

about 6400 years ago. This is at least 1500 years after major changes in lake level or vegetation occurred in or near the study area.

The grain size profile of 23P is too noisy to compare with 24P (Fig. 1), perhaps because of local current eddies in the vicinity of site 23P. The profile was smoothed by plotting a running three-point average of median grain size—that is, the median grain size at each depth was changed to the average of the medians of the sample above, the one at the depth, and the one below. This profile compares roughly with the unaltered profile in core 24P (Fig. 2). No strong episodic signal emerges of grain size versus depth other than downcore coarsening. There appears to be a slight correlation of two relatively fine-grained intervals (A and B in Fig. 2). Assignment of absolute ages to the intervals on the basis of their magnetic profiles is not possible because the upper parts cannot be correlated with confidence to other dated profiles (15, 16). Linear interpolation between core top and the postglacial-glacial contact date events A and B at about 500 and 3500 years ago, respectively.

In summary, the five cores from Lake Superior that show a downcore increase in the median grain size indicate that bottom-water circulation in Lake Superior was stronger between 9500 and 6500 years ago than afterward. Because the bottom currents are driven primarily by large storms, the data on grain size suggest more frequent or more intense storms in the early Holocene as the Laurentide ice sheet retreated than after its disappearance at about 6500 years ago. Grain size and paleomagnetic data for the late Holocene provide weak evidence for periods of decreased storm activity about 500 and 3500 years ago.

JOHN D. HALFMAN
THOMAS C. JOHNSON

Duke University Marine Laboratory,
Beaufort, North Carolina 28516

References and Notes

1. H. H. Lamb, *Palaeogeogr. Palaeoclimatol. Palaeoecol.* **10**, 125 (1971).
2. J. Williams, R. G. Barry, W. M. Washington, *J. Appl. Meteorol.* **13**, 305 (1974).
3. W. L. Gates, *J. Atmos. Sci.* **33**, 1844 (1976).
4. S. Manabe and D. G. Hahn, *J. Geophys. Res.* **82**, 3889 (1977).
5. H. T. U. Smith, *J. Geol.* **73**, 557 (1965).
6. P. F. McDowell, *Quat. Res. (N.Y.)* **19**, 100 (1983).
7. T. C. Johnson, J. D. Halfman, W. Busch, R. D. Flood, *Bull. Geol. Soc. Am.*, in press.
8. R. D. Flood and T. C. Johnson, *Sedimentology*, in press.
9. T. C. Johnson, T. W. Carlson, J. E. Evans, *Geology* **8**, 437 (1980).
10. The Keweenaw current, however, has only been studied during summer months when the lake is stratified and the current is restricted to the epilimnion [R. A. Ragotzkie, *Univ. Wis. Dep. Meteorol. Tech. Rep.* **29** (1966)].
11. The postglacial-glacial contact is marked by a change from varved, glacial-lacustrine clay to homogeneous postglacial mud. The contact is interpreted to be 9500 years old (20).
12. Model 80XY, manufactured by Particle Data.
13. The analytical procedures developed for the particle analyzer are described in J. D. Halfman [thesis, University of Minnesota, Minneapolis (1982)].
14. A phi unit is equal to the negative logarithm (base 2) of the grain's diameter in millimeters.
15. K. M. Creer and P. Tucholka, *Can. J. Earth Sci.* **19**, 1106 (1982).
16. T. C. Johnson and J. Fields, *Chem. Geol.*, in press.
17. Oriented sediment samples (2 cm³) were obtained at approximately 2.5-cm intervals for the length of each core. Magnetic measurements were made with a two-axis, superconducting magnetometer, manufactured by SCT Corporation. The noise per channel was 1×10^{-5} A/m, and the natural remanent magnetic intensities ranged between 1.2×10^{-3} and 27.8×10^{-3} . Any viscous remanent magnetization was removed by demagnetization at 100 Oe. The remaining natural remanent magnetization (nrm₁₀₀) is reported.
18. J. D. Halfman and T. C. Johnson, *Geol. Soc. London Spec. Publ.*, in press.
19. E. J. Graham and D. K. Rea, *J. Great Lakes Res.* **6**, 129 (1980); D. K. Rea, R. A. Bourbonniere, P. A. Meyers, *ibid.*, p. 321; D. K. Rea, R. M. Owen, P. A. Meyers, *Rev. Geophys. Space Phys.* **19**, 635 (1981).
20. W. R. Farrand, *Proc. Conf. Great Lakes Res.* **12**, 18 (1969).
21. M. Saarnisto, *Can. J. Earth Sci.* **12**, 300 (1975).
22. R. J. Wold, D. R. Hutchinson, T. C. Johnson, *Geol. Soc. Am. Mem.* **156**, 257 (1982).
23. C. W. Drexler, thesis, University of Michigan, Ann Arbor (1981).
24. H. E. Wright, Jr., *Proc. Conf. Great Lakes Res.* **12**, 397 (1969).
25. T. Webb, *Am. Midl. Nat.* **92**, 12 (1974).
26. H. J. B. Birks, *Quat. Res. (N.Y.)* **14**, 60 (1980).
27. C. H. Mortimer, *Mitt. Int. Ver. Theor. Angew. Limnol.* **20**, 123 (1974).
28. G. Oman, personal communication.
29. R. A. Bryson, W. M. Wendland, J. D. Ives, J. J. Andrews, *Arct. Alp. Res.* **1**, 1 (1969).
30. We thank W. E. Dean, D. A. Livingstone, and H. Wright, Jr., for their helpful reviews and S. R. Banerjee, J. Marvin, J. Fields, and J. King for the paleomagnetic analyses. Supported by NSF grants OCE-8018339 and OCE-8109833 to T. C. Johnson while he was at the University of Minnesota.

