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Dimethylsulfoniopropionate (DMSP) assimilation by *Synechococcus* in the Gulf of Mexico and northwest Atlantic Ocean

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Abstract

A variety of bacterial phylogenetic groups assimilate dimethylsulfoniopropionate (DMSP), an organic sulfur compound that can satisfy most of the sulfur (S) demand of bacteria in the surface waters of the ocean. Marine *Synechococcus* are capable of utilizing some forms of dissolved organic matter, but it is unknown if *Synechococcus* also assimilate DMSP. To better understand the role of *Synechococcus* in the flux of DMSP, we used microautoradiography to follow the assimilation of ^{35}S-DMSP and ^{35}S-methanethiol, an intermediate in DMSP assimilation, by *Synechococcus* in the surface waters of the Gulf of Mexico and the northwest Atlantic Ocean. About 85% of *Synechococcus* cells assimilated S from DMSP and methanethiol in these environments, and *Synechococcus* assimilated more DMSP per cell than other bacteria. On average, *Synechococcus* accounted for roughly 20% of prokaryotic DMSP assimilation in the northwest Atlantic and the Gulf of Mexico. Tests with axenic cultures of *Synechococcus* revealed that two phycoerythrin-containing strains (WH8102 and WH7803) were capable of DMSP transport, although these strains did not produce dimethylsulfide (DMS). These data indicate that DMSP could provide a significant amount of S to *Synechococcus* and that *Synechococcus* are important consumers of DMSP in the ocean.

Dimethylsulfoniopropionate (DMSP) can play an important role in marine food webs and in the regulation of global temperature (Kiene et al. 2000; Simó 2001). DMSP is produced by various groups of phytoplankton (Yoch 2002) and is released into the water column through phytoplankton senescence, viral lysis, and grazing (Wolfe et al. 1994; Bratbak et al. 1995). After its release, marine bacteria quickly metabolize dissolved DMSP in a variety of ways. Bacteria can lyse DMSP to produce dimethylsulfide (DMS), a climatically active gas hypothesized to mitigate changes in global temperature (Charlson et al. 1987). Most DMSP, however, is quickly assimilated into bacterial biomass or transformed into nonvolatile compounds. Marine bacteria are thought to assimilate sulfur (S) from DMSP through a demethylation–demethiolation process that produces methanethiol (MeSH), a volatile compound that can be used in the synthesis of methionine and cysteine (Kiene et al. 1999). The assimilation of DMSP is estimated to satisfy as much as 90% of the S demand of bacterial communities in surface waters of the ocean (Kiene and Linn 2000*a*; Simó et al. 2002).

The capacity to assimilate S from DMSP appears to be spread among a variety of bacterial groups, such as the alpha-, beta-, and gamma-proteobacteria and *Cytophaga-Flavobacteria* (González et al. 1999; Malmstrom et al. 2004*b*; Vila et al. 2004). The *Roseobacter* clade, a subgroup of the alpha-proteobacteria, can be particularly important to the flux of DMSP when they are abundant (Zubkov et al. 2002; Malmstrom et al. 2004*b*; Vila et al. 2004). However, the capacity to assimilate DMSP into biomass or to produce DMS is not uniformly distributed among members of the *Roseobacter* clade (González et al. 1999). If the capacity to metabolize DMSP also varies among other phylogenetic groups, then the composition of bacterial communities could influence whether DMSP is assimilated into biomass or lysed to produce DMS. To better understand the biogeochemical flux of DMSP, it is necessary to identify the bacterial groups capable of metabolizing DMSP.

It is unknown if *Synechococcus,* an abundant group of autotrophic bacteria (Waterbury et al. 1979), metabolize DMSP, but *Synechococcus* is known to utilize other organic

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compounds. For example, many *Synechococcus* use urea as a source of nitrogen (Moore et al. 2002), and *Synechococcus* strains WH7803, WH8101, and WH8113 can take up amino acids (Willey and Waterbury 1989; Paerl 1991). In addition, genomic analyses of *Synechococcus* strain WH8102 revealed genes homologous to those for transporting amino acids, oligopeptides, and cyanate (Palenik et al. 2003). If *Synechococcus* consume other components of the dissolved organic matter (DOM) pool, then they may also consume DMSP.

