Iodine is an essential trace element for humans and animals because of its important role as a constituent of thyroid hormones. If the anthropogenic iodine-129 (\(^{129}\)I, half-life: \(1.6\times10^7\) years), which is released from nuclear facilities into the environment and has a long half-life, participates in the biogeochemical cycling of iodine, it potentially accumulates in the human thyroid gland and might cause thyroid cancer. Therefore, it is necessary to obtain better information on the behavior of iodine in the environment for accurate safety assessments of \(^{129}\)I. Major pathways of iodine cycling are the volatilization of organic iodine compounds into the atmosphere, accumulation of iodine in living organisms, oxidation and reduction of inorganic iodine species, and sorption of iodine by soils and sediments. Considerable geochemical evidence has indicated that these processes are influenced or controlled by microbial activities, although the precise mechanisms involved are still unclear. This review summarizes current knowledge on interactions between microorganisms and iodine, with special emphasis on newly isolated bacteria possibly contributing to the cycling of iodine on a global scale.

**Key words:** iodine, radionuclide, ozone destruction, biogeochemical cycling, microorganisms

### What is iodine?

Iodine is an essential trace element for vertebrates because of its role as a constituent of thyroid hormones, *i.e.* thyroxine and triiodothyronine. An insufficient amount of iodine in diet leads to iodine deficiency disorders such as endemic goiter and cretinism\(^{18,39}\), which are still the most common causes worldwide of mental retardation and brain damage\(^{67}\). The average person needs about 0.2 mg of iodine daily, most of which comes from foods. Seafood is a good source of iodine because seawater contains considerable amounts of iodine. Seaweed including kelp contains substantial amounts of iodine, occasionally more than 4% of dry weight\(^{48}\). However, the distribution of iodine in terrestrial and freshwater areas is uneven. Iodine deficiency is especially common in alpine areas (Himalayas, Andes and Alps), but many other areas including Central Africa, Central Asia and Europe remain problematic\(^{77}\).

Japanese frequently eat kelp (konbu) and other seaweeds (nori, hijiki and wakame), and thus are among the world’s greatest consumers of iodine. Japan is also the world’s second largest iodine-producer. Some 80 to 90% of the iodine produced in Japan comes from Chiba prefecture. In this area, ancient underground seawater called “brine” is pumped out to produce natural gas, the iodine concentration (approximately 1 mM) of which is 2,000 times that in natural seawater\(^{61}\). Annual production of iodine in this area accounts for 6,000 tons, constituting one third of the world’s production. Iodine has a number of applications in a wide variety of industries, including in X-ray contrast media, biocides, catalysts and feed additives. Currently, Chile, Japan, Turkmenistan, China, Russia, Indonesia, Azerbaijan Republic and the USA are the world’s major producers of iodine.

Iodine-129 (\(^{129}\)I, half-life: \(1.6\times10^7\) years) is one of the most persistent anthropogenic radionuclides, and has been released into the environment as a fission product from nuclear weapon tests and the operation of nuclear facilities\(^{12,41,39,69}\). This nuclide has become widely distributed in the global environment and can be expected, given its long half-life, to behave in similar way to stable iodine (\(^{127}\)I) over long periods\(^{21}\). Ultimately, it may accumulate in the human thyroid gland and cause thyroid cancer\(^{22}\). In a recent report by the United Nations Scientific Committee on the Effects of Atomic Radiation\(^{86}\), \(^{129}\)I is regarded as one of the most important radionuclides to be assessed in terms of global circulation. However, the biogeochemical cycling of iodine is not well understood, mainly due to a lack of data on the complex behavior of iodine in the environment. Thus, it is necessary to study the behavior of iodine throughout the biosphere to understand its biogeochemical cycles and also to ensure the safety of \(^{129}\)I\(^{67}\).

