Limnol. Oceanogr., 51(1), 2006, 280–287

Vertical distributions of thiosulfate and sulfite in the Cariaco Basin

Meredith K. Hayes[1] *and Gordon T. Taylor*
Marine Sciences Research Center, Stony Brook University, Stony Brook, New York 11794-5000

Yrene Astor
Estación de Investigaciones Marinas de Isla Margarita, Fundación de la Salle de Ciencias Naturales, Apartado 144,
Punta de Piedras, Edo. Nueva Esparta, Venezuela

Mary I. Scranton[2]
Marine Sciences Research Center, Stony Brook University, Stony Brook, New York 11794-5000

Abstract

Water column profiles of $S_2O_3^{2-}$ and SO_3^{2-} were obtained in January and May 2004 in the eastern portion of the Cariaco Basin (10°30′N 64°40′W). Mean concentrations in the chemocline (250 to 450 m) decreased from 2.3 ± 1.1 μmol L^{-1} $S_2O_3^{2-}$ in January to 1.4 ± 0.3 μmol L^{-1} $S_2O_3^{2-}$ in May and from 4.8 ± 2.9 μmol L^{-1} SO_3^{2-} in January to 2.7 ± 0.7 μmol L^{-1} SO_3^{2-} in May. Integrated over this same depth interval, $S_2O_3^{2-}$ was 459 mmol m^{-2} in January and 287 mmol m^{-2} in May and SO_3^{2-} was 799 mmol m^{-2} in January and 574 mmol m^{-2} in May. During this time, the integrated chemoautotrophic production in the chemocline decreased from 26 to 13 mmol C m^{-2} d^{-1}. The concurrent decreases in $S_2O_3^{2-}$ and SO_3^{2-} inventories and chemoautotrophic production are consistent with a decrease in the frequency or intensity of intrusion events. $S_2O_3^{2-}$ and SO_3^{2-} were present in the oxic portion of the water column, suggesting lateral transport from shallower areas containing H_2S. Calculations suggest that sufficient O_2 and H_2S can be supplied to the interface by advection and diffusion to form $S_2O_3^{2-}$ in sufficient quantities to support the observed levels of chemoautotrophic production. However, apparently $<2\%$ of the H_2S needed to form the observed amount of $S_2O_3^{2-}$ can be produced from the degradation of organic carbon sinking through the interface at this site. This suggests either that the H_2S or carbon flux must be supplied laterally, perhaps from shallower areas (not yet sampled), or that chemoautotrophic production near the interface is overestimated.

The Cariaco Basin, located on the continental shelf of Venezuela, is the largest truly marine anoxic basin on earth. The basin is made up of two 1,400-m-deep subbasins separated by a saddle reaching 900 m and is connected to the southern Caribbean Sea by two channels at the eastern and western ends of the basin that reach depths of no more than ~140 m. The waters above sill depth interact with the open sea, but vertical mixing below ~100 m is inhibited by the geometry of the basin and a strong pycnocline (Astor et al. 2003). Seasonal upwelling is usually observed in the surface waters between November and May, and annual primary production is estimated to vary between 500 and 600 g C m^{-2} yr^{-1} (Muller-Karger et al. 2001). Because of the restricted circulation and the degradation of the large amount of organic matter produced in the region, oxygen is completely consumed, and the waters below ~300 m are permanently sulfidic.

The pathways of organic matter decomposition in the basin are reflected in vertical distributions of the major electron acceptors and their reduced products (Ho et al. 2004). Autotrophic H_2S- and $S_2O_3^{2-}$-oxidizing denitrifiers, $S_2O_3^{2-}$-oxidizing Mn reducers, S^0 disproportionaters, and SO_4^{2-} reducers have all been cultivated from selective enrichment experiments with waters from near the O_2/H_2S interface in the Cariaco Basin (Madrid 2000). Elevated concentrations of autotrophic and heterotrophic bacteria have also been found near the interface of other anoxic water columns, including the Black Sea and Framvaren Fjord (Karl 1978; Sorokin et al. 1995; Vetriani et al. 2003).

Because the interface is located well below the illuminated layer, the interface community of the Cariaco Basin must be supported by chemoautotrophs that use a variety of oxidants and reductants to obtain energy for the fixation of CO_2 (Taylor et al. 2001; Ho et al. 2004). Chemoautotrophs conserve chemical energy and reducing power in the form of ATP and NADPH, respectively. Three moles of ATP and 2 mol of NADPH are needed for each mole of CO_2 fixed, at a basic cost of 191 kJ (Thauer et al. 1977). ATP synthesis is coupled to the oxidation of an electron donor, whereas

---

[1] Present address: GZA GeoEnvironmental of New York, 440 Ninth Avenue, 18th Floor, New York, New York 10001.

[2] Corresponding author (mscranton@notes.cc.sunysb.edu).

Acknowledgments

We are indebted to the personnel of the Fundación La Salle de Ciencias Naturales, Estación de Investigaciones Marinas Isla Margarita (FLASA/EDIMAR) and the crew of the R/V *Hermano Ginés* for their enthusiasm and professional support. Ramon Varela provided unwavering leadership in coordinating and carrying out field expeditions. Russell Cuhel provided valuable advice regarding development of the HPLC method. We also thank Laura Lorenzoni for her assistance with the figures and the reviewers of this manuscript for their many helpful comments. This work was supported by the National Science Foundation through grants OCE 0118491 and OCE 0326175 to M.I.S. and G.T.T. and, in Venezuela, by Proyecto Cariaco by FONACIT (Fondo Nacional de Ciencia, Tecnologia e Innovacion) under grant 2000001702 to Ramon Varela of Fundacion La Salle. This is contribution 1305 from the Marine Sciences Research Center, Stony Brook University.

NADPH is generated from reverse electron transport reactions, metabolic mechanisms required for generating the necessary negative redox potential. A variety of reactions that yield enough energy for the fixation of CO_2 are listed in Eqs. 1 to 4. In particular, the most common sulfur compounds used as electron donors for ATP synthesis in chemoautotrophs are H_2S, S^0, and $S_2O_3^{2-}$ (Madigan et al. 2003).

$$H_2S + 2O_2 \rightarrow SO_4^{2-} + 2H^+ \qquad \Delta G^\circ = -798 \text{ kJ mol}^{-1} \tag{1}$$

$$H_2S + \frac{1}{2}O_2 \rightarrow S_0 + H_2O \qquad \Delta G^\circ = -209 \text{ kJ mol}^{-1} \tag{2}$$

$$S^0 + H_2O + \frac{3}{2}O_2 \rightarrow SO_4^{2-} + 2H^+ \qquad \Delta G^\circ = -587 \text{ kJ mol}^{-1} \tag{3}$$

$$S_2O_3^{2-} + H_2O + 2O_2 \rightarrow 2SO_4^{2-} + 2H^+ \qquad \Delta G^\circ = -818 \text{ kJ mol}^{-1} \tag{4}$$

In these reactions, O_2 is the electron acceptor. However, alternative electron acceptors, such as NO_3^-, can also be used for chemoautotrophic H_2S oxidation (Jannasch and Mottl 1985).