To better understand the role of *Synechococcus* in the flux of DMSP, we used microautoradiography to follow the assimilation of ^{35}S-DMSP and ^{35}S-MeSH by *Synechococcus* in the northwest Atlantic Ocean and the Gulf of Mexico. In addition, the capacity to take up and lyse DMSP was examined in axenic cultures of four strains of *Synechococcus.* We found that *Synechococcus* could take up DMSP and were responsible for 20% of total DMSP assimilation by prokaryotes, on average.

Materials and methods

Surface seawater samples were collected in the Gulf of Mexico (November 2001, RV *Pelican*), the Sargasso Sea (April 2002, RV *Oceanus*), and the Mid-Atlantic Bight (November 2002, RV *Cape Henlopen*). Whole seawater (30–60 mL) was incubated with additions of either ^{35}S-DMSP ($<$0.1 nmol L^{-1}; specific activity 12–43 TBq mmol^{-1}) or ^{35}S-MeSH ($<$0.01 nmol L^{-1}; specific activity 43 TBq mmol^{-1}) for 24 h in glass serum vials or acid-washed polycarbonate bottles. Samples were incubated in flow-through water baths at in situ temperatures (13–25°C) and 60% surface light levels (minus ultraviolet). Identical dark incubations were conducted at four stations in the Sargasso Sea and North Carolina coastal waters. Live incubations were terminated by the addition of 20% fresh paraformaldehyde (final concentration of 2%). Samples were fixed for 24 h at 4°C before filtration onto 0.2-μm white, polycarbonate filters. Filters were rinsed twice with 5 mL of deionized, filter-sterilized water and stored at $-$20°C. Identical incubations with tracer additions of ^{35}S-DMS ($<$0.1 nmol L^{-1}; specific activity 12–43 TBq mmol^{-1}) were conducted to examine possible uptake of ^{35}S-DMS produced from ^{35}S-DMSP. Killed controls were treated with paraformaldehyde 10 min prior to addition of radiolabeled substrates and were incubated simultaneously with live incubations. Assimilation of radioactive compounds was negligible in the killed control incubations.

Microautoradiography preparations followed the method described elsewhere (Malmstrom et al. 2004*b*). Microautoradiography samples were examined with a semiautomated microscopy and image analysis system described in detail previously (Cottrell and Kirchman 2003). Briefly, images of 4′,6′-diamidino-2-phenylindole (DAPI) fluorescence, phycoerythrin autofluorescence, and silver grain clusters formed during microautoradiography were acquired from 10–30 fields of view and the images were overlaid. Objects that matched the dimensions of *Synechococcus* (ca.1-μm–diameter spheroids) and exhibited both DAPI and phycoerythrin fluorescence were counted as *Synechococcus* (Waterbury et al. 1979). Silver grain clusters overlapping with DAPI-positive bacteria indicated cell-specific assimilation of radiolabeled compounds. Since microautoradiography samples were fixed with paraformaldehyde and filtered onto polycarbonate filters, which causes cells to lose unincorporated ^{35}S-DMSP (Kiene and Linn 1999), the formation of silver grains was indicative of assimilation of ^{35}S into high-molecular-weight biomass. *Synechococcus* and substrate-assimilating cells were enumerated during automated image analysis, and the area of silver grains associated with cells was measured. These automated counts of the fraction of *Synechococcus* assimilating DMSP and MeSH were consistent with manual counts. A comparison of four preparations with and without microautoradiography confirmed that the microautoradiography treatment did not affect the fraction of cells identified as *Synechococcus* (data not shown).

Radio-labeled *Synechococcus* from three locations (Sta. 26, 36, and 39) were analyzed using flow cytometry–cell sorting. Samples were fixed with 1% paraformaldehyde + 0.05% glutaraldehyde (final concentration), frozen in liquid nitrogen, and stored at $-$80°C. *Synechococcus* were identified and sorted based on chlorophyll *a* ($>$670 nm; FL3 instrument channel) and phycoerythrin (585 nm centered; FL2 instrument channel) autofluorescence using a FACSCalibur flow cytometer–cell sorter (excitation 488 nm) (Li 1994). Duplicate samples of 30,000–50,000 *Synechococcus* were collected onto 0.2-μm nylon filters and assayed by liquid scintillation counting.

Determination of DMSP concentrations and flux in these samples was described in detail previously (Malmstrom et al. 2004*b*). MeSH concentrations were determined by gas chromatography as described elsewhere (Kiene 1996).

