SULFATE REDUCTION IN GROUND WATER OF SOUTHEASTERN MONTANA

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UNITED STATES DEPARTMENT OF THE INTERIOR

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CONTENTS

ILLUSTRATIONS

TABLES

METRIC CONVERSION TABLE

The following factors may be used to convert inch-pound units to the International System (SI) of metric units.

Multiply inch-pound unit	By	To obtain SI unit
acre	4047	square meter (m^2)
foot (ft)	0.3048	meter (m)

SULFATE REDUCTION IN GROUND WATER OF SOUTHEASTERN MONTANA

By

William S. Dockins[1], Gregory J. Olson[1], Gordon A. McFeters[1], Susan C. Turbak[1], and Roger W. Lee[2]

ABSTRACT

Ground water in southeastern Montana was investigated to determine if sulfide production was bacterially mediated. Sulfate-reducing bacteria were detected in 25 of 26 ground-water samples in numbers ranging from 2.0×10^1 to greater than 2.4×10^4 bacteria per 100 milliliters. Stable sulfur isotope fractionation studies indicate a biological role in sulfate reduction. However, sulfate-reducing activity as determined by use of a radioactive sulfur isotope was observed in only 1 of 16 samples. Bacterial dissimilatory sulfate reduction is postulated to be responsible for a major part of the sulfide produced in these ground waters. These bacteria are most likely active in the adsorbed state, possibly in subsurface microzones where environmental conditions are conducive to sulfate reduction.

INTRODUCTION

Much of the surficial geology of southeastern Montana consists of the Tertiary Fort Union Formation, a continental deposit of sandstone, siltstone, and shale, which contains low sulfur subbituminous and lignite coal deposits of economic interest. Coal seams 25 to 60 ft (8-20 m) thick are not uncommon in this area and several aquifers consist of coal-bearing strata.

Hydrogen sulfide (H_2S or HS^-) is present in ground water in many areas including those associated with coal and sulfur deposits. Most studies have been concerned with removal of this toxic gas from potable and coal-mine waters. Because the odor threshhold of H_2S is low, this compound can be objectionable in ground water in nontoxic concentrations. Sulfide is dissolved in ground waters throughout southeastern Montana at concentrations that may reach 3 to 4 mg/L (milligrams per liter), but generally are less than 1.0 mg/L.

The major source of the sulfide ion in ground water is thought to be reduction of the sulfate ion (Riffenburg, 1925), but often, as in the case of southeastern Montana ground waters, the presence of sulfide cannot be explained by the slow kinetics of abiotic sulfate reduction. In many studies, various biochemical reactions, which could overcome chemical kinetic restrictions, have been postulated as promoters of sulfate reduction, usually with little experimental evidence to support these contentions.

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Sulfate-reducing bacteria (Desulfovibrio desulfuricans) are widely distributed in nature and they have been extensively studied from a physiological standpoint. These micro-organisms live anaerobically by oxidizing certain simple organic compounds and produce sulfide from reduction of sulfate ion:

$$2CH_2O + SO_4^{=} = H_2O + CO_2 + HCO_3^{-} + HS^{-}. \quad (1)$$

A strong case for their presence and activity in ground water has been constructed by several authors, as reviewed by McNabb and Dunlap (1975). Li (1975) reported that the major source of hydrogen sulfide in ground water was the anaerobic reduction of sulfate. He quantified the population of sulfate-reducing bacteria in aquifer sediments. Al-Sawaf (1977) studied the distribution of sulfate-reducing bacteria in ground water and their importance in the formation of minable sulfur deposits in northern Iraq. A series of experiments to explain the origin of hydrogen sulfide in ground waters of the Carpathian region of the USSR, an area of extensive sulfur deposition, was undertaken by Ivanov (1964). A survey of sulfate-reducing bacteria in both ground water and host rock, measurements of the rate of sulfate reduction in ground water, and a consideration of hydrogen sulfide evolution from purely chemical processes supported his argument that the hydrogen sulfide present in these ground waters was due to bacterial sulfate reduction.