For chemoautotrophic production to proceed, both electron donors and acceptors are required. If vertical transport of these species controls supply, fluxes of H_2S, O_2, and NO_3^- calculated from vertical concentration profiles and estimates of vertical eddy diffusion should balance biological demand calculated from the measured chemoautotrophic production. However, in the Cariaco Basin, Taylor et al. (2001) found that measured rates of chemoautotrophic production were much higher than could be supported by the diffusive supply of H_2S, O_2, and NO_3^- to the interface. Vertical diffusive H_2S fluxes could provide between 0.7% and 2.8% of the demand for reductant, whereas O_2 and NO_3^- supply could provide 2.5–4.3% and 1.0–1.4%, respectively, of the demand for oxidant.

Horizontal intrusions of O_2-rich water from the Caribbean Sea have been reported in the Cariaco Basin (Scranton et al. 2001; Astor et al. 2003), and these could increase supply of oxidant. In addition, we considered it probable that reductants other than H_2S, including intermediate oxidation sulfur species like $S_2O_3^{2-}$ and SO_3^{2-}, might be important in chemoautotrophic production and thus that measurement of H_2S alone would underestimate reductant supply. The reaction of O_2 with H_2S in the basin could form sulfur species of intermediate oxidation states, such as $S_2O_3^{2-}$, SO_3^{2-}, and S^0, that could be used in disproportionation reactions, such as those in Eqs. 5 and 6, in which a compound is transformed into two compounds of lower and higher oxidation states, or as a reductant in reactions like Eqs. 7 or 8, in which metal oxides serve as the oxidant (Nealson and Myers 1992; Zhang and Millero 1993*a*; Janssen et al. 1996).

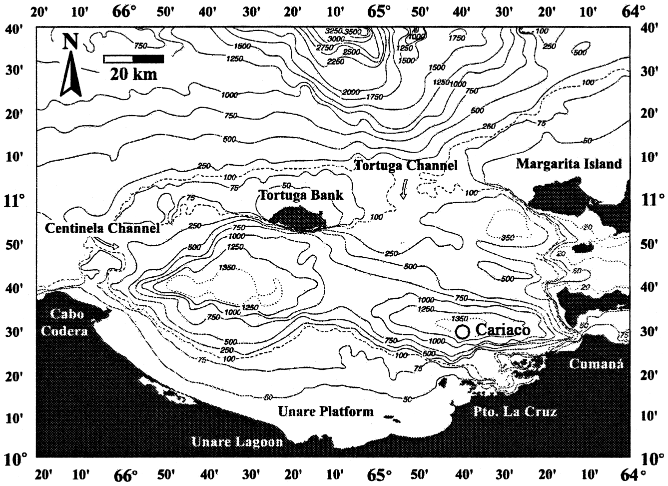

Fig. 1. Map of Cariaco Basin with station for CARIACO time series (10°30′N, 64°40′W) indicated by the bull's-eye. Isobaths are in meters.

$$S_2O_3^{2-} + H_2O \rightarrow SO_4^{2-} + H_2S \qquad \Delta G_f^\circ = -21.9 \text{ kJ mol}^{-1} \tag{5}$$

$$SO_3^{2-} + \frac{1}{2}H^+ \rightarrow \frac{3}{4}SO_4^{2-} + \frac{1}{4}H_2S \qquad \Delta G_f^\circ = -58.9 \text{ kJ mol}^{-1} \tag{6}$$

$$S^0 + \frac{2}{3}FeOOH \rightarrow \frac{1}{3}SO_4^{2-} + \frac{2}{3}FeS + \frac{2}{3}H^+ \qquad \Delta G_f^\circ = -34.4 \text{ kJ mol}^{-1} \tag{7}$$

$$S_2O_3^{2-} + 4MnO_2 + 6H^+ \rightarrow 4Mn^{2+} + 2SO_4^{2-} + 3H_2O \qquad \Delta G_f^\circ = -733 \text{ kJ mol}^{-1} \tag{8}$$

In these examples, ΔG_f° is the standard Gibb's free energy at pH 7. The above reactions are schematic and might not reflect exact reaction pathways. However, these and similar reactions would yield sufficient energy to support chemoautotrophic growth. To investigate the potential importance of $S_2O_3^{2-}$ and SO_3^{2-} in the redox budget of the Cariaco Basin, measurements of $S_2O_3^{2-}$ and SO_3^{2-} were made during January and May 2004.

Methods

The CARIACO time series station is in the eastern subbasin at 1,400 m depth (10°30′N, 64°40′W; Fig. 1). Water samples were collected from the Cariaco Basin during 20 January 2004 (CAR-96) and 18 May 2004 (CAR-100) as part of the ongoing CARIACO time series program. All discrete water samples were collected with Teflon-lined 8-liter Niskin bottles mounted on a Seabird Rosette system. Before drawing samples from waters depleted in oxygen, the Niskin bottles were fitted at the top with a source of nitrogen gas to minimize oxygen contamination.

Profiles of temperature, salinity, and O_2 were obtained with a Seabird CTD system with attached YSI oxygen probe.

In addition to measurements made by the YSI probe, O_2 concentrations were also determined in discrete samples by standard Winkler titrations (Aminot 1983). Detection limit for O_2 in this system has been estimated at ~2–5 μmol L^{-1} (Scranton et al. 2001).

Samples for H_2S were collected with a glass gas-tight syringe from a continuously flowing stream of water to minimize contact with the atmosphere and then immediately injected into a vial containing $ZnCl_2$. The resulting ZnS precipitate was derivatized and analyzed with a modification of the method of Cline (1969). The detection limit was estimated to be 1–2 μmol L^{-1} (Scranton et al. 2001).

Two techniques were used for $S_2O_3^{2-}$ and SO_3^{2-} samples. In January, triplicate samples for $S_2O_3^{2-}$ and SO_3^{2-} were collected in plastic rubberless syringes, derivatized, and analyzed following the procedure of Vairavamurthy and Mopper (1990). Time between collection and derivatization was on the order of 2–3 min. For each sample, 20 mL of water was added to a vial containing sodium acetate buffer and immediately derivatized with 2,2′dithiobis(5-nitropyridine). The derivatives were stored on Sep-Pak tC_{18} cartridges and frozen onboard. After return to the lab in Venezuela, argon gas was blown through the cartridges and field blanks were prepared from distilled water and the same reagents used onboard. $S_2O_3^{2-}$ and SO_3^{2-} analysis by high-performance liquid chromatography (HPLC) was completed at Stony Brook within 3 weeks of sample collection.

