified bacteria. Because of these differences, the identified bacteria could be categorized into seven different groups (Supplementary Table S1; Figure 3).

Group I bacteria contained a dmsEFAB gene cluster and a mtrCAB gene cluster. An additional gene for the outer membrane multiheme c-Cyt is also associated with the mtrCAB gene cluster (Group I, Figure 3). This group of bacteria included S. japonica KCTC 22435, S. livingstonensis LGM 19866, S. putrefaciens CN32, S. woodyi ATCC 51908, Shewanella sp. ARC9_LZ, Shewanella sp. SUN WT4, and Shewanella sp. WPAGA9 (Supplementary Table S1). Notably, the dmsEFAB and mtrCAB gene clusters of S. japonica KCTC 22435 were 99.5%–100% identical to those of Shewanella sp. WPAGA9, respectively (Figures 2A–E; Supplementary Tables S2–S6).

Like Group I bacteria, Group II bacteria also contained a dmsEFAB gene cluster and a mtrCAB gene cluster. However, the mtrCAB gene cluster of this group of bacteria also contained a mtrDEF gene cluster and the genes for other outer membrane multiheme c-Cyt (Group II, Figure 3). Group II bacteria included S. fidelis ATCC-BAA-318, S. marisflavi EP1, S. piezotolerans WP3, S. schlegeliania JCM 11561, S. xiamenensis NUITEM-VS1, Shewanella sp. 8A, Shewanella sp. ISTPL2, Shewanella sp. LZH-2, Shewanella sp. MBLT60-112-B1, Shewanella sp. MBLT60-112-B2, and Shewanella sp. MR-4 (Supplementary Table S1). Among this group of bacteria, Shewanella sp. MBLT60-112-B1 and Shewanella sp. MBLT60-112-B2 shared identical dmsEFAB and mtrCAB gene clusters (Figures 2A–E; Supplementary Tables S2–S6). These results suggest that they are very closely related. Results also showed that dmsEFAB and mtrCAB gene clusters of S. xiamenensis NUITEM-VS1, Shewanella sp. 8A, and Shewanella sp. LZH-2 were 97.6%–100% identical, respectively (Figures 2A–E; Supplementary Tables S2–S6), which suggest that these Shewanella spp. acquire dmsEFAB and mtrCAB gene clusters from a common ancestor.

Both Group III and IV bacteria contained two dmsEFAB gene clusters and a mtrCAB gene cluster. The major difference between these two groups of bacteria was that the mtrCAB gene of Group IV bacteria also had a mtrDEF gene cluster and one to three genes for the outer membrane c-Crys (Group III and IV, Figure 3). Group III bacteria included Ferrimonas sp. SCSIO 43195, S. frigidimarina NCIMB 400, Shewanella sp. Actino-trap-3 and Shewanella sp. XX20019, among which an additional gene for the outer membrane multiheme c-Cyt associated with the mtrCAB gene cluster of S. frigidimarina NCIMB 400, Shewanella sp. Actino-trap-3 and Shewanella sp. XX20019 (Group III, Figure 3; Supplementary Table S1). The identified Group IV bacteria were Ferrimonas balericus DSM 9799 and S. oneidensis MR-1 (Supplementary Table S1).

Group V bacteria all possessed three dmsEFAB gene clusters and a mtrCAB gene cluster with an additional gene for the outer membrane c-Cyt (Group V, Figure 3), which included F. lipolytica S7 and S. psychromarinicola M2 (Supplementary Table S1). One of the dmsEFAB gene clusters, the 06735–06750 gene cluster, of S. psychromarinicola M2 was 100% identical to the 15625–15640 gene cluster, one of the dmsEFAB gene clusters of Shewanella sp. Actino-trap-3 from Group III (Figures 2A–D; Supplementary Tables S2–S5). Similarly, mtrCAB gene cluster of S. psychromarinicola M2 was 97.1%–99.7% identical to that of Shewanella sp. Actino-trap-3 (Figures 2C–E; Supplementary Tables S4–S6).