The degree of isotope fractionation of various light elements such as sulfur can be used to examine the likelihood of biological transformation in natural systems. The two most common stable isotopes of sulfur in nature are ^{32}S and ^{34}S. Thode, Kleerekoper, and McElcheran (1951) and Harrison and Thode (1958) demonstrated that biological sulfate reduction by the bacterium Desulfovibrio desulfuricans enhanced the preferential selection of the lighter stable isotope of sulfur (^{32}S) during the sulfate-reduction process. The magnitude of sulfur-isotope fractionation in natural environments often surpasses the abiotic limits of fractionation and has only been adequately explained by assuming the presence of a biological process. The fractionation of sulfur isotopes has been quantified in several natural systems including salt dome sulfur deposits, marine waters and sediments, sulfur springs, and lakes. Thode, MacNamara, and Collins (1949) determined the isotope abundance in some sulfide-containing ground waters and found the sulfur isotopes to be fractionated, but in this early work they were not concerned with biological reactions.

Purpose and scope

This investigation was designed to determine if sulfide production in southeastern Montana ground water was bacterially mediated. The problem was approached using three techniques: (1) the detection and quantification of sulfate-reducing bacteria, (2) demonstration of sulfate-reducing activity in ground water using a radioactive sulfur isotope, and (3) sulfur isotope fractionation studies.

Description of study site

All wells sampled were located in southeastern Montana in areas lying within the northern Powder River Basin. The wells were chosen to allow sampling of several different aquifers in various source materials. Figure 1 is a map of this area with the sampling sites marked. Table 1 lists the locations of the sites.

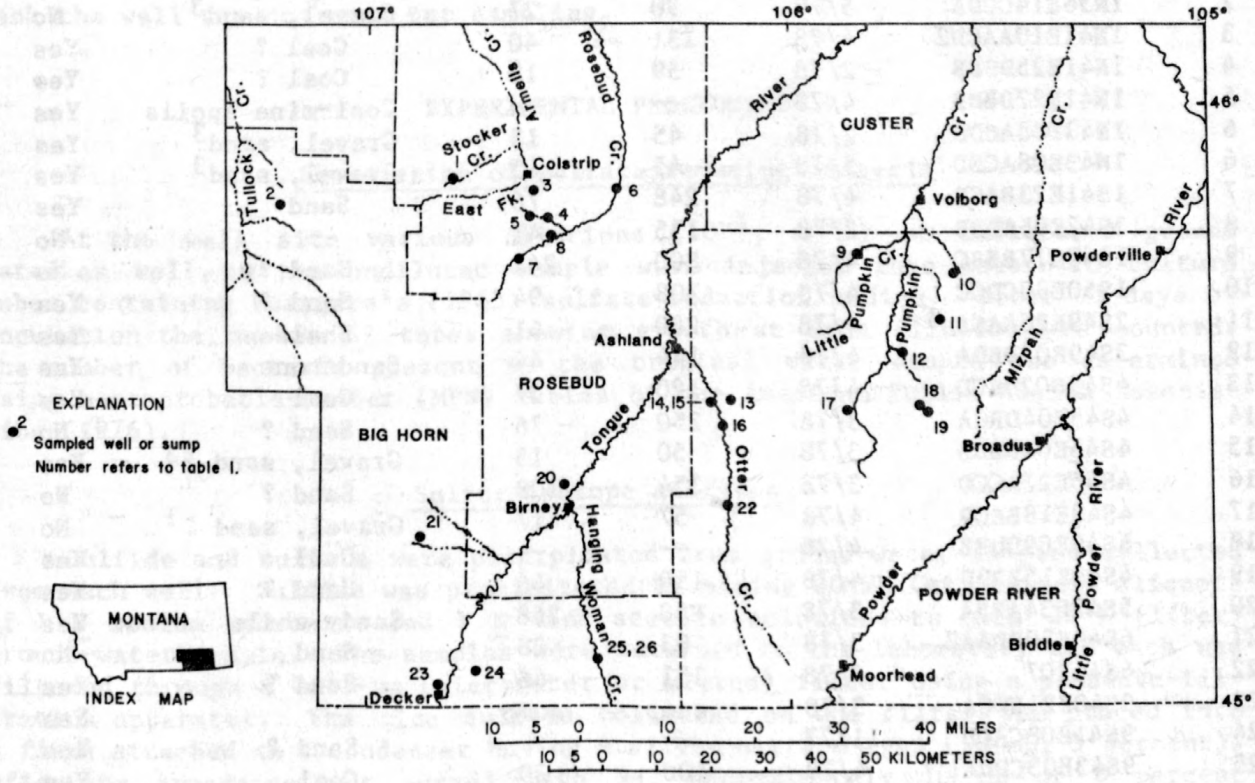

Figure 1.--Locations of sampling sites.