Axenic cultures of *Synechococcus* sp. WH8102, *Synechococcus* sp. WH7803, *S. elongatus* Naegeli CCMP 1630, and *S. bacillaris* Butcher WH5701 (CCMP) were grown at 24°C on seawater media with f/2-Si nutrient amendments. Light levels alternated on a 14 : 10-h light : dark cycle. During DMSP uptake and transformation experiments, unlabeled DMSP was added at 50 nmol L^{-1} along with tracer additions of ^{35}S-DMSP ($<$1 nmol L^{-1}), and cultures were incubated for 48 h. Uptake of DMSP was determined by filtering 1 mL of culture sample into 0.2-μm nylon filter, rinsing with 10 mL of isotonic seawater, and measuring ^{35}S-activity by liquid scintillation counting. Production of DMS was measured as described previously (Kiene and Linn 2000*b*).

Results

The abundance of *Synechococcus* and assimilation of sulfur from DMSP were examined in a variety of environments ranging from the estuarine waters of the Delaware Bay to the oligotrophic Sargasso Sea. *Synechococcus* made up 1–9% of total prokaryotes in this study, and their abundance ranged from 1.1 $\times$ 10^{4} cells mL^{-1} in the Mid-Atlantic Bight to 10.8 $\times$ 10^{4} cells mL^{-1} in North Carolina coastal waters (Table 1). *Synechococcus* assimilated DMSP at all stations, and $\geq$90% of *Synechococcus* assimilated DMSP at 10 of the 12 stations (Table 1). Because of the large fraction of *Synechococcus* taking up DMSP and their abundance, these cyanobacteria often accounted for a significant component

Table 1. DMSP assimilation in the light by *Synechococcus* in the Gulf of Mexico and northwest Atlantic Ocean. Mean ± standard error (SE) of 30 fields of view. NC, North Carolina; nd, not determined.

Location	Station	Lat. N	Long. W	Dissolved DMSP flux (nmol L^{-1} d^{-1})	*Synechococcus* abundance (10^4 cells mL^{-1})	*Synechococcus* abundance (% of prokaryotes)	% DMSP-assimilating cells	% DMSP silver grain area	% *Synechococcus*-assimilating DMSP
Gulf of Mexico	3	29°04′	89°45′	7.7	8.0±1.2	2±0.3	3±0.6	4±1.4	57±4.3
Gulf of Mexico	4	29°27′	86°51′	7.3	4.0±0.9	4±0.9	5±1.5	29±11	92±2.0
Gulf of Mexico	6	27°55′	83°41′	20.5	5.5±0.9	5±0.8	<1	<1	19±4.3
Gulf of Mexico	7	29°70′	87°23′	nd	8.0±1.2	8±1.2	13±2.0	30±5.7	91±2.2
Sargasso Sea	8	34°47′	68°59′	5.9	2.8±0.7	7±1.8	10±2.7	22±6.4	92±2.2
Sargasso Sea	10	31°30′	69°00′	6.8	0.8±0.2	2±0.4	3±0.7	36±6.8	89±2.6
Sargasso Sea	12	31°30′	68°59′	1.1	4.5±0.4	9±0.8	9±2.5	35±12.2	95±2.1
Sargasso Sea	14	33°33′	74°28′	5.3	2.0±0.6	2±0.6	4±1.2	18±6.0	95±2.9
NC coast	15	34°14′	76°56′	9.1	10.8±1.4	6±0.8	9±1.3	25±4.4	99±0.8
Delaware Bay	26	38°55′	75°06′	nd	1.9±0.2	1±0.1	1±0.2	2±0.5	90±3.0
Delaware coast	36	38°11′	72°59′	nd	1.6±0.2	4±0.5	5±0.6	15±2.8	99±1.0
Mid-Atlantic Bight	39	38°20′	73°28′	nd	1.1±0.7	1±0.6	2±0.7	3±1.1	97±1.7

of the DMSP-assimilating community in a variety of environments. For example, at Sta. 8 in the Sargasso Sea, 92% of *Synechococcus* assimilated DMSP, and they comprised 10% of cells assimilating DMSP (Table 1). On average, 5% of DMSP-assimilating cells were identified as *Synechococcus.*

Variation in the size (μm^2) of silver grain clusters formed during microautoradiography indicates that there were substantial differences in the per-cell assimilation of DMSP (Fig. 1). For example, at Sta. 15, the average silver grain cluster associated with *Synechococcus* was 5.9 μm^2, while the average silver grain cluster associated with other prokaryotes was significantly smaller, at 1.9 μm^2 (*t*-test; $p < 0.05$). These data indicate that DMSP assimilation per cell was higher for *Synechococcus* than for other prokaryotes. Additionally, the distribution of per-cell assimilation was different for *Synechococcus* than for the other prokaryotes. Most prokaryotes had small silver grain areas, indicating low per-cell assimilation, while a few cells had large silver grain areas (Fig. 2A). In contrast, the size of silver grain clusters was distributed more evenly among the *Synechococcus,* indicating that a large fraction of *Synechococcus* had moderate to high per-cell assimilation (Fig. 2B).