### Volatilization of iodine

The most significant feature of the biogeochemical cycling of iodine is its volatilization from the ocean into the atmosphere\(^{59,89}\). Atmospheric iodine may participate in the destruction of ozone like other halogen species, such as chlorine and bromine\(^{26,81}\). Iodine atoms react with ambient ozone to form iodine oxide (IO), which is converted back to iodine atoms by photodissociation or by reactions with halogen oxides (ClO, BrO and IO)\(^{23}\). Iodine is thought to volatilize from seawater in the form of organic iodine compounds, such as methyl iodide (CH\(_3\)I)\(^{18,70,73}\), diiodomethane (CH\(_2\)I\(_2\))\(^{17,63}\), chloriodomethane (CH\(_3\)I\(_2\))\(^{27,45,59}\), iodoethane...
(C₃H₇I)²⁷,⁹⁹ and iodopropane (C₃H₇I)⁴⁵. Among these, CH₃I has the highest atmospheric mixing ratio (around 0.5–1.0 parts per trillion by volume)¹⁷ and may play a significant role in global iodine transfer. A series of ozone-depleting chain reactions caused by CH₃I proceeds as follows:

\[ \text{CH}_3\text{I} + h\nu \rightarrow \text{CH}_3 + \text{I} \\
\text{I} + \text{O}_3 \rightarrow \text{IO} + \text{O}_2 \\
\text{IO} + \text{I} \rightarrow 2\text{I}_2 + \text{O}_2 \]

Several lines of evidence have shown that CH₃I is produced by living organisms in the ocean⁴⁹,⁷⁰, although some terrestrial sources (rice paddies⁶³,⁷², higher plants⁷⁴,⁹⁸, and fungi⁴⁶,⁷¹,⁹⁸) have been proposed. Annual global CH₃I production in the ocean has been estimated at 1 to 4×10¹¹ g⁷⁷,⁷⁹. Marine algae and phytoplanktons are well-known CH₃I producers, but laboratory experiments have revealed that their production (10⁶ to 10¹⁰ g year⁻¹) is insufficient to account for global CH₃I production (Table 1)⁴³,⁵²,⁵⁴,⁶⁵,⁷⁶. Thus, it has been pointed out that bacteria are also involved in the processes⁵³. In 2001, it was found that a wide variety of bacteria, including marine and terrestrial bacteria, are capable of methylating iodide (I⁻, oxidation state; −1) to form CH₃I³. Aerobic bacteria such as Alteromonas macroeolida, Vibrio splendidus, Rhizobium sp. and Methyllosinus trichosporium showed significant production of CH₃I, whereas anaerobic microorganisms (Clostridium sp. and methanogens) did not produce it. Bacterial CH₃I production depended greatly on surrounding iodide levels, and the iodide-methylating reaction was mediated enzymatically with S-adenosyl-L-methionine as the methyl donor⁵¹. In several eukaryotic organisms such as marine algae⁶⁰, wood-rotting fungi⁵⁷,⁹⁸ and terrestrial higher plants⁷₄,⁹⁸, similar enzymes catalyzing the methylation of halides have been characterized. Annual bacterial CH₃I production in the ocean can be roughly assessed based on the mean rate of CH₃I production by resting cells of 10 marine bacterial strains (2.5 pg day⁻¹ 10¹⁰ cells⁻¹, ranging from 0 to 8.1 pg day⁻¹ 10¹⁰ cells⁻¹)³, and the total number of prokaryotic cells in the upper 100 m of the open ocean (1.8×10¹⁰ cells) estimated by Whitman et al.⁹⁰. The estimated value (1.6×10¹⁰ g year⁻¹) is at least one order of magnitude greater than annual algal production (10⁶–10⁸ g year⁻¹) and comparable to that produced by phytoplanktons (10⁶–10¹⁰ g year⁻¹), suggesting that bacteria are a potential new source of CH₃I in the ocean (Table 1). However, it should be noted that estimated bacterial production as well as algal and phytoplanktonic production is still significantly less than global CH₃I production (1 to 4×10¹¹ g year⁻¹).