In May, quadruplicate 10-mL samples were taken with a 10-mL glass gas-tight syringe from a continuously flowing stream of water. Samples were derivatized immediately (on the order of seconds) after collection, and derivatives were transferred to Sep-Pak tC_{18} cartridges and frozen onboard. As before, after return to the lab in Venezuela, argon gas was blown through the cartridges. In May, field blanks were prepared onboard ship after each cast with the reagents used for that cast. The derivatives were analyzed at Stony Brook by HPLC within 6 weeks of collection.

The detection limit for $S_2O_3^{2-}$ and SO_3^{2-} was calculated as the average of quadruplicate field blanks. In January, the field blank was 0.8 ± 0.3 and 1.5 ± 0.5 μmol L^{-1} for $S_2O_3^{2-}$ and SO_3^{2-}, respectively. In May, the values were 0.6 ± 0.03 and 2.2 ± 0.2 μmol L^{-1} for $S_2O_3^{2-}$ and SO_3^{2-}, respectively.

Chemoautotrophic production was measured by ^{14}C-bicarbonate incorporation into particles as described previously by Taylor et al. (2001). After addition of the sample into a 40-mL ground glass-stoppered bottle, 200 μL of chilled N_2-purged ^{14}C-bicarbonate in an alkaline brine (pH 9.5, salinity 60) was injected (Tuttle and Jannasch 1973) into it. Samples were incubated for 14 to 20 h and particles were collected on 0.22-μm cellulosic membranes. Filters were purged of unassimilated ^{14}C in a saturated HCl atmosphere for >1 h, then dried and suspended in Hionic-Fluor scintillation cocktail and radioassayed. Data were corrected for abiotic sorption by use of samples processed immediately after introduction of the radiotracer. Rates of dark ^{14}C-assimilation were corrected for isotopic discrimination and normalized (μmol C L^{-1} d^{-1}) by use of values of dissolved inorganic carbon derived from pH, temperature, and alkalinity measurements obtained as a part of the standard CARIACO data set (unpubl. data).

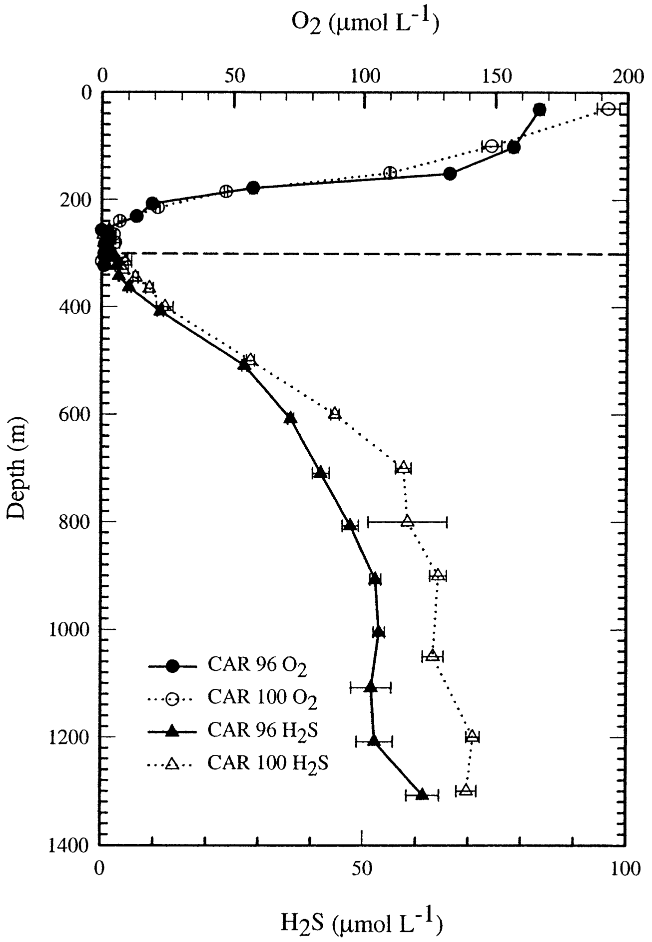

Fig. 2. O_2 and H_2S profiles on 20 January (CAR-96) and 18 May 2004 (CAR-100). Horizontal dashed line represents the suboxic–anoxic interface.

Results

O_2 and H_2S—In January, O_2 concentrations decreased from ~190 μmol L^{-1} in the surface water to <5 μmol L^{-1} at 250 m (Fig. 2). The O_2 profile in May was similar, with concentrations decreasing from ~196 μmol L^{-1} at the surface to <5 μmol L^{-1} at 290 m. During both cruises, the interface (defined as the first appearance of H_2S) was at a depth of 300 m, leaving a zone 50 (January) to 10 m (May) thick where neither O_2 nor H_2S was detected. Below the interface, H_2S concentrations increased to a bottom water concentration of ~60 μmol L^{-1}. May H_2S concentrations increased more rapidly with depth, reaching a bottom water concentration of 68 μmol L^{-1}.

Chemoautotrophic production—In January, chemoautotrophic production was highest (0.31 μmol C L^{-1} d^{-1}) at 360 m and exhibited a secondary maximum (0.14 μmol C L^{-1} d^{-1}) at 310 m (Fig. 3). In May, a broad maximum was observed at and slightly below the interface, with a value of