Table 1.--Description of sampled wells and sump

Site number (fig. 1)	Location[1]	Date sampled	Depth (feet)	Depth (meters)	Aquifer material[2]	Odor H_2S
1	2N40E31DCCD	2/78	165	50	Shale	No
2	1N36E14CCDA	5/78	90	27	Gravel, sand[3]	No
3	1N41E10AACD2	4/78	131	40	Coal ?	Yes
4	1N41E25DDBB	2/78	59	18	Coal ?	Yes
5	1N41E27DBDB	4/78	--	--	Coal-mine spoils	Yes
6	1N43E08ACDD	2/78	45	15	Gravel, sand[3]	Yes
6	1N43E08ACDD	5/78	45	15	Gravel, sand[3]	Yes
7	1S41E23BACB	4/78	248	76	Sand	Yes
8	1S42E05ADBB	2/78	135	41	--	No
9	1S48E17BBBC	4/78	800	244	Sand ?	No
10	1S50E30CDCC	4/78	308	94	Sand ?	Yes
11	2S49E26AACA	4/78	200	61	Shale	Yes
12	3S49E07DBDA	4/78	144	44	Sandstone	Yes
13	4S45E02DACD	4/78	120	37	Coal	Yes
14	4S45E04DBCA	3/78	250	76	Sand ?	No
15	4S45E04DBDB	3/78	50	15	Gravel, sand ?[3]	Yes
16	4S45E27ACCD	3/78	354	108	Sand ?	No
17	4S48E18BACD	4/78	57	17	Gravel, sand ?[3]	No
18	4S49E09DDBB	4/78	--	--	Coal	Yes
19	4S49E15BDDD	4/78	150	46	Sand ?	Yes
20	5S42E34ABBA	3/78	880	268	Sandy shale	Yes
21	6S40E30DDAA2	3/78	93	28	Sand	No
22	6S46E07	3/78	151	46	Sand ?	Yes
23	9S40E21BDCA	3/78	sump	sump	--	Yes
24	9S41E08CACD	11/77	--	--	Sand ?	Yes
25	9S43E03CDDA1	4/78	100	30	Coal	Yes
26	9S43E03CDDA2	4/78	60	18	Coal	Yes

[1]Location number based on Federal system of land subdivision. The first number indicates the township; the second, the range; and the third, the section. The first number following the section denotes the 160-acre tract; the second, the 40-acre tract; the third, the 10-acre tract; and the fourth, the 2-1/2 acre tract. Letters are assigned in a counterclockwise direction, beginning with "A" in the northeast quadrant.

[2]Lithology followed by query was determined indirectly; other lithology was determined from well logs.

[3]Gravel and sand sources are most likely river alluvium.

General sampling techniques

All efforts were made to ensure that the ground-water sample removed from each well was representative of the formational water in that area. Wells that were not flowing or in continuous operation were pumped for 0.5 to 3.0 hours, depending upon depth and clarity of the sample, before sampling. In some cases, conductivity was measured at intervals during pumping and the sample was collected when the conductivity stabilized. Sampling of wells having pressure tanks was avoided where possible, and if water supplies had a storage tank the well was not used for sampling.

EXPERIMENTAL PROCEDURES

Enumeration of sulfate-reducing bacteria

At the well site various dilutions (10^{-1}, 10^{-2}) of unfiltered ground water as well as the undiluted sample were injected into anaerobic culture tubes containing Postgate's (1963) sulfate-reduction medium. After 21 days of incubation the number of tubes showing growth at each dilution was counted. The number of bacteria present in the original water sample was determined using most-probable-number (MPN) tables of the American Public Health Association (1976).