The bacterial contribution to CH₃I production was also determined using environmental samples such as soil slurries and seawater⁴³,⁴⁴. The amount of iodine volatilized from various soil slurries was variable, but autoclaving resulted in a marked decrease in volatilization in all samples⁴⁴. Yeast extract, added to stimulate microbial activities, sometimes enhanced the volatilization more than 10 times. When the slurries were incubated with the antibiotics streptomycin and tetracycline, specific inhibitors for prokaryotes, iodine volatilization was completely inhibited. On the other hand, the addition of cycloheximide, a specific inhibitor for eukaryotes, did not have a significant inhibitory effect. These results suggest that bacteria contribute significantly to the volatilization of iodine from soil environments⁴⁴. Similar experiments were carried out using surface seawater collected from several locations⁵³. Iodine volatilization from seawater was completely inhibited by autoclaving or filtration through a membrane filter with a pore size of 0.22 μm. However, both prokaryotic and eukaryotic inhibitors showed significant inhibitory effects, though the prokaryotic inhibitors were less effective than in the soil slurries. Thus, it was presumed that iodine in seawater is volatilized by the actions of both bacteria and eukaryotic microorganisms⁵³.

**Table 1.** Estimated annual CH₃I production by various marine organisms

<table>
<thead>
<tr>
<th>Organisms</th>
<th>Annual production (×10⁹ g year⁻¹)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Algae</td>
<td>0.2</td>
<td>52</td>
</tr>
<tr>
<td></td>
<td>0.003 to 0.17</td>
<td>76</td>
</tr>
<tr>
<td></td>
<td>0.008 to 0.05</td>
<td>65</td>
</tr>
<tr>
<td>Phytoplanktons</td>
<td>0.3</td>
<td>43</td>
</tr>
<tr>
<td></td>
<td>5 to 38</td>
<td>43</td>
</tr>
<tr>
<td></td>
<td>1.2</td>
<td>54</td>
</tr>
<tr>
<td>Bacteria</td>
<td>1.6</td>
<td>This study</td>
</tr>
</tbody>
</table>

**Accumulation of iodine**

Iodine is a biophilic element, and accumulates in various organisms including vertebrates, algae, coral, sponges, lobworms, and shellfishes. To date, however, the mechanisms of iodine uptake by living organisms have only been characterized in the thyroid gland in mammals and in brown algae. In the thyroid gland, iodide is taken up by a sodium/iodide symporter²⁶,²⁷,⁸⁰. This transmembrane protein co-transport sodium ion with iodide ion into the thyroid against a concentration gradient. The driving force for the process is an electrochemical gradient of sodium ion across the membrane. As a result of this active transport system, the normal thyroid concentrates iodide by a factor of 20 to 40⁹⁰. On the other hand, brown algae such as Laminaria digitata often accumulate iodide from seawater with a concentration factor of 1.5×10⁵. Küpper et al.⁴⁰ showed that iodide in seawater was first oxidized to hypioiodous acid (HIO, oxidation state; +1) by a cell wall haloperoxidase in L. digitata, and that this non-ionized iodine species then freely penetrated algal cells by means of facilitated diffusion.

In 2005, an iodine-accumulating bacterium, designated strain C-21, was isolated from surface marine sediment⁷. This strain was phylogenetically closely related to a marine aerobic bacterium, Arenibacter troitsensis, a member of the family Flavobacteriaceae. When cultured in a liquid medium containing 0.1 μM iodide, C-21 removed 80% of the iodine from the medium within 24 h, and accumulated it in its cells with a maximum concentration factor of 5.5×10⁶. The initial rate of by washed cells showed substrate saturation kinetics with an apparent affinity constant for transport of 0.073 μM. Iodine transport assays revealed that glucose and oxygen were necessary for the uptake of iodine⁴⁶. The uptake was not or only partially inhibited by various metabolic inhibitors (ionophores, protonophores and ATPase inhibitors), whereas reducing agents and catalase strongly inhibited the uptake. When exogenous glucose oxidase was
Interactions between Iodine and Microbes

Mechanism of iodine uptake by *Arenibacter* sp. C-21. GO, membrane-bound glucose oxidase; HPO, haloperoxidase; Org-I, organic iodine compound. For clarity, the periplasmic space and outer membrane are not shown.

![Mechanism of iodine uptake by *Arenibacter* sp. C-21](image)

Mechanism of iodine uptake by *Arenibacter* sp. C-21. GO, membrane-bound glucose oxidase; HPO, haloperoxidase; Org-I, organic iodine compound. For clarity, the periplasmic space and outer membrane are not shown.