17 October 1983; accepted 21 February 1984

Precipitation of Sulfide Ores and Organic Matter: Sulfate Reactions at Pine Point, Canada

Abstract. *Bitumen is a common associate of carbonate-hosted lead-zinc deposits. On the Pine Point lead-zinc property, Northwest Territories, Canada, there are two forms of bitumen. Unaltered bitumens have atomic hydrogen/carbon ratios of about 1.4, sulfur contents of about 7.8 percent, and sulfur isotope ratios ($\delta^{34}S$) of approximately +4.6 per mil. Altered bitumens occur in proximity to sulfide ore bodies and white sparry dolomite. Their hydrogen/carbon ratios are about 1.02, the sulfur contents average 22 percent, and the $\delta^{34}S$ values are about +12.4 per mil. These data indicate that some bitumen has participated in the thermochemical reduction of sulfate to produce hydrogen sulfide required to precipitate the ores. Mass balance considerations show that the amount and degree of alteration of bitumen is more than adequate to account for the reduced sulfur species (lead, zinc, and iron sulfides) deposited at Pine Point. These reactions may provide an important means of generating the large volumes of sulfide necessary to precipitate ore bodies in carbonate rocks.*

Lead-zinc deposits hosted by unmetamorphosed sedimentary carbonate rocks are thought to originate by normal sedimentary and diagenetic processes (1). Nevertheless, these deposits present many perplexing problems, including the sources of metals and sulfides, the precipitation mechanisms, and the environment and timing of precipitation (2). Bitumen and heavy oil are often intimately associated with the ore bodies, and it has been a continuing question whether such organic matter is involved in the formation of the sulfide (1, 3).

The Pine Point lead-zinc field, Northwest Territories, Canada, is located in a Middle Devonian carbonate barrier complex (4). It has served as a model for the popular Jackson-Beales hypothesis of a sedimentary-diagenetic origin for carbonate-hosted lead-zinc deposits (5). As is common with this type of deposit, bitumen is intimately associated with the ore. Organic geochemical studies (6) have shown that the host rocks at Pine

Point occur at the threshold of petroleum generation (60°C or less), and that the heavy oil-bitumen has originated, more or less in situ, from relatively immature organic-rich rocks in the barrier complex. Fluid inclusion data indicate that the dolomitized and mineralized zones represent thermal anomalies [up to 100°C (7)] with respect to the host rocks. Within these zones, heavy oils and bitumens have been altered by heat and reaction with sulfur to form an insoluble pyrobitumen (6). Sulfur isotopic measurements on a few bitumens have suggested that thermochemical reduction of sulfate (8) by bitumen was the mechanism for formation of hydrogen sulfide and the precipitation of the metals (6).