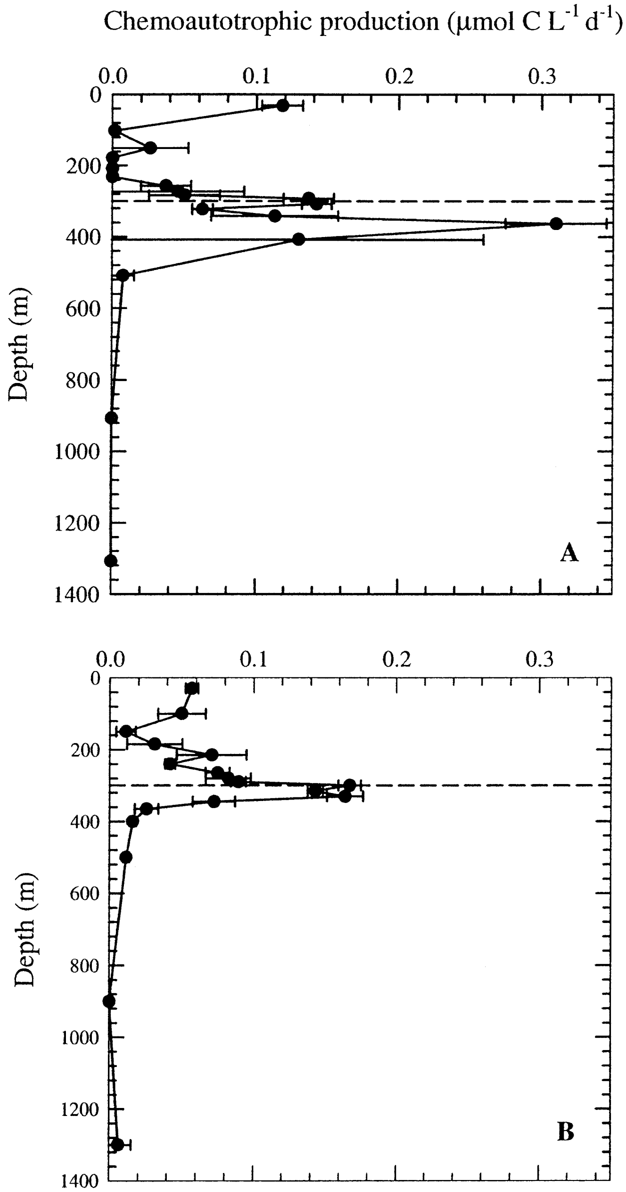

Fig. 3. Chemoautotrophic production on (A) 20 January 2004 (CAR-96) and (B) 18 May 2004 (CAR-100). Horizontal dashed lines represent the suboxic–anoxic interface. Symbols and error bars represent means and 1 standard deviation of triplicate incubation vials.

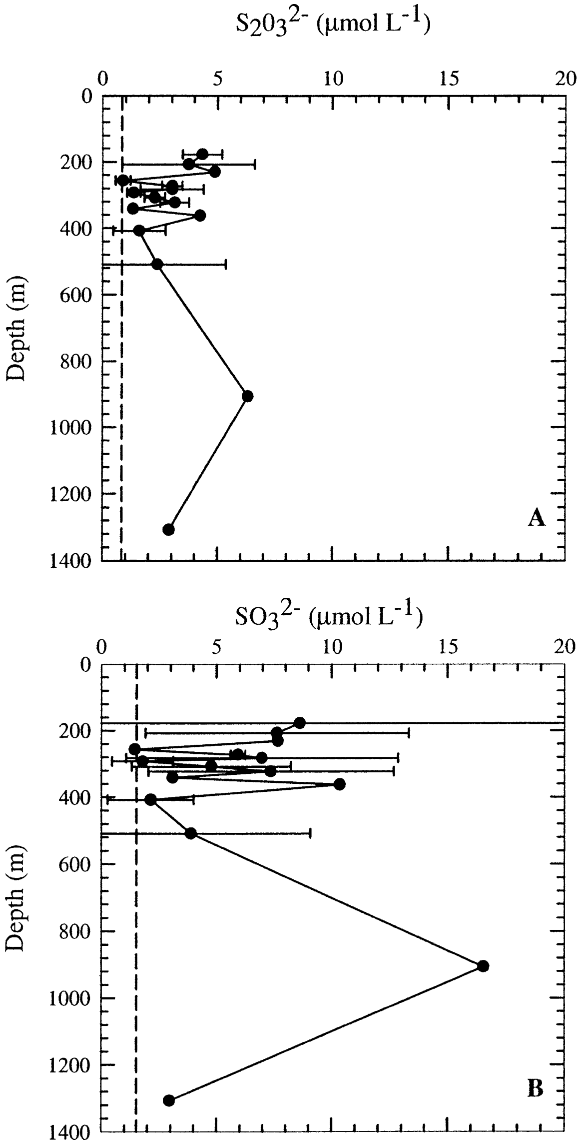

Fig. 4. (A) $S_2O_3^{2-}$ and (B) SO_3^{2-} profiles from 20 January 2004 (CAR-96). Plotted data are not blank corrected. Vertical dashed lines represent the values of the field blank. At depths with no error bar, the value was obtained from a single sample.

0.14–0.16 μmol C L^{-1} d^{-1} (Fig. 3). Integrated production in the chemocline (250–450 m) decreased from 26 to 13 mmol C m^{-2} d^{-1} between January and May.

$S_2O_3^{2-}$ and SO_3^{2-}—$S_2O_3^{2-}$ and SO_3^{2-} were present at all measured depths in January, with values ranging from 0.9 to 6.3 and 1.5 to 16.6 μmol L^{-1}, respectively (Fig. 4A). Although $S_2O_3^{2-}$ and SO_3^{2-} also seemed to be present in the bottom waters, concentrations of H_2S below 400 m were 10 μmol L^{-1} and higher. Laboratory analysis of seawater solutions of 10 μmol L^{-1} H_2S, treated like the shipboard samples, showed concentrations of $S_2O_3^{2-}$ and SO_3^{2-} at or below our detection limits (data not shown). At higher concentrations of H_2S, it is possible that $S_2O_3^{2-}$ and SO_3^{2-} could have been formed by oxidation during sampling. Therefore, data below 400 m are not discussed here.

Ignoring the deeper samples, the maximum in $S_2O_3^{2-}$ was found at 360 m, coincident with the peak in chemoautotrophic production. In the oxic and suboxic zone, where no H_2S was present, $S_2O_3^{2-}$ concentrations were as high as 4.9 μmol L^{-1}. The SO_3^{2-} values are harder to interpret than the $S_2O_3^{2-}$ data because of the large uncertainty associated with the concentrations and the relatively high blanks (indicated by the vertical line in Fig. 4B). However, the average values for each depth seem to follow the same trends observed in the $S_2O_3^{2-}$ profile.

In May 2004, $S_2O_3^{2-}$ concentrations were similar to those found during January, ranging from 0.7 to 3.2 μmol L^{-1}