Fig. 1. Mechanism of iodine uptake by *Arenibacter* sp. C-21. GO, membrane-bound glucose oxidase; HPO, haloperoxidase; Org-I, organic iodine compound. For clarity, the periplasmic space and outer membrane are not shown.

Added to the cell suspension, enhanced uptake of iodine was observed. The uptake occurred even in the absence of glucose and oxygen, if hydrogen peroxide was added to the cell suspension. These findings indicate that hydrogen peroxide produced by glucose oxidase plays a key role in the uptake of iodine. Actually, significant activity of glucose oxidase was found in the membrane fraction of C-21. Furthermore, enzymatic oxidation of iodide strongly stimulated uptake in the absence of glucose. Based on these results, the mechanism illustrated in Fig. 1 is proposed. First, glucose is oxidized by the membrane-bound glucose oxidase, and hydrogen peroxide is produced. Second, iodide is oxidized to hypoiodous acid probably by an unidentified haloperoxidase. Finally, hypoiodous acid is probably transported via carrier-mediated facilitated diffusion. In the cells, hypoiodous acid should be reduced to iodide again, or be associated with certain organic compounds, to avoid a release of iodine from the cells. At present, however, the chemical form of iodine in the cells is still unclear. It is interesting that such a mechanism of iodine uptake is very similar to that found in brown algae.

The physiological reason why C-21 takes up and accumulates iodine remains unclear. Recently, Küpper et al. found that the major form of iodine accumulated in *L. digitata* was iodide, and proposed that iodide functions as an organic antioxidant, which efficiently scavenges reactive oxygen species upon oxidative stress. Considering the resemblance in uptake systems between C-21 and *L. digitata*, the uptake of iodine by C-21 could function in the detoxification of hydrogen peroxide generated by the membrane-bound glucose oxidase. In this case, however, the role of the hydrogen peroxide production by glucose oxidase is still unclear.

Reduction of iodate

The average total concentration of dissolved iodine in seawater is 0.45 μM, and the predominant chemical forms are iodide and iodate (IO3−, oxidation state; +5)2,89,91. Thermodynamically, the concentration ratio between iodate and iodide (IO3−/I−) in oxygenated seawater (at pH 8.1 and pE 12.5) should be 3.2×10^14, indicating that iodate is the more stable form. However, significant quantities of iodide, up to 0.3 μM, are observed at the surface6,82,83. It is widely speculated that this apparent disequilibrium is caused by the biological reduction of iodate to iodide, and marine microorganisms such as *bacterial24,28,84* and *phytoplankton19,88,95* may play significant roles in the process. Iodide is also the dominant form of iodine in deep oxygenated water84, anoxic basins20,29,30,50,85,94,96, and porewater of marine sediment11,44,60,66. In deep water, iodide is often highly enriched at concentrations of several micromolars to more than 1 mM. In addition to the abiotic chemical reduction of iodate and microbial remineralization of organic iodine compounds, the bacterial reduction of iodate is expected to be an important process to maintain the reduced form of iodine in these environments28–30,44,50.

To date, only a few studies have been conducted on the bacterial reduction of iodate. Tsunogai and Sase reported that several laboratory strains of nitrate-reducing bacteria reduced iodate. They also found that cell-free extracts of *Escherichia coli*, which included nitrate reductase activity, reduced iodate. They proposed that iodate in seawater is reduced by the nitrate reductases of marine organisms84, although this hypothesis has not been investigated further in bacteria. *Desulfovibrio desulfuricans* ATCC 2957724 and *Shewanella oneidensis* (formerly *S. putrefaciens*) MR-428 have been reported to reduce iodate, but the enzymes involved in the reaction were not determined. Despite these early studies, much is still unclear about the physiological and biochemical mechanisms of bacterial iodate reduction. For instance, it is not fully understood whether the reduction is a dissipatory process or not. In addition, little information is available about the key enzymes catalyzing the reduction of iodate.