Sulfur isotope analyses

Sulfide and sulfate were precipitated from ground-water samples collected from each well. Sulfide was precipitated by adding 20 mL (milliliter) aliquots of 1 N sodium hydroxide and 1 N zinc acetate solutions to each 20 L (liter) ground-water sample. The samples were returned to the laboratory and each was filtered through a 0.45-µm (micrometer or micron) filter using a pressure-filtration apparatus. The zinc sulfide collected on the filter was placed into a flask attached to a condenser having a silver nitrate trap (10 mL, 5 percent). After the apparatus was purged with N_2, approximately 10 mL of 8 percent (weight to volume) $SnCl_2 \cdot 2H_2O$ in 6 N HCl was added to the zinc sulfide, and the mixture was heated to boiling and stirred for about 30 minutes while the gassing continued. The trap was disconnected and released sulfide trapped as silver sulfide was allowed to settle, washed several times in distilled water, placed in serum vials, and air dried.

Sulfate was precipitated from the original filtrate by adding barium chloride (3 g) to 1 L of sample, which had been acidified to pH 3 and heated to boiling. Barium sulfate was collected on a 0.45-µm filter, placed in a serum vial, and dried for 15 minutes at 105°C.

Isotope fractionation analyses of these samples were performed by Global Geochemistry Corp., Santa Monica, Calif. Data are expressed as $\delta^{34}S$, as compared to standard troilite (FeS). The data, which are reported per mil (parts per thousand), are calculated by the formula:

$$\delta^{34}S = \frac{(^{34}S/^{32}S)_{sample} - (^{34}S/^{32}S)_{standard} \times 1000}{(^{34}S/^{32}S)_{standard}} \quad (2)$$

Sulfate-reduction rates

Radioactive sulfate ($^{35}SO_4$) was added to replicate tubes containing unfiltered ground-water samples from each of the sites. Some of the tubes also received spikes of lactate (a source of energy for sulfate-reducing bacteria) and sulfide (a reducing agent). After a 7-to 12-day period of incubation, the tubes were assayed for radioactive hydrogen sulfide production using a scintillation counter. The daily rate of hydrogen sulfide production was calculated by the formula of Ivanov (1964):

$$\text{Rate (mg } H_2S \text{ generated per liter per day)} = \frac{r \times (SO_4) \times 24 \times 1.06}{R \times t}. \qquad (3)$$

In this formula, R is the radioactivity of the added sulfate in counts per minute, r is the radioactivity of the hydrogen sulfide produced by the bacteria in counts per minute, (SO_4) is the sulfate concentration of the sample in milligrams per liter, t is the duration of the experiment in hours, and 1.06 is a correction factor for converting sulfide sulfur to hydrogen sulfide.

RESULTS

Detection of sulfate-reducing bacteria

Sulfate-reducing bacteria were detected in 25 of 26 ground-water samples. Numbers of these organisms ranged from 2.0×10^1 bacteria per 100 milliliters to a number exceeding the limit of maximum detection that was employed (2.4×10^4 bacteria per 100 milliliters, table 2). No correlation was observed between the depth or geographic location of the wells sampled and the number of sulfate-reducing bacteria. These organisms were present in several samples that did not have a hydrogen sulfide odor.

Sulfur isotope fractionation

Generally the stable sulfur isotopes recovered from the ground-water samples were fractionated as compared to the standard meteoritic troilite (table 2). Values for $\delta^{34}S$ ranged from -38.50 to +6.68 per mil for the sulfides and -0.20 to +45.17 per mil for the sulfates, and averaged -23.80 and +7.96 per mil, respectively (fig. 2). In each individual water sample differential fractionation of the ^{34}S in the sulfate and sulfide species was measured, and a preference for the lighter isotope (^{32}S) by the bacteria during the sulfate-reduction process was indicated. The magnitude of the difference between the sulfate and sulfide values from each sample pair ranged from 9.54 to 48.13 per mil and averaged 32.31 per mil. In figure 3 the $\delta^{34}S$ values for sulfates and sulfides from each sample are plotted against each other. Single samples of both pyrite and gypsum were collected in the study area and the sulfur isotope fractionation values were -24.42 and -9.15 per mil, respectively.