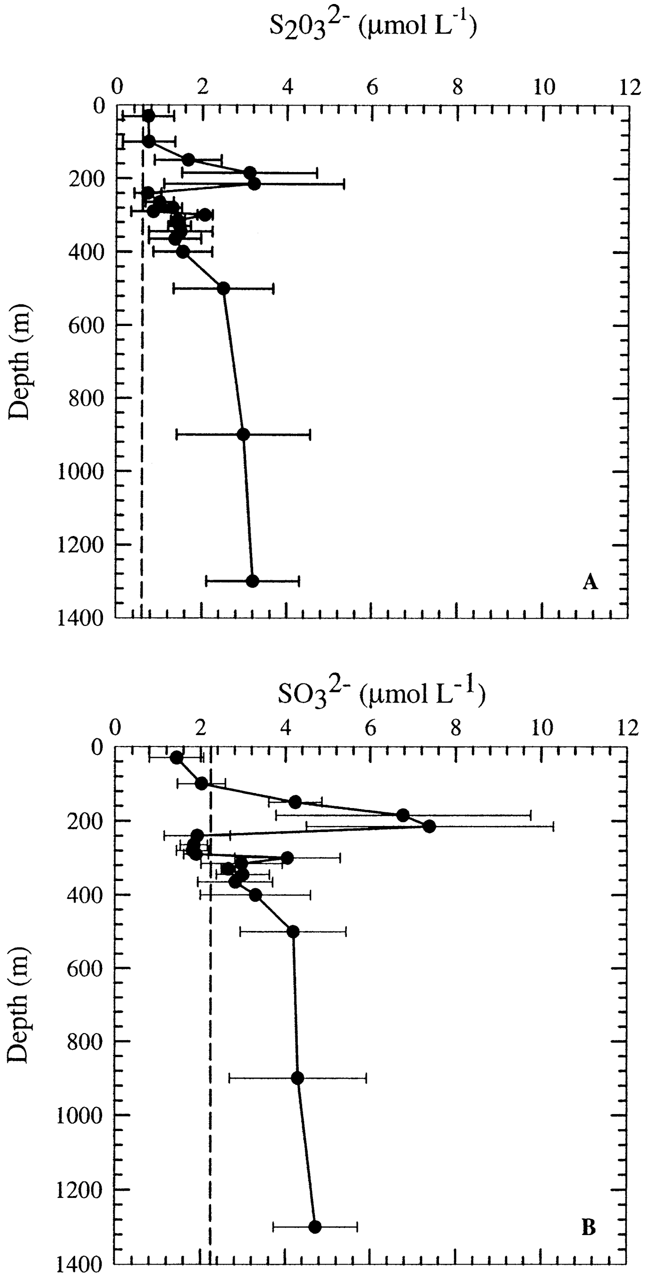

Fig. 5. (A) $S_2O_3^{2-}$ and (B) SO_3^{2-} profiles from 18 May 2004 (CAR-100). Plotted data are not blank corrected. Vertical dashed lines represent the values of the field blanks.

(Fig. 5A). The reproducibility of SO_3^{2-} measurements was better in May, presumably because of the different sampling method, and concentrations ranged from 1.4 to 7.4 μmol L^{-1} (Fig. 5B). The trends in both the $S_2O_3^{2-}$ and SO_3^{2-} profiles were similar. A broad maximum was apparent above the interface at 200 m, a secondary peak occurred at the interface, and concentrations remained relatively constant from the interface to 400 m. Linear regressions of $S_2O_3^{2-}$ concentration against SO_3^{2-} concentration showed that the two species were highly correlated, with $r = 0.988$ ($p < 0.0001$) for January and $r = 0.983$ ($p < 0.0001$) for May.

In both Figs. 4 and 5, the error bars are a measure of analytical variability. Triplicate samples were collected in January and quadruplicate samples were collected in May. However because of instrument problems, in some cases data are not available for all replicates. The error bars represent the range of values for duplicate analyses and the standard deviation when three or more samples were measured from a single depth.

Discussion

Microbiological implications of $S_2O_3^{2-}$ and SO_3^{2-}—Previous researchers have reported the growth of bacteria by disproportionation of $S_2O_3^{2-}$, SO_3^{2-}, or S^0 into H_2S and SO_4^{2-} (Bak and Cypionka 1987; Jørgensen and Bak 1991; Janssen et al. 1996). Most of the organisms known to metabolize inorganic sulfur compounds by a disproportionation mechanism are gram-negative bacteria that cluster within the delta subclass of Proteobacteria (Kramer and Cypionka 1989). Bak and Pfenning (1987) showed that *Desulfovibrio sulfodismutans* strain ThAc01 and strain NTA3 obtain energy for growth from $S_2O_3^{2-}$ or SO_3^{2-} disproportionation. Strain ThAc01 required acetate in addition to CO_2 but was unable to use acetate as an oxidizable energy source. Strain NTA3 was able to grow without an added carbon source and therefore exhibited true chemoautotrophic growth. For growth by S^0 disproportionation, free H_2S had to be removed from the cultures. Jørgensen et al. (1991) also demonstrated the presence of truly autotrophic sulfur-oxidizing bacteria in the Black Sea that used S^0 or $S_2O_3^{2-}$ as energy sources.

Sulfur isotope data has confirmed the environmental importance of disproportionation in the sulfur cycle of euxinic waters. Because of mass-related differences in the properties of the isotopes, which result in different reaction rates, disproportionation preferentially oxidizes ^{34}S and reduces ^{32}S. In culture studies, isotope fractionations during disproportionation are 7–20‰ (Canfield et al. 1998). Sørensen and Canfield (2004) showed that isotopic depletion of water column H_2S in the Mariager Fjord could be explained by $S_2O_3^{2-}$ or S^0 formation followed by disproportionation to H_2S and SO_4^{2-}. Most probable number counts confirmed that cells capable of growing by disproportionation of these compounds were present at all depths in the fjord.

In addition to disproportionation reactions, sulfur compounds of intermediate oxidation states might also be important in reactions involving particulate metal oxides as the electron acceptors. Culture studies have shown that organisms are capable of anaerobic growth through the reduction of Mn and Fe oxides (DiChristina et al. 1988; Lovley and Phillips 1988; Myers and Nealson 1988). Madrid (2000) demonstrated that cultures from the chemocline of the Cariaco Basin could reduce MnO_2 autotrophically at the expense of $S_2O_3^{2-}$. Thus, in addition to $S_2O_3^{2-}$ and SO_3^{2-}, it might be important to consider the distributions of iron and manganese oxides when comparing chemoautotrophic production to the supply of electron acceptors and donors to the interface.

$S_2O_3^{2-}$ and SO_3^{2-} in the Cariaco Basin—$S_2O_3^{2-}$ and SO_3^{2-} in the Cariaco Basin might be formed as a result of the reaction between O_2 in water entrained from the southern

Caribbean Sea with H_2S from waters at intermediate depths in the basin (Scranton et al. 2001; Taylor et al. 2001; Astor et al. 2003). Zhang and Millero (1993*a*) examined sulfur chemistry in the Cariaco Basin, showing that the main products of abiotic H_2S oxidation are $S_2O_3^{2-}$ and SO_3^{2-}.

$$2H_2S + 2O_2 \rightarrow S_2O_3^{2-} + H_2O + 2H^+ \quad (9)$$

$$H_2S + \frac{3}{2}O_2 \rightarrow SO_3^{2-} + 2H^+ \quad (10)$$

Lateral intrusions of O_2-rich water are known to occur in the Cariaco, as demonstrated by the presence in some months of subsurface maxima in O_2 profiles (Astor et al. 2003). Ventilation events observed in 1997 and 1998 were associated with eddies near the shelf in the southeastern Caribbean Sea, and Astor et al. (2003) hypothesized that this would result in colder and denser Caribbean Sea water being drawn up over the sill, subsequently sinking within the Cariaco Basin and spreading along isopycnals. Other intrusions, not associated with eddies, have also been observed (Astor unpubl. data).