As a result of substantial differences in per-cell assimilation, the contribution of *Synechococcus* to total DMSP assimilation was determined based on the percentage of total silver grain area associated with *Synechococcus.* Using this estimate, *Synechococcus* were typically responsible for a large fraction of the total DMSP assimilation by prokaryotes. *Synechococcus* accounted for 15–36% of DMSP assimilation at 8 out of 12 stations and ≤4% at four stations (Table 1). Using flow cytometry–cell sorting, *Synechococcus* accounted for 3%, 15%, and 3% of DMSP assimilation at Sta. 26, 36, and 39, respectively, a result that agrees with estimates based on silver grain area (Table 1). The contribution of *Synechococcus* was consistently high in the Sargasso Sea but was more variable in the Gulf of Mexico (Table 1). The large contributions were surprising considering that *Synechococcus* accounted for only ≤9% of the total prokaryotic abundance. For example, *Synechococcus* made up 4% of the prokaryotic community at Sta. 6 in the Gulf of Mexico but was responsible for 29% of DMSP assimilation. *Synechococcus* accounted for a larger fraction of DMSP assimilation than would be indicated by their overall abundance at 8 of 12 stations (*t*-test; $p < 0.05$) (Fig. 3A). These data indicate that *Synechococcus* make a disproportionately large contribution to DMSP assimilation in the ocean compared to other prokaryotes.

MeSH assimilation—In addition to DMSP, the capacity to assimilate MeSH was also examined in the Gulf of Mexico and the Sargasso Sea. We found that 94–98% of *Synechococcus* assimilated MeSH at four of the seven stations (Table 2). The fraction of *Synechococcus* that assimilated MeSH was generally similar to the fraction that assimilated DMSP at each station (Table 1), although there were some differences at Sta. 4 and 6.

As with DMSP, *Synechococcus* mediated a substantial fraction of total MeSH assimilation. *Synechococcus* accounted for 2–18% of the MeSH-assimilating cells, and they were

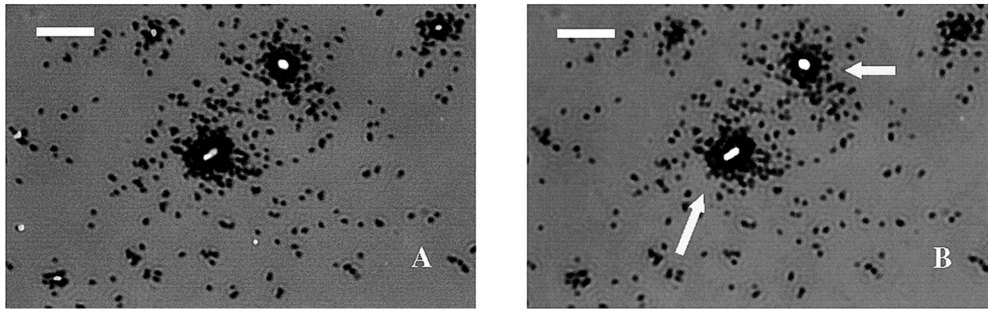

Fig. 1. A representative microautoradiogram of ^{35}S-labeled cells viewed using transmitted light and DAPI fluorescence (A) and phycoerythrin autofluorescence (B). White arrows point to phycoerythrin-containing cells identified as *Synechococcus.* The formation of dark silver grain clusters around cells indicates assimilation of ^{35}S. The scale bar = 5 μm.

responsible for 4–58% of MeSH assimilation based on total silver grain area (Table 2). MeSH assimilation per cell was higher for *Synechococcus* than for the rest of the prokaryotic community. For example, at Sta. 15, the average silver grain cluster associated with *Synechococcus* was 4.9 μm^2, whereas other prokaryotes had a significantly smaller average silver grain cluster size of 1.3 μm^2 (t-test; $p < 0.05$). Because of their high assimilation per cell, *Synechococcus* mediated a larger fraction of MeSH assimilation than would be predicted by their overall abundance at five stations (t-test; $p < 0.05$) (Fig. 3B). In contrast to DMSP and MeSH, *Synechococcus* did not assimilate DMS in the Gulf of Mexico or the Sargasso Sea (data not shown).