Table 2.--Chemical and microbiological data from sampled wells and sump

Site number (fig. 1)	Location	Sulfate-reducing bacteria (MPN/100mL)	$\delta^{34}S$ (parts per thousand)		Fractionation difference ($\delta^{34}S_{SO_4^=} - \delta^{34}S_{S^=}$)	$SO_4^=$ (mg/L)
			Ag_2S	$BaSO_4$		
1	2N40E31DCCD	2.0×10^1	--	--	--	700
2	1N36E14CCDA	1.6×10^4	--	--	--	640
3	1N41E10AACD2	3.5×10^3	--	--	--	638
4	1N41E25DDBB	1.4×10^2	-36.82	+3.08	39.90	1,100
5	1N41E27DBDB	2.2×10^3	-8.43	+2.41	10.84	1,764
6	1N43E08ACDD	2.4×10^3	--	+3.33	--	1,075
6	1N43E08ACDD	3.5×10^3	-6.15	+3.39	9.54	--
7	1S41E23BACB	2.8×10^3	-35.11	+1.85	36.96	570
8	1S42E05ADBB	1.3×10^3	--	--	--	1,700
9	1S48E17BBBC	1.4×10^2	--	--	--	--
10	1S50E30CDCC	2.4×10^4	-36.43	+4.11	40.54	1,600
11	2S49E26AACA	3.5×10^3	-27.68	+3.33	31.01	860
12	3S49E07DBDA	3.5×10^3	--	--	--	--
13	4S45E02DACD	2.4×10^4	--	--	--	1,000
14	4S45E04DBCA	0	--	--	--	3.8
15	4S45E04DBDB	5.4×10^3	-3.43	+35.82	39.25	48
16	4S45E27ACCD	7.9×10^2	--	--	--	6.0
17	4S48E18BACD	1.4×10^3	--	--	--	830
18	4S49E09DDBB	2.4×10^4	-34.65	+2.68	37.33	--
19	4S49E15BDDD	2.4×10^4	-38.50	+9.63	48.13	1,100
20	5S42E34ABBA	2.4×10^4	-11.43	+6.07	17.50	79
21	6S40E30DDAA2	1.1×10^3	--	--	--	370
22	6S46E07	7.0×10^2	--	--	--	--
23	9S40E21BDCA	3.5×10^3	-32.90	-0.20	32.70	350
24	9S41E08CACD	1.7×10^3	+6.68	+45.17	38.49	7
25	9S43E03CDDA1	2.4×10^4	-31.47	+0.79	32.26	130
26	9S43E03CDDA2	5.4×10^3	-36.94	+0.90	37.84	3,900

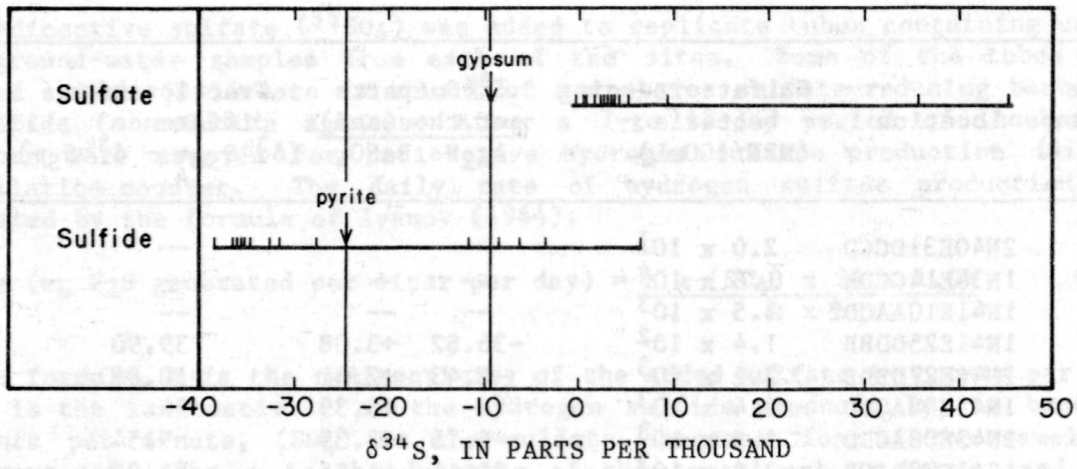

Figure 2.--Range and distribution of $\delta^{34}S$ values in sulfates and sulfides relative to $\delta^{34}S$ values for possible sources of sulfate in selected ground-water samples. The possible sources of sulfate were pyrite and gypsum collected in the study area.