Recently, an iodate-reducing bacterium, designated strain SCT, was newly isolated from marine sediment5. Strain SCT was phylogenetically closely related to a denitrifying bacterium, *Pseudomonas stutzeri*, and reduced 200 μM iodate to iodide within 12 h in an anaerobic culture containing 10 mM nitrate. The strain did not reduce iodate under the aerobic conditions. An anaerobic washed cell suspension of SCT reduced iodate, when the cells were pregrown anaerobically with 10 mM nitrate and 200 μM iodate. However, cells pregrown without iodate did not reduce it. Although both types of cells showed similar levels of nitrate reductase activity, only the former cells (pregrown with iodate) showed methyl viologen-dependent iodate reductase activity, which was located predominantly in the periplasmic space. Furthermore, SCT was capable of anaerobic growth with 2 to 4 mM iodate as the sole electron acceptor. The growth was nearly proportional to the iodate concentration in the medium. In the presence of 3 mM iodate and 10 mM malate, a 520-fold increase in the number of cells was observed with a generation time of 14 h (Fig. 2). The growth of SCT coincided with the reduction of iodate, and iodate was transformed completely into iodide. Malate, lactate, glycerol, glucose, succinate and acetate could serve as the electron donor. These results suggest that SCT is a dissimilatory iodate-reducing bacterium, and that its periplasmic iodate reductase is induced by iodate under anaerobic growth conditions5. Considering the high redox potential of iodate reduction...
oxidizing bacteria produced not only I$_2$ but also volatile organic iodine compounds such as CH$_2$I$_2$, CH$_2$ClI and iodoform (CHI$_3$). Fuse et al.$^{35}$ suggested that I$_2$ and organic acids released into the media were important sources of these organic iodine compounds.

While attempting to isolate iodide-oxidizing bacteria from the environment, Amachi et al.$^8$ made an interesting discovery. They first tried to isolate iodide-oxidizing bacteria directly from seawater samples, but were unsuccessful because the population sizes of the bacteria were always below the detection limit (10 CFU mL$^{-1}$). However, the population sizes increased to 10$^3$ to 10$^5$ CFU mL$^{-1}$ after the seawater samples were incubated with a high concentration of iodide (1 mM), and this “enrichment” process enabled them to isolate iodide-oxidizing bacteria repeatedly (Fig. 3). A similar phenomenon was observed during the isolation of “P. iodoxidans” from seawater aquaria.$^{34}$ Gozlan$^{34}$ reported that seawater of aquaria contained 60 $\mu$M of iodine. Since this is 120 times the level in natural seawater, the aquaria must have been supplemented with exogenous iodine. Fuse et al.$^{35}$ also carried out an enrichment of iodide-oxidizing bacteria by inoculating marine samples into a liquid medium containing 0.9 mM of iodide. Furthermore, Amachi et al.$^8$ found that iodide-oxidizing bacteria could be isolated without any enrichment steps from natural gas brines, whose iodide levels were very high (60 $\mu$M to 1.2 mM). These results strongly suggest that iodide-oxidizing bacteria prefer iodide-rich environments.$^8$ The physiological reason why iodide-oxidizing bacteria in seawater can be enriched with iodide is still uncertain. One possibility is that iodide is advantageous to iodide-oxidizing bacteria, through the exclusion of competing microorganisms by toxic iodine species. I$_2$ produced by iodide-oxidizing bacteria is a highly active oxidizing agent, and has strong bactericidal, fungicidal, and sporicidal activities.$^{56}$ Therefore, it is possible that iodide-oxidizing bacteria attack competing microorganisms with I$_2$, and increase their population sizes within iodide-enriched seawater. Considering that iodide-oxidizing bacteria were only isolated from natural gas brines without enrichment, they probably occupy their ecological niche in this iodide-rich environment using a similar strategy.
Interactions between Iodine and Microbes