Influence of light on assimilation—The effect of light on DMSP and MeSH assimilation was examined in the Sargasso Sea and North Carolina coastal waters. The proportion of *Synechococcus* assimilating DMSP was 15–16% greater when incubated in the light than in the dark at Sta. 8 and 10 in the Sargasso Sea (t-test; $p < 0.05$), but there was no difference between light and dark incubations at Sta. 14 (Fig. 4A). In North Carolina coastal waters (Sta. 15), 99% of *Synechococcus* assimilated DMSP in light incubations, compared to only 21% in the dark incubations. Similarly, a larger fraction of *Synechococcus* assimilated MeSH in the light (96%) than in the dark (26%) at Sta. 15 (Fig. 4B). However, there were no differences in the number of *Synechococcus* assimilating MeSH during light and dark incubations at Sta. 8 and 14. These data indicate that light can stimulate the assimilation of DMSP and MeSH by *Synechococcus,* but the effect varies.

DMSP uptake by axenic cultures—The capacity to take up or lyse DMSP was examined in four axenic strains of *Synechococcus. Synechococcus elongatus* (CCMP 1630) and *S. bacillaris* (WH5701) did not take up DMSP. In contrast, the phycoerythrin-containing strains *Synechococcus* WH7803 and *Synechococcus* WH8102 did transport DMSP. Of the four strains tested, only *S. elongatus* lysed DMSP to produce DMS in culture (data not shown).

Discussion

Synechococcus are often abundant, widely distributed, and contribute substantially to primary production in tropical and temperate waters (Waterbury et al. 1979; Li 1994). Some *Synechococcus* strains also have the capacity to consume certain dissolved organic compounds (Cuhel and Waterbury 1984; Paerl 1991), but because of their autotrophic lifestyle, the contribution of *Synechococcus* to the turnover of dis-

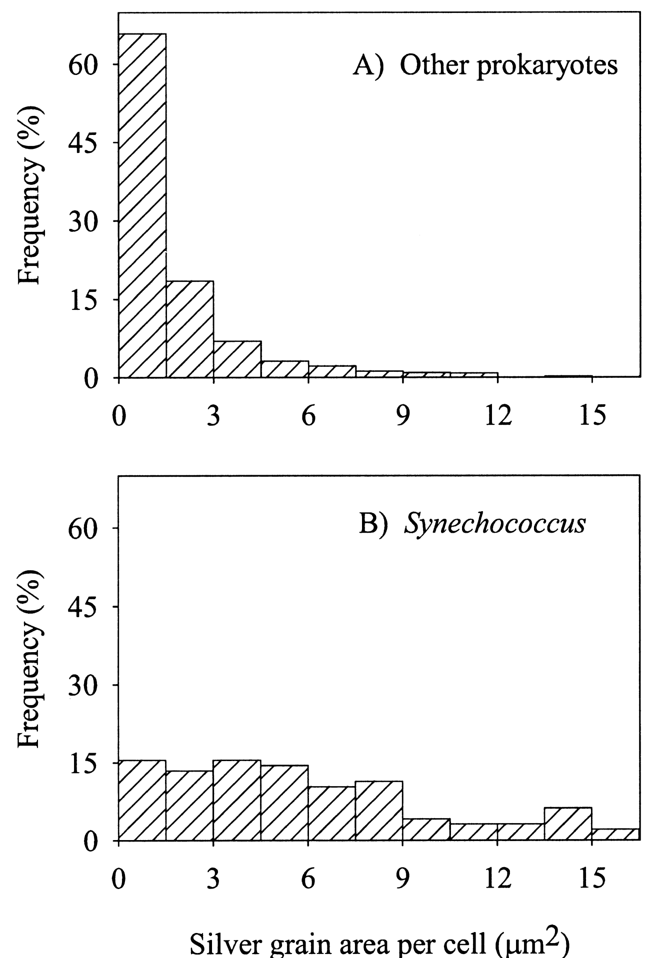

Fig. 2. Frequency distribution of silver grain sizes (μm^2) per cell in (A) other prokaryotes and (B) *Synechococcus* in North Carolina coastal waters (Sta. 15). Silver grain areas were grouped into bins at 1.5-μm^2 increments. These distributions are representative of those from other stations.