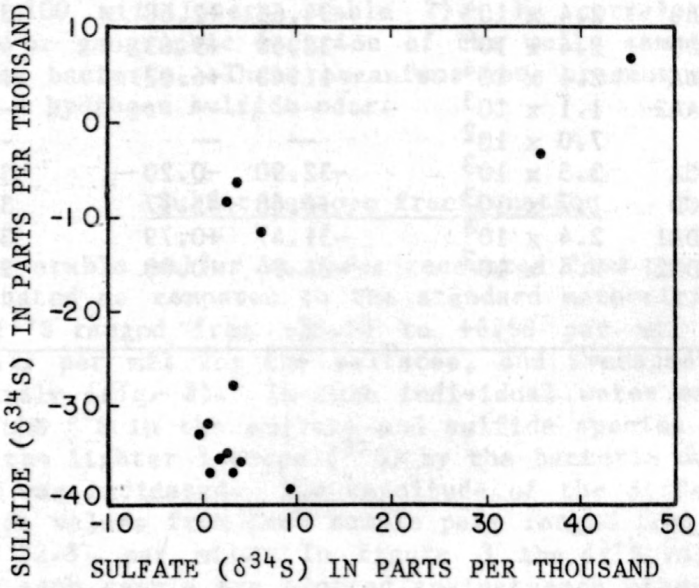

Figure 3.--Relationship of $\delta^{34}S$ between sulfate and sulfide ions for selected ground-water samples.

Sulfate-reduction rates

Radioactively labeled sulfate was reduced in only 1 of the 16 samples tested. This ground-water sample was unusual in that it contained a large quantity of coal particulate. The rate of sulfate reduction [(mg/L)/day H_2S] in this sample was 0.0036 in the untreated sample and 0.0053 when lactate was added. In the presence of added sulfide the rate increased to 0.047 and with lactate and sulfide it was 0.034. No measurable sulfate reduction was observed in the control tube.

DISCUSSION

Sulfate-reducing bacteria may cause the characteristics and quality of natural waters to be altered. This effect may be beneficial or detrimental depending upon the initial quality and projected use of the water in question. For example, the presence of hydrogen sulfide may decrease the potability of a water supply and may be responsible for pipe corrosion. In other instances this compound may serve to precipitate toxic heavy metals from mine effluents (Tuttle and others, 1969; Ilyaletdinov and others, 1977). Al-Sawaf (1977) suggested that cultivation of sulfate-reducing bacteria in aquifers in the Lower Fars Formation in Iraq could rid the ground water of excessive sulfates that are making the water unfit for agricultural or human use.

Sulfate-reducing bacteria were recovered in relatively high numbers from all but one of the wells sampled during this study. Repeated detection of a physiological group of organisms generally indicates that these organisms are native to and show activity in that habitat. The numbers of organisms obtained compared favorably with numbers reported in ground waters associated with sulfur deposits in the Carpathian region of the USSR by Ivanov (1964) and in ground waters in Iraq by Al-Sawaf (1977). It is interesting to note that these organism counts were comparable in spite of the fact that the dissolved-sulfide concentrations were much lower in the ground waters sampled in this study.

The number of sulfate-reducing bacteria in the only sample to show activity as previously discussed was comparable to the values obtained from the measurements of the ground-water samples. However, because greater numbers of bacteria could be attached to particulate matter, the count may not be representative of the actual population. An increase of sulfate-reducing activity in this sample was noted when sodium sulfide (a reducing agent) was added, suggesting that either oxygenation of the sample had occurred during experimental manipulation or the natural water environment did not contain a redox potential favorable to sulfate-reducing bacteria.