The reaction of O_2 and H_2S and subsequent formation of $S_2O_3^{2-}$ and SO_3^{2-} has been found in the chemocline of other anoxic water columns. Zopfi et al. (2001) observed $S_2O_3^{2-}$ and SO_3^{2-} in the anoxic Mariager Fjord, presumably produced as a result of mixing and oxygenation of the water column. Peak concentrations of 5.3 μmol L^{-1} $S_2O_3^{2-}$ and 1.2 μmol L^{-1} SO_3^{2-} were observed in the chemocline, with concentrations decreasing to 2 μmol L^{-1} $S_2O_3^{2-}$ and 0.7 μmol L^{-1} SO_3^{2-} 1 day after an intrusion and to 1.3 μmol L^{-1} for both $S_2O_3^{2-}$ and SO_3^{2-} after 2 days. The decrease in concentrations over time suggests that elevated concentrations of $S_2O_3^{2-}$ and SO_3^{2-} might be transient, reflecting microbial consumption and the length of time since oxygenation of the water column.

A few earlier measurements of $S_2O_3^{2-}$ and SO_3^{2-} are available for the Cariaco Basin. Tuttle and Jannasch (1979) reported maximum values of 14 μmol L^{-1} $S_2O_3^{2-}$ and 2 μmol L^{-1} SO_3^{2-} at the same depth as that at which elevated levels of chemoautotrophic production were found. Zhang and Millero (1993*a*) found that both species increased from <0.5 μmol L^{-1} at the interface to 1 μmol L^{-1} $S_2O_3^{2-}$ and 1.5 μmol L^{-1} SO_3^{2-} at 600 m but did not find distinct maxima. During this study, $S_2O_3^{2-}$ and SO_3^{2-} concentrations were similar to those found by Zhang and Millero (1993*a*) but lower than the maximum values reported by Tuttle and Jannasch (1979). Maximum values in the chemocline during January 2004 were 4.2 μmol L^{-1} for $S_2O_3^{2-}$ and 10.4 μmol L^{-1} for $SO_3{}^2$. May values were 2.1 μmol L^{-1} for $S_2O_3^{2-}$ and 4.0 μmol L^{-1} for SO_3^{2-}.

Integrated $S_2O_3^{2-}$ (between 250 and 450 m) was 459 mmol m^{-2} in January and 287 mmol m^{-2} in May, whereas integrated SO_3^{2-} was 799 mmol m^{-2} in January and 574 mmol m^{-2} in May. The decrease from January to May coincides with a decrease in integrated chemocline chemoautotrophy from 26 to 13 mmol C m^{-2} d^{-1}, and a decrease in the thickness of the suboxic zone from 50 to 10 m. The simultaneous decreases in $S_2O_3^{2-}$ and SO_3^{2-} inventory, chemoautotrophic production, and thickness of the suboxic zone are consistent with the idea that intrusions of O_2-rich water are required to form $S_2O_3^{2-}$ and SO_3^{2-} and that these compounds are then used by chemoautotrophs. No intrusions of water were observed in the oxygen profiles between January and May 2004.

During both January and May, the highest concentrations of $S_2O_3^{2-}$ and SO_3^{2-} occurred near 200 m, where O_2 was ~20 μmol L^{-1} and H_2S was not present. If $S_2O_3^{2-}$ and SO_3^{2-} are formed by the reactions in Eqs. 9 and 10, then the depth of the maxima suggests that $S_2O_3^{2-}$ and SO_3^{2-} must be formed in shallower areas containing H_2S and transported laterally.

$S_2O_3^{2-}$ and SO_3^{2-} can also be formed by reaction with sedimenting Fe or Mn oxides (Millero 1991; Yao and Millero 1993; Zhang and Millero 1993*b*; Eqs. 11–14).

$$6H^+ + 4MnO_2 + 2H_2S \rightarrow 4Mn^{2+} + 5H_2O + S_2O_3^{2-} \quad (11)$$

$$4H^+ + 3MnO_2 + H_2S \rightarrow 3Mn^{2+} + SO_3^{2-} + 3H_2O \quad (12)$$

$$14H^+ + 8FeOOH + 2H_2S \rightarrow 8Fe^{2+} + S_2O_3^{2-} + 13H_2O \quad (13)$$

$$10H^+ + 6FeOOH + H_2S \rightarrow 6Fe^{2+} + SO_3^{2-} + 9H_2O \quad (14)$$

The importance of H_2S oxidation by MnO_2 has been suggested for anoxic portions of the Chesapeake Bay and Black Sea (Luther et al. 1988; Konovalov et al. 2003). In addition to reacting directly with H_2S, Fe and Mn oxides might also provide solid surfaces than can act as catalysts for the oxidation of H_2S. The concentrations of metal oxides present, as well as the presence of O_2, could affect the distribution of sulfur compounds (Zhang and Millero 1993*b*). Unfortunately, to date, we have inadequate data on either Fe or Mn oxides to assess the importance of these processes in $S_2O_3^{2-}$ and SO_3^{2-} formation in the Cariaco Basin.

Estimates of O_2 and H_2S transport to the interface—If $S_2O_3^{2-}$ and SO_3^{2-} are necessary to fuel the chemoautotrophy occurring near the interface in the Cariaco Basin, then enough O_2 needs to enter the basin to form these compounds. If we assume that all chemoautotrophy is supported by $S_2O_3^{2-}$ or SO_3^{2-}, using the May integrated production in the chemocline (13 mmol C m^{-2} d^{-1}), we can estimate the amount of O_2 required to produce these compounds. It is possible that some chemoautotrophic production is associated with aerobic H_2S oxidation, which requires less oxygen to fix 1 mole of CO_2 than does $S_2O_3^{2-}$ or SO_3^{2-}. However Taylor et al. (2001) have already shown that only a small fraction of chemoautotrophy can be supported by vertical eddy diffusive fluxes of H_2S.