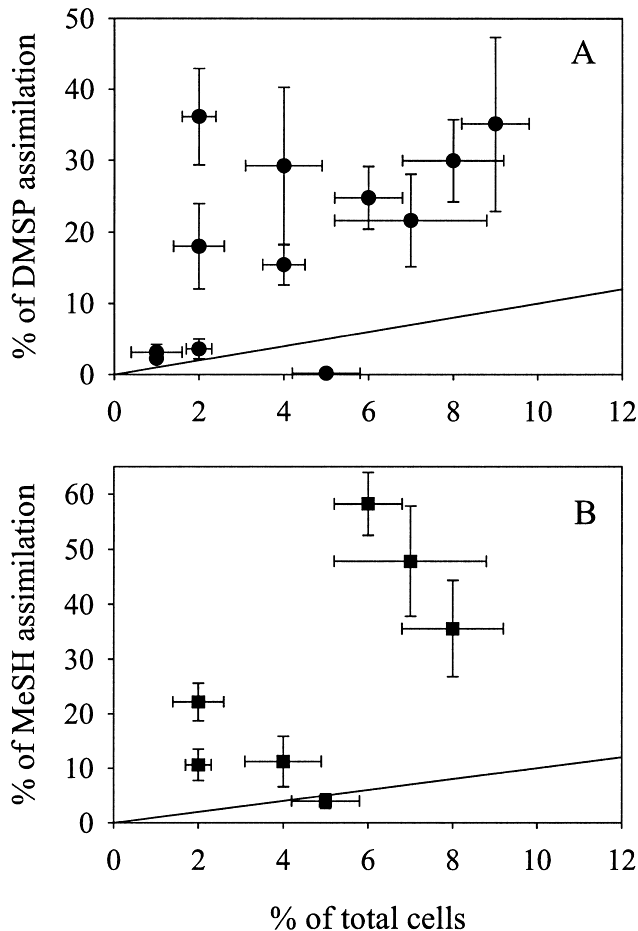

Fig. 3. Contribution of *Synechococcus* to DMSP and MeSH assimilation in the northwest Atlantic Ocean and Gulf of Mexico. (A) The percentage of DMSP assimilation mediated by *Synechococcus* in the light versus the abundance of *Synechococcus* in the total prokaryotic community (DAPI-positive cells). (B) The percentage of MeSH assimilation mediated by *Synechococcus* in the light versus abundance of *Synechococcus* in the total prokaryotic community. Error bars indicate standard error (SE) of 30 fields of view. A 1 : 1 line bisects the graph.

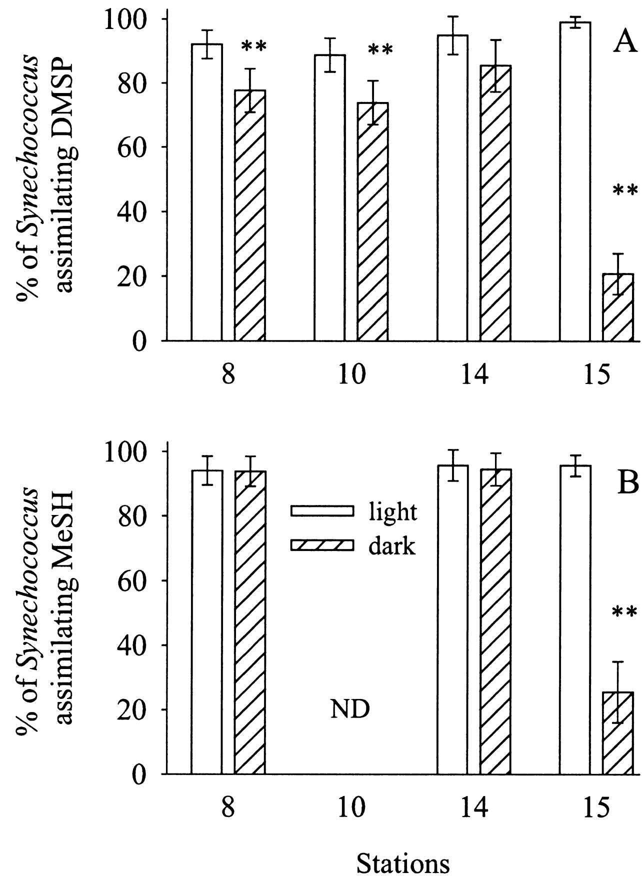

Fig. 4. Assimilation of (A) DMSP and (B) MeSH by *Synechococcus* during light and dark incubations. Statistically significant differences (*t*-test; $p < 0.01$) are indicated (**). Data presented as mean ± 95% confidence intervals. ND, not determined.