To further evaluate this process, we collected 50 additional samples ranging from liquid heavy oil to pyrobitumen from the Pine Point property. The bitumens were analyzed for carbon, hydrogen, nitrogen, and sulfur (9), and oxygen was determined by difference after cor-

rection for ash content (Table 1). Soluble bitumens and heavy oils have high atomic H/C ratios (~ 1.44). They occur in the host rocks and are largely unaltered (6). Insoluble pyrobitumens have low atomic H/C ratios (~ 1.02) and are altered (6). They occur in white coarsely crystalline dolomite (10) or sulfide mineralization. Other bitumens have intermediate atomic H/C ratios (~ 1.20), indicating partial alteration.

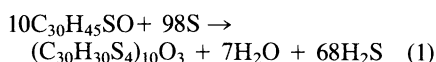
The altered bitumens have higher sulfur contents (~ 22 percent) than the unaltered bitumens (~ 7.8 percent) (Table 1). Samples with the highest atomic S/C ratios have the lowest atomic H/C ratios (Fig. 1). Analysis of the sulfur isotopic compositions of selected bitumens (Table 1 and Fig. 2) shows that there is an increase in $\delta^{34}\text{S}$ value with increasing sulfur content and that the most altered

bitumens have the most positive $\delta^{34}\text{S}$ values. These data indicate that isotopically heavy sulfur was added during the alteration process. Furthermore, the increase in sulfur content and $\delta^{34}\text{S}$ values in some of the "unaltered" bitumens shows that they also have had isotopically heavy sulfur added to them. All bitumens have low oxygen and nitrogen contents with the altered forms having slightly lower values than the unaltered forms (Table 1).

Thermal alteration of organic matter in the geological environment involves processes such as dehydration, decarboxylation, and cracking (11). In carbonate rocks, sulfurization becomes important because reactive iron is not available to remove the sulfur as iron sulfides (8). Hydrogen sulfide can react by nucleophilic substitution of oxygen and nitro-

gen, whereas elemental sulfur is an oxidizing agent and reacts readily with organic matter to form H_2S (12). These reactions can be distinguished in the bitumens if one plots elemental ratios on Van Krevelen type diagrams [Fig. 1 (13)]. Simple reactions such as loss of CO_2 and addition of sulfur are represented by lines of different slope from the point representing the sample under consideration (Fig. 1).

Figure 1 shows that the composition of the altered bitumens is due primarily to reaction of the precursor bitumen with elemental sulfur. The transformation of an unaltered bitumen to an altered bitumen (transformation A to B in Fig. 1) by reaction with sulfur must have the form:



if the proportions of carbon, hydrogen, sulfur, and oxygen in altered and unaltered materials are to be maintained. Nitrogen contents are minimal and can be neglected in this discussion (Table 1).

Hydrogen sulfide is a major product of Eq. 1 and could be the means for precipitating the metals at Pine Point. The addition of sulfur in Eq. 1 is accompanied by a decrease in the atomic O/C ratio. Part of the variation in atomic O/C ratios of the bitumens may be due to analytical error at these low oxygen contents. However, the plot of atomic O/C versus S/C ratios (Fig. 1) and the changes in $\delta^{34}\text{S}$ of the "unaltered" bitumens (Fig. 2) show that the variation in O/C ratios can also be due to nucleophilic substitution of sulfur for oxygen by H_2S . In the transformation A to B (Fig. 1), the atomic S/C ratio increased from 0.027 to 0.138 and the atomic O/C ratio decreased from 0.026 to 0.009. Thus, 15 percent of the increase could be accounted for by nucleophilic substitution of oxygen by sulfur derived from H_2S . In contrast, in the partial transformations that occur in the unaltered bitumens (for example, transformation A to C in Fig. 1), 71 percent of the small increase in sulfur content can be accounted for by substitution of oxygen by sulfur. Cracking and loss of hydrocarbons did occur and accounts for the composition of some partly altered bitumens (Fig. 1).