With the use of ΔG_f° from Eq. 5 and 191 kJ mol^{-1} for CO_2 fixation, 9 mol of $S_2O_3^{2-}$ should provide enough energy to fix 1 mol of CO_2 through disproportionation. Culture studies have shown that growth yields are low for bacteria grown autotrophically by $S_2O_3^{2-}$ disproportionation (Bak and Cypionka 1987). Assuming 9 mol of $S_2O_3^{2-}$ are required to fix 1 mol of CO_2 and that the entire CO_2 fixation is supported by $S_2O_3^{2-}$ disproportionation, then 117 mmol $S_2O_3^{2-}$ m^{-2} d^{-1} would be required to support the chemoautotrophy occurring in the chemocline. Because all of the chemoautotrophic production in the chemocline probably does not result from the disproportionation of $S_2O_3^{2-}$, this calculation likely will overestimate the amount of $S_2O_3^{2-}$ required. The O_2 concentration outside the basin at sill depth is ~115 μmol L^{-1} (Scranton et al. 2001). If $S_2O_3^{2-}$ is produced according to the reaction in Eq. 9, then 2 mol of O_2 are required to produce 1 mol of $S_2O_3^{2-}$. The area of the entire Cariaco Basin at 320 m is 8.22 × 10^9 m^2 (Scranton et al. 1987), so if chemoautotrophy were uniform throughout the basin, which we suspect is untrue, then 1 × 10^{11} mmol d^{-1} of carbon would be fixed basinwide, requiring 1.7 × 10^{13} L d^{-1} of water to enter the basin.

If water from the Caribbean Sea is entering the basin through the bottom 50 m of the 1.9 × 10^4-m-wide eastern channel at Canal de la Tortuga, then 1.7 × 10^{13} L d^{-1} must enter through 9.5 × 10^5 m^2, giving a velocity of 19 cm s^{-1}. This value is in general agreement with past observations of current velocities in the Cariaco Basin, which usually range from 0 to 20 cm s^{-1}, as measured at a mooring at the Cariaco site (Weisberg pers. comm.). Unfortunately, the detailed geometry of the sill area and the thickness and velocity of the layer of intruding water are not known in adequate detail. Nonetheless, this estimate supports the idea that enough O_2 could enter the basin to produce enough $S_2O_3^{2-}$ to support the chemoautotrophic growth of bacteria.

In addition to O_2, H_2S also needs to be supplied to the chemocline to produce $S_2O_3^{2-}$. The supply of H_2S to the interface can be estimated indirectly from the flux of organic matter because bacteria consume the organic matter directly during biological SO_4^{2-} reduction. At 275 m, there is an average flux of 0.1 g C m^{-2} d^{-1} (0.008 mol C m^{-2} d^{-1}) on the basis of sediment trap data from 2003 (Thunell pers. comm.). Assuming the organic matter starts out with a Redfield ratio of 106 C : 16 N : 1 P, then 53 mol H_2S would be produced from 106 mol C, resulting in a potential H_2S flux of 4.2 mmol H_2S m^{-2} d^{-1}. This is similar to the H_2S flux from the sediments of 3.8 ± 2.4 mmol m^{-2} d^{-1} calculated by Scranton et al. (1987). An H_2S flux of 4.2 mmol m^{-2} d^{-1} would produce 2.1 mmol $S_2O_3^{2-}$ m^{-2} d^{-1}. This is $<2\%$ of the 117 mmol of $S_2O_3^{2-}$ m^{-2} d^{-1} needed to support chemoautotrophic production near the interface. Thus, the particulate sinking flux at the CARIACO site is too low to support the required H_2S flux.

One possible explanation is that the carbon (or H_2S or both) is supplied laterally to the CARIACO site from another location in shallower waters where surface productivity and carbon flux are much higher than predicted from the sediment trap data at the CARIACO site. SeaWiFS satellite production and transport models clearly illustrate that carbon flux to the sediments is higher in other areas of the Cariaco Basin than at the CARIACO site (Muller-Karger et al. 2004) and that H_2S fluxes out of the sediments are likely to be higher as well. During both January and May, the highest concentrations of $S_2O_3^{2-}$ and SO_3^{2-} occurred near 200 m, where no H_2S was present. Horizontal transport of water in the basin could be bringing H_2S, $S_2O_3^{2-}$, and SO_3^{2-} from shallower areas to the CARIACO site.

It has been shown previously that vertical diffusive supplies of H_2S, O_2, and NO_3^- to the chemocline in the Cariaco Basin is insufficient to support the observed chemoautotrophic production (Taylor et al. 2001). Horizontal intrusions of southern Caribbean Sea water could be a mechanism to supply O_2 into the basin. The reaction of H_2S with oxidized species, such as O_2 or metal oxides, could form $S_2O_3^{2-}$ and SO_3^{2-}, which have not been routinely measured and could be used by chemoautotrophs. Estimates of O_2 transport to the interface suggest that enough O_2 could enter the basin to form $S_2O_3^{2-}$ and SO_3^{2-} and support the observed chemoautotrophic production. However, the particulate sinking flux at the CARIACO site is not large enough to support the required H_2S flux. The horizontal transport of water within the basin could bring reduced sulfur species from shallow areas to the CARIACO site, as supported by the measurement of $S_2O_3^{2-}$ and SO_3^{2-} in the oxic zone. Further studies investigating horizontal variability within the basin are needed.

References

AMINOT, A. 1983. Dosage de l'oxygène dissous, p. 75–92. *In* A. Aminot and M. Chaussepied [eds.], Manuel des analyses chimiques en milieu marin. Centre National pour l'Exploitation des Oceans.

ASTOR, Y., F. MULLER-KARGER, AND M. I. SCRANTON. 2003. Seasonal and interannual variation in the hydrography of the Cariaco Basin: Implications for basin ventilation. Cont. Shelf Res. **23:** 125–144.

BAK F., AND H. CYPIONKA. 1987. A novel type of energy metabolism involving fermentation of inorganic sulphur compounds. Nature **326:** 891–892.

———, AND N. PFENNIG. 1987. Chemolithotrophic growth of *Desulfovibrio sulfodismutans* sp. nov. by disproportionation of inorganic sulfur compounds. Arch. Microbiol. **147:** 184–189.

CANFIELD, D. E., B. THAMDRUP, AND S. FLEISCHER. 1998. Isotope fractionation and sulfur metabolism by pure and enrichment cultures of elemental-sulfur disproportionating bacteria. Limnol. Oceanogr. **43:** 253–264.

CLINE, J. D. 1969. Spectrophotometric determination of hydrogen sulfide in natural waters. Limnol. Oceanogr. **14:** 454–458.

DICHRISTINA, D. J., R. G. ARNOLD, M. E. LIDSTROM, AND M. R. HOFFMANN. 1988. Dissimilative iron reduction by the marine eubacterium *Alteromonas-putrefaciens* strain-200. Water Sci. Technol. **20:** 69–79.

HO, T., Y. ASTOR, R. VARELA, G. T. TAYLOR, AND M. I. SCRANTON. 2004. Vertical and temporal variability of redox zonation in the water column of the Cariaco Basin: Implications for organic oxidation pathways. Mar. Chem. **86:** 89–104.