solved organic compounds was expected to be minimal. However, we found *Synechococcus* were major consumers of the organic-sulfur compounds DMSP and MeSH. On average, *Synechococcus* were responsible for about 20% of DMSP assimilation by prokaryotes, which is similar to the contributions by other abundant bacterial groups, such as the *Roseobacter* and SAR11 clade (Malmstrom et al. 2004*a,b*; Vila et al. 2004). Because of their high degree of assimilation and widespread distribution, *Synechococcus* are likely to be an important sink for DMSP in the ocean.

To better understand the contribution of *Synechococcus* to DMSP flux, it is informative to identify the strains of *Synechococcus* that metabolize DMSP. In this study, two of four axenic strains of *Synechococcus* took up DMSP. *Synechococcus* WH8102 and WH7803, which can transport DMSP, are phycoerythrin-containing strains that belong to the marine cluster A phylogenetic group of cyanobacteria (Rocap et al. 2002). In contrast, the two phycocyanin-containing strains, *S. elongatus* and *S. bacillaris,* did not take up DMSP. Results from field studies indicate that a variety of *Synechococcus* strains metabolize DMSP. Roughly 90% of *Synechococcus* assimilated DMSP at 10 of 12 stations, and it is likely that these *Synechococcus* populations differed among these locations. Indeed, previous work using immunofluorescent labeling indicates that various *Synechococcus* strains are not distributed uniformly among different environments (Toledo and Palenik 2003). If in fact *Synechococcus* strains differed among stations, then the capacity to utilize DMSP was common to many *Synechococcus* strains in this study. However, only 57% and 19% of *Synechococcus* assimilated DMSP at Sta. 3 and 6, respectively, indicating that the capacity to assimilate DMSP may not be universally distributed among strains of *Synechococcus.* Additional work on axenic cultures and further characterization of DMSP-assimilating *Synechococcus* in the oceans will be useful in determining the distribution of DMSP metabolism within the *Synechococcus.*

Genomic analyses may also provide information about DMSP use by *Synechococcus.* The genes involved in the DMSP assimilation pathway are currently unknown, although some data are available regarding genes encoding DMSP-transporters. Investigations on cultured heterotrophic

Table 2. Assimilation of MeSH by *Synechococcus* in the Gulf of Mexico and northwest Atlantic Ocean. Mean ± standard error (SE) of 30 fields of view. NC, North Carolina; nd, not determined.

Location	Station	MeSH (nmol L^{-1})	% MeSH-assimilating cells	% MeSH silver grain area	% *Synechococcus*-assimilating MeSH
Gulf of Mexico	3	nd	7±1.6	11±2.9	44±6.9
Gulf of Mexico	4	0.18	10±3.3	11±4.6	77±3.7
Gulf of Mexico	6	0.13	2±0.7	4±1.4	52±4.4
Gulf of Mexico	7	nd	7±2.4	36±8.8	98±1.4
Sargasso Sea	8	0.57	13±2.3	48±10	94±2.2
Sargasso Sea	10	0.32	nd	nd	nd
Sargasso Sea	12	0.31	nd	nd	nd
Sargasso Sea	14	0.81	6±1.0	22±3.4	96±2.4
NC coast	15	0.34	18±1.0	58±5.7	96±1.6

bacteria confirm that DMSP and glycine betaine can share the same transporter (Kempf and Bremer 1998), and competitive inhibition assays indicate that DMSP and glycine betaine are taken up into cells by the same transport system in marine bacterial communities (Kiene et al. 1998). Palenik et al. (2003) reported that the genome of *Synechococcus* sp. WH8102 contains putative glycine betaine transporter genes. Additional BLAST analyses reveal that these transporter genes are homologous to opuAA, opuAB, and opuAC (51%, 46%, and 31% identity based on protein sequence, respectively), which are components of the OpuA transporter that recognizes DMSP and glycine betaine in *Bacillus subtilis* (Kempf and Bremer 1998). In this study, work with axenic cultures confirmed that *Synechococcus* WH8102 can transport DMSP and indicates that this strain can utilize DMSP. The presence of similar DMSP/glycine betaine transporters in other cyanobacteria genomes may provide insight into the role of marine cyanobacteria in DMSP metabolism.