Ivanov (1964) was able to demonstrate sulfate reduction using a radioactive isotope in many ground-water samples. He found the rates to be limited by the concentration of organic compounds capable of being utilized by sulfate-reducing bacteria, but the samples in that study contained much more dissolved sulfide. One reason for the inability to demonstrate appreciable rates of sulfate reduc-

tion in the majority of the samples may be because sulfate-reducing bacteria are more active in the attached state, possibly in subsurface microzones where environmental conditions (for example, nutrients, redox potential) conducive to sulfate reduction occur. If this is true, the bacteria recovered from the ground water may represent only a small part of the total sulfate-reducing population that either is suspended in an unfavorable environment that may cause physiological damage to the cells or is too few in number for detection of sulfate-reduction rate. Evidence exists to support the contention that sulfate-reducing bacteria may be more active in the attached state. Li (1975) reported numbers of sulfate-reducing bacteria in aquifer sediment that were greater than those usually obtained in ground water. Ivanov (1961) found that the number of sulfate-reducing bacteria in sedimentary rocks associated with sulfur deposits was in most instances one or two orders of magnitude higher than in the ground water in these regions. Bacteria have been found to adhere to favorable surfaces in nearly every competitive natural environment examined, and aquifer material should be no exception to these observations.

The sulfur isotopes in both the sulfate and sulfide forms isolated from the ground water during this study were generally fractionated when compared to standard meteoritic troilite. More important is that the sulfate and sulfide pairs recovered from each individual sample showed substantial differences in fractionation. The magnitude of this difference was comparable to and in many instances greater than that found in previously reported studies where the authors attributed the formation of sulfide to biological transformations (Thode and others, 1951; Kaplan, 1975).

The sulfate in these ground waters originates from several sources including pyrite oxidation and the solubilization and leaching of gypsum. The observation that all sulfate samples were more enriched in ^{34}S than either of these minerals is consistent with the hypothesis that these are likely source materials. Olson, Turbak, and McFeters (1979) have found that aquifers in coal and associated overburden strata exposed by strip-mining operations in southeastern Montana harbor appreciable numbers of bacteria of the genus Thiobacillus that are capable of oxidizing reduced sulfur compounds. The sulfur isotopes from secondary gypsum and secondary pyrite collected in the area were fractionated enough to lead the authors to suspect that sulfate-reducing bacteria may have been active at the time of formation and deposition of these minerals. Much of the secondary gypsum present in porous aquifer materials is likely the result of pyrite oxidation, and the sulfur isotope fractionation data obtained for these two minerals tend to support this hypothesis. The grouping of data points when the fractionation values for the sulfates and sulfides from each sample are plotted (fig. 3) may reflect differences in the sulfate source and the extent of sulfate-reducing activity within a localized system. Two ground-water samples contained sulfates that were considerably enriched in ^{34}S. These samples also had low sulfate concentrations (table 2). The data imply that in these ground waters low sulfate concentrations are likely due to depletion by bacterial sulfate reduction.

The abiotic production of sulfide in these waters appears unlikely. Although the reaction is thermodynamically feasible, the extreme reaction conditions or the reaction rate necessary for abiotic sulfate reduction prob-

ably does not exist in this system. Therefore, the high fractionation values and the ubiquitous presence of sulfate-reducing bacteria in this system indicate that these organisms are responsible for a major part of the sulfide present in southeastern Montana ground waters.

A knowledge of sulfur transformation in ground water is important in the understanding of basic biological and geochemical phenomena in southeastern Montana. Coal mining operations which may alter ground-water quality and flow are expanding rapidly in this area and applications of knowledge of microbiological aspects of the sulfur cycle could alleviate some of the associated water-pollution problems. For example, the rapid establishment of a sulfate-reducing bacterial population in backfilled coal overburden could decrease the problem of sulfate loading of the ground waters by leaching processes. The activity of sulfate-reducing bacteria could perhaps be enhanced by adjusting conditions in potential problem areas to provide a favorable environment. The factors limiting growth of these bacteria in aquifers examined in this study are not known. Ivanov (1964) found that in some ground waters the supply of simple organic compounds utilized by sulfate-reducing bacteria was limiting. These compounds were supplied only during aquifer recharge resulting in seasonal variability of sulfate-reduction rates. There has been some speculation that sulfate-reducing bacteria may use complex organic compounds as sole carbon sources, or alternatively that a consortium of different types of bacteria may break down hydrocarbons into forms usable by the sulfate reducers (Zobell, 1958).