JANNASCH, H. W., AND M. J. MOTTL. 1985. Geomicrobiology of deep-sea hydrothermal vents. Science **4715:** 717–725.

JANSSEN, P. H., A. SCHUHMANN, F. BAK, AND W. LIESACK. 1996. Disproportionation of inorganic sulfur compounds by the sulfate-reducing bacterium *Desulfocapsa thiozymogenes* gen. nov., sp. nov. Arch. Microbiol. **166:** 184–192.

JØRGENSEN, B. B., AND F. BAK. 1991. Pathways and microbiology of thiosulfate transformations and sulfate reduction in marine sediment (Kattegat, Denmark). Appl. Environ. Microbiol. **57:** 847–856.

———, H. FOSSING, C. O. WIRSEN, AND H. W. JANNASCH. 1991. Sulfide oxidation in the anoxic Black Sea chemocline. Deep-Sea Res. **38:** S1083–S1103.

KARL, D. M. 1978. Distribution, abundance, and metabolic states of microorganisms in the water column and sediments of the Black Sea. Limnol. Oceanogr. **23:** 936–949.

KONOVALOV, S. K., AND OTHERS. 2003. Lateral injection of oxygen with the Bosporus plume—fingers of oxidizing potential in the Black Sea. Limnol. Oceanogr. **43:** 2369–2376.

KRAMER, M., AND H. CYPIONKA. 1989. Sulfate formation via ATP sulfurylase in thiosulfate-disproportionating and sulfite disproportionating bacteria. Arch. Microbiol. **151:** 232–237.

LOVLEY, D. R., AND E. J. P. PHILLIPS. 1988. Novel mode of microbial energy-metabolism—organic carbon oxidation coupled to dissimilatory reduction of iron or manganese. Appl. Environ. Microbiol. **54:** 1472–1480.

LUTHER, G. W. III, T. FERDELMAN, AND E. TSAMAKIS. 1988. Evidence suggesting anaerobic oxidation of the bisulfide ion in Chesapeake Bay. Estuaries **11:** 281–285.

MADIGAN, M. T., J. MARTINKO, AND J. PARKER. 2003. Brock biology of microorganisms. Prentice Hall.

MADRID, V. M. 2000. Characterization of the bacterial communities in the anoxic zone of the Cariaco Basin, M.S. thesis, State Univ. of New York, Stony Brook.

MILLERO, F. J. 1991. The oxidation of H_2S in Framvaren Fjord. Limnol. Oceanogr. **36:** 1007–1014.

MULLER-KARGER, F., R. VALERA, R. THUNELL, Y. ASTOR, H. ZHANG, R. LUERSSEN, AND C. HU. 2004. Processes of coastal upwelling and carbon flux in the Cariaco Basin. Deep-Sea Res. II **51:** 927–943.

———, AND OTHERS. 2001. Annual cycle of primary production in the Cariaco Basin: Response to upwelling and implications for vertical export. J. Geophys. Res. **106:** 4527–4542.

MYERS, C. R., AND K. H. NEALSON. 1988. Microbial reduction of manganese oxides—interaction of iron and sulfur. Geochim. Cosmochim. Acta **52:** 2727–2732.

NEALSON, K. H., AND C. R. MYERS. 1992. Microbial reduction of manganese and iron: New approaches to carbon cycling. Appl. Environ. Microbiol. **58:** 439–443.

SCRANTON, M. I., Y. ASTOR, R. BOHRER, T. Y. HO, AND F. MULLER-KARGER. 2001. Controls on the temporal variability of the geochemistry of the deep Cariaco Basin. Deep-Sea Res. I **48:** 1605–1625.

———, F. L. SAYLES, M. P. BACON, AND P. G. BREWER. 1987. Temporal changes in the hydrography and chemistry of the Cariaco Trench. Deep-Sea Res. **34:** 945–963.

SØRENSEN, K. B., AND D. E. CANFIELD. 2004. Annual fluctuations in sulfur isotope fractionation in the water column of a euxinic marine basin. Geochim. Cosmochim. Acta **68:** 503–515.

SOROKIN, Y. I., P. Y. SOROKIN, V. A. AVDEEV, D. Y. SOROKIN, AND S. V. ILCHENKO. 1995. Biomass, production, and activity of bacteria in the Black Sea, with special reference to chemosynthesis and the sulfur cycle. Hydrobiologia **308:** 61–76.

TAYLOR, G. T., M. IABICHELLA, T. HO, M. I. SCRANTON, F. MULLER-KARGER, AND R. VARELA. 2001. Chemoautotrophy in the redox transition zone of the Cariaco Basin: A significant source of organic carbon production. Limnol. Oceanogr. **46:** 148–163.

THAUER, R. K., K. JUNGERMANN, AND K. DECKER. 1977. Energy conservation in chemotrophic anaerobic bacteria. Bacteriol. Rev. **41:** 100–180.

TUTTLE, J. H., AND H. W. JANNASCH. 1973. Sulfide and thiosulfate-oxidizing bacteria in anoxic marine basins. Mar. Biol. **20:** 64–70.

———, AND ———. 1979. Microbial dark assimilation of CO_2 in the Cariaco Trench. Limnol. Oceanogr. **24:** 746–753.

VAIRAVAMURTHY, A., AND K. MOPPER. 1990. Determination of sulfite and thiosulfate in aqueous samples including anoxic seawater by liquid chromatography after derivatization with 2,2′-dithiobis(5-nitropyridine). Environ. Sci. Technol. **24:** 333–337.

VETRIANI, C., H. V. TRAN, AND L. J. KERKHOF. 2003. Fingerprinting microbial assemblages from the oxic/anoxic chemocline of the Black Sea. Appl. Environ. Microbiol. **69:** 6481–6488.

YAO, W. S., AND F. J. MILLERO. 1993. The rate of sulfide oxidation by δMnO_2 in seawater. Geochim. Cosmochim. Acta **57:** 3359–3365.

ZHANG, J., AND F. J. MILLERO. 1993*a*. The chemistry of the anoxic waters of the Cariaco Trench. Deep-Sea Res. I **40:** 1023–1041.

——— AND ———. 1993*b*. The products of the oxidation of H_2S in seawater. Geochim. Cosmochim. Acta **57:** 1705–1718.

ZOPFI, J., T. G. FERDELMAN, B. B. JØRGENSEN, A. TESKE, AND B. THAMDRUP. 2001. Influence of water column dynamics on sulfide oxidation and other major biogeochemical processes in the chemocline of Mariager Fjord (Denmark). Mar. Chem. **74:** 29–51.

Received: 7 March 2005
Accepted: 30 August 2005
Amended: 16 September 2005