One challenge in investigating DMSP metabolism by *Synechococcus* is determining if the *Synechococcus* assimilated DMSP directly or if some *Synechococcus* assimilated byproducts of ^{35}S-DMSP, such as ^{35}S-DMS and ^{35}S-MeSH produced by other organisms. In this study, *Synechococcus* did not assimilate ^{35}S-DMS. *Synechococcus* did assimilate ^{35}S-MeSH, which is consistent with the capacity to assimilate DMSP, since the incorporation of DMSP-derived S into biomass is thought to proceed through a MeSH intermediate (Kiene et al. 1999). It is unlikely that a large fraction of *Synechococcus* were only assimilating MeSH because *Synechococcus* incorporated large amounts of ^{35}S-label compared to nearly all other prokaryotes (Fig. 2). Indeed, the other DMSP-assimilating bacteria would have to assimilate DMSP with very low efficiency and release most of the MeSH they produce in order to account for the large differences between ^{35}S found in *Synechococcus* and other prokaryotes. In addition, the ^{35}S-label found *Synechococcus* was not the result of retention of DMSP as an osmolyte, because the microautoradiography preparation causes cells to lose un-incorporated solutes (Kiene and Lynn 1999). Although assimilation of MeSH produced by other prokaryotes remains a possibility, direct DMSP assimilation by *Synechococcus* is the most parsimonious interpretation of the data.

An important implication of DMSP use by *Synechococcus* is the possible impact of light on DMSP consumption rates. As a phototrophic organism, *Synechococcus* experiences diel variations in energy supply over the daily light : dark cycle, and many physiological processes are coupled to this periodicity. For example, DNA synthesis and cell division in *Synechococcus* are linked to light : dark cycles (Jacquet et al. 2001). Transcription of ribulose bisphosphate carboxylase/oxygenase and glutamine synthetase also follows diel rhythms (Wyman 1999). If carbon and nitrogen assimilation are affected by light : dark cycles, then rates of DMSP assimilation may also vary on a diel basis. In this study, light affected DMSP assimilation by *Synechococcus* at three of four locations. Diel variation in *Synechococcus* activity could produce variation in total DMSP consumption rates over the light : dark cycle.

High uptake of DMSP was surprising in part because concentrations of sulfate are typically 10^7-fold greater than concentrations of DMSP, indicating that *Synechococcus* would rely on sulfate as a source of S. However, to assimilate S from sulfate into protein, bacteria must first expend energy to reduce sulfate. Heterotrophic bacteria are hypothesized to use DMSP over sulfate because it is more energy efficient to incorporate the reduced S from DMSP directly than to assimilate S from sulfate (Kiene et al. 2000). As with heterotrophic bacteria, *Synechococcus* may also utilize DMSP and MeSH because it is energetically efficient to assimilate these reduced compounds. By analogy, *Synechococcus* preferentially take up reduced nitrogen in the form of ammonium, as compared to nitrate and nitrite (Moore et al. 2002), and *Prochlorococcus,* a close relative of *Synechococcus* (Rocap et al. 2002), cannot use nitrate at all (Moore et al. 2002). These data indicate that the assimilation of reduced compounds, both inorganic and organic, is an effective strategy for marine cyanobacteria to increase metabolic efficiency.

The observation of organic sulfur uptake by *Synechococcus* furthers our understanding of the role photoheterotrophy plays in marine biogeochemical cycles. Recent reports indicate that photoheterotrophic activities by cyanobacteria could be important to the flux of DOM. For example, photoheterotrophy in *Prochlorococcus* may explain light stimulation of leucine incorporation in the North Pacific gyre

(Church et al. 2004). *Prochlorococcus* were also responsible for about 30% of methionine turnover in the Arabian Sea (Zubkov et al. 2003). Some *Synechococcus* also have the capacity to assimilate amino acids (Willey and Waterbury 1989; Paerl 1991), although the contribution of *Synechococcus* to methionine uptake was <5% in the Arabian Sea (Zubkov et al. 2003). These data indicate that substantial heterotrophic uptake in *Synechococcus* may be limited to a few components of the DOM pool. Determining the contribution of *Synechococcus* and other cyanobacteria to the turnover of specific DOM components, as well as to the total DOM pool, is necessary to better understand the role of cyanobacteria in marine biogeochemical cycles.

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