Both Group VI and VII bacteria had a mtrCAB gene cluster that also included a mtrDEF gene cluster and three genes for the outer membrane c-Crys (Group VI and VII, Figure 3). However, the Group VI bacterium S. eurypsychrophilus YLB-08 possessed four dmsEFAB gene clusters (Group VI, Figure 3; Supplementary Table S1), while the Group VII bacteria S. sediminis HAW-EB3 and Shewanella sp. YLB-09 contained six dmsEFAB gene clusters (Group VII, Figure 3; Supplementary Table S1). Notably, among the six dmsEFAB gene clusters of Shewanella sp. YLB-09, the 01235–01250 and 01530–01515 gene clusters were 100% identical to the 02855–02870 and 03150–03135 gene clusters, respectively, which demonstrates duplications of dmsEFAB gene clusters in Shewanella sp. YLB-09 (Figures 2A–D; Group VII, Figure 3; Supplementary Tables S2–S5). Furthermore, the mtrCAB gene cluster and its associated genes (07500–07460) of S. eurypsychrophilus YLB-08 were 100% identical to the mtrCAB gene cluster and its associated genes (09075–09035) of Shewanella sp. YLB-09 (Group VI and VII, Figure 3; Supplementary Tables S4–S6). Similarly, dmsEFAB gene clusters 01250–01265, 01545–01530, 23480–23495, and 23515–23530 of S. eurypsychrophilus YLB-08 were 100% identical to dmsEFAB gene clusters 01235–01250/02855–02870, 01530–01515/03150–03135, 25010–25025 and 25045–25060 of Shewanella sp. YLB-09, respectively (Group VI and VII, Figure 3; Supplementary Tables S2–S5). Thus, the dmsEFAB and mtrCAB gene clusters of S. eurypsychrophilus YLB-08 and Shewanella sp. YLB-09 must be acquired from the same ancestor.

Conclusion

To investigate to what extent the extracellular IO₃⁻−-reducing organisms were distributed around world, the bacteria with both dmsEFAB and mtrCAB gene clusters were systemically searched. A total of 29 bacteria were identified to possess both gene clusters. They belonged to the genus of Ferrimonas and Shewanella. Possession of both dmsEFAB and mtrCAB gene clusters suggests the ability to mediate extracellular reduction of IO₃⁻− by these bacteria. The identified bacteria with capability for extracellular reduction of IO₃⁻− were widespread around the world. Although some were found in freshwater lakes and rivers as well as the rocks of deep continental subsurface, most of them were derived from geographically distributed marine environments. The latter included those found in the Arctic, Atlantic, Indian, and Pacific
Widespread occurrence of the bacteria with capability for extracellular reduction of IO$_3^-$ suggests a crucial role of this group of bacteria in global biogeochemical cycling of iodine. The genetic organizations of identified dmsEFAB and mtrCAB gene clusters varied significantly. The mtrCAB gene clusters often associated with genes for the outer membrane c-Cyt of multiheme. Some of the mtrCAB gene clusters also contained a mtrDEF gene cluster. The numbers of dmsEFAB gene cluster detected in a given bacterial genome ranged from one to six. Duplications of the detected dmsEFAB gene clusters also occurred. Thus, this group of bacteria acquire their capability for extracellular reduction of iodate differently.
Collectively, the results from this investigation provide new insights into the distribution and evolution of as well as the role in global biogeochemical cycling of iodine by the bacteria with capability for extracellular reduction of iodate. Physiological characterization of the iodate-reducing capacity for the predicted strains and their ecological roles on iodine cycling in different ecosystems are in need of further investigation.

Data availability statement

The original contributions presented in the study are included in the article/Supplementary material, further inquiries can be directed to the corresponding authors.

Author contributions

LS designed the experiment and acquired funding. JG, JJ, and YZ performed the experiment. ZP, YJ, ZJ, YH, YD, and LS analyzed the data and prepared manuscript. ALL authors contributed to the article and approved the submitted version.

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Conflict of interest

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Supplementary material

The Supplementary material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fmicb.2022.1070601/full#supplementary-material
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