CONCLUSIONS

Sulfate-reducing bacteria (Desulfovibrio desulfuricans) exist in relatively high numbers in ground water sampled in southeastern Montana. These bacteria promote sulfate to sulfide reduction in ground water with a preference for the lighter ^{32}S isotope over the ^{34}S isotope. Thus, ^{34}S is enriched in dissolved sulfate and ^{32}S in sulfide beyond their natural abundances. This disequilibrium ($\delta^{34}S$) occurs in all types of shallow sediments throughout southeastern Montana.

Bacteria adsorbed on the sediment may have higher activity than in the unadsorbed state. An apparent H_2S production was positive during only one test; the rate was 0.0036 (mg/L)/day. Ground water having low sulfate concentration shows high $\delta^{34}S$ values for the sulfate phase. Fractionation of ^{34}S is apparent in aquifer minerals. Pyrite (FeS_2) shows low concentrations of ^{34}S, whereas gypsum ($CaSO_4 \cdot 2H_2O$) shows higher values, which are still low by comparison to data from previous studies. The data indicate possible pyrite oxidation as the source of the sulfate in the gypsum.

REFERENCES

Al-Sawaf, F. D. S., 1977, Sulfate reduction and sulfur deposition in the Lower Fars Formation, Northern Iraq: Economic Geology, v. 72, p. 608-618.

American Public Health Association, 1976, Standard methods for the examination of water and wastewater [14th ed.]: Washington, D.C., American Public Health Association, Inc., 1193 p.

Harrison, A. G., and Thode, H. G., 1958, Mechanism of the bacterial reduction of sulfate from isotope fractionation studies: Transactions of Faraday Society, v. 54, p. 84-92.

Ilyaletdinov, A. N., Enker, P. B., and Loginova, L. V., 1977, Role of sulfate reducing bacteria in the precipitation of copper: Microbiology, v. 46, p. 92-95.

Ivanov, M. V., 1961, Microbiological studies of Carpathian sulfur deposits, Part V, Occurrence of sulfate reducing bacteria in sedimentary rocks composing sulfur deposits: Microbiology, v. 30, p. 428-430.

_____1964, Microbial processes in the formation of sulfur deposits: Publications of Israel Program for Scientific Translation for U.S. Department of Agriculture and National Science Foundation (translated from Russian in 1968), 298 p.

Kaplan, I. R., 1975, Stable isotopes as a guide to biogeochemical processes: Proceedings Royal Society of London B, v. 189, p. 183-211.

Li, E. C. C., 1975, Significance of hydrogen sulfide in groundwater: Water and Sewage Works, v. 122, p. 66-67.

McNabb, J. F., and Dunlap, W. J., 1975, Subsurface biological activity in relation to groundwater pollution: Ground Water, v. 13, p. 33-34.

Olson, G. J., Turbak, S. C., and McFeters, G. A., 1979, Impact of western coal mining, Part II, Microbiological studies: Water Research, no. 13, p. 1033-1041.

Postgate, J. R., 1963, Versatile medium for the enumeration of sulfate reducing bacteria: Applied Microbiology, v. 11, p. 265-267.

Riffenburg, H. B., 1925, Chemical character of ground-waters of the Northern Great Plains: U.S. Geological Survey Water-Supply Paper 560-B, p. 31-52.

Thode, H. G., Kleerekoper, H., and McElcheran, D., 1951, Isotopic fractionation in the bacterial reduction of sulphate: London, Research, v. 4, p. 581-582.

Thode, H. G., MacNamara, J., and Collins, C. B., 1949, Natural variations in the isotopic content of sulphur and their significance: Canadian Journal of Research, v. B27, p. 361-373.

Tuttle, J. H., Dugan, P. R., and Randles, C. I., 1969, Microbial sulfate reduction and its potential utility as an acid mine water pollution abatement procedure: Applied Microbiology, v. 17, p. 297-302.

Zobell, L. E., 1958, Ecology of sulfate reducing bacteria: Producers Monthly, v. 22, p. 12-29.