273

Sorption of iodine by soils and sediments

In terrestrial environments, iodine is strongly adsorbed by soils, and high iodine concentrations in soils have been reported, e.g. 5 mg kg\(^{-1}\) as a world-wide average\(^{14}\) and about 30 mg kg\(^{-1}\) as a representative value for Japanese soil\(^{62}\). These values are much greater than those for rocks and plants (0.05 to 0.5 mg kg\(^{-1}\))\(^{89}\), indicating that iodine is highly accumulated in soils. Although the sorption by soils is affected by various physico-chemical parameters including soil type, pH, Eh, salinity, and organic matter content\(^{32,89}\), a number of studies have indicated that soil microorganisms are also involved in the process\(^{11,13,46,62}\). Muramatsu and Yoshida\(^{62}\) found that autoclaving of soils significantly reduced the sorption of iodide. The decreased sorption was recovered by incubation of autoclaved soils with a smaller amount (1%) of fresh soil, suggesting an effect of increased microbial activities. Koch et al.\(^{46}\) observed increased iodide sorption in soils treated with glucose, and decreased sorption in soils treated with thymol (an antiseptic). Decreased iodine sorption by fumigated, air-dried, or gamma-irradiated soil has also been reported\(^{13}\). Similarly, several studies on marine sediment (oxic sediment) showed the presence of very high concentrations of iodine\(^{51,67,68}\). For instance, 96 to 1,990 mg kg\(^{-1}\) of iodine was observed in surface sediment of the Southwest African Shelf\(^{68}\). Since the total dissolved iodine concentration in seawater is only 60 μg L\(^{-1}\)\(^{91}\), it is apparent that such sediment is highly enriched with iodine. Malcolm and Price\(^{51}\) speculated that the accumulation of iodine in sediment occurs as a result of bacterial activities. Bacterial importance in iodine sorption was also suggested in freshwater sediment\(^{12}\).

The sorption of iodine by soils and sediments is of great importance when considering the fate of long-lived \(^{129}\)I released from nuclear facilities. However, details regarding the microbial effects on iodine sorption remain to be elucidated. There are two possible ways to explain how microorganisms are involved in the sorption of iodine in the environment. The first one is that iodide in soil or sediment is oxidized to I\(_2\) by a microbial enzyme, and subsequently incorporated into organic compounds\(^{11}\). The second one is that iodine is bound to a microbial cell envelope or taken up by the cells\(^{11,13}\). As described above, bacteria catalyzing these processes have been isolated and characterized, i.e. iodide-oxidizing bacteria and iodine-accumulating bacteria. Interestingly, both types of bacteria oxidize iodide upon catalyzing the process, with oxygen and hydrogen peroxide as the electron acceptor, respectively\(^{6,8}\). Thus, it is presumed that iodide oxidation, at least in part, plays a role in the sorption of iodine. This hypothesis is supported by the fact that iodine sorption in soils occurs more easily under oxic conditions than anoxic conditions\(^{11-13,62,77}\).

Concluding remarks

The biogeochemical cycling of iodine is summarized in Fig. 4. As described in this review, microorganisms (particularly bacteria) participate in the cycling through processes such as volatilization, accumulation, reduction, oxidation and sorption. Another possible significant pathway
may include the degradation or dehalogenation of iodinated aliphatic- and aromatic-compounds. Several aerobic bacteria capable of growing on methyl halides (CH3Cl, CH3Br and CH3I) as a sole source of carbon and energy have been isolated, including *Methyllobacterium chloromethanicum CM4* [54], *Hyphomicrobium chloromethanicum CM2* [55], *Aminobacter* sp. IMB-1 [52] and *Aminobacter* sp. CC495 [53], Anaerobic microorganisms capable of reductive de-iodination, such as dehalorespiring “Dehalococciodes” [40], may also exist, but have not been isolated so far. The characterization of such “iodine-related microorganisms” at physiologic, phylogenetic and genetic levels will help us to understand the global circulation of this essential element, and thus enable us to assess more accurately the safety of radioactive iodine. Currently, the underground disposal of high-level radioactive waste is underway in many countries. It could possibly leak from subterranean burial sites into surrounding groundwater, and ultimately into the human environment. Therefore, interactions between subsurface microorganisms and iodine with special emphasis on microbial functions as a “natural barrier” should be evaluated in the near future.

Acknowledgements

The author acknowledges Dr. Yasuyuki Muramatsu (Gakushuin Univ., Dr. Yoichi Kamagata (AIST) and Dr. Takaaki Fuji (Chiba Univ.) for continuous encouragement and support. This work was supported in part by KAKEN to S. A. (no. 20780049).

References

Interactions between Iodine and Microbes