The stoichiometry of Eq. 1 can be used to calculate the isotopic composition of the sulfur-sulfide required to convert the least altered bitumen ($\delta^{34}\text{S} = +3.15$ per mil; point A in Figs. 1 and 2) to an altered bitumen ($\delta^{34}\text{S} = +14.5$ per mil; point B in Figs. 1 and 2). The calculated value for this reactant sulfur is $+18.3$ per mil. In a similar manner, equations for transformations A to C, A to D,

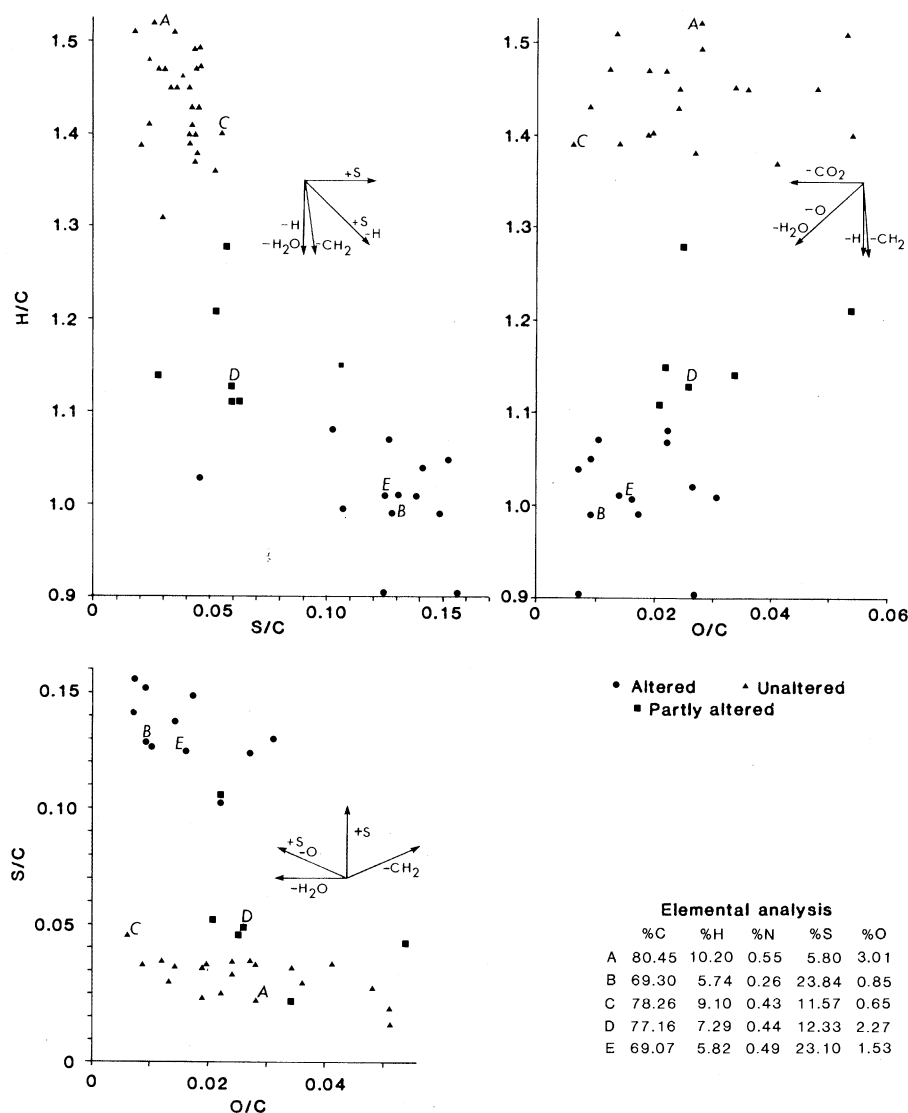
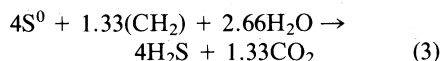
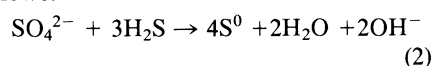


Fig. 1. Relations between atomic ratios of carbon, hydrogen, oxygen, and sulfur in the Pine Point bitumens. Lettered samples refer to samples mentioned in the text and to elemental analyses. Arrows represent trends of composition due to the following reactions: $-\text{H}_2\text{O}$ = dehydration; $-\text{CH}_2$ = cracking of hydrocarbons; $+\text{S}$ = addition of sulfur; $-\text{H}$ = dehydrogenation; $+\text{S}-\text{O}$ = substitution of sulfur for oxygen; and $-\text{CO}_2$ = decarboxylation.

and A to E (Figs. 1 and 2) give $\delta^{34}\text{S}$ values for the reactant sulfur-sulfide of +14, +31.1, and +11.9 per mil, respectively. The isotopic composition of the reactant sulfur and the native sulfur at Pine Point varies from values above to values below that for coeval sulfate [$\delta^{34}\text{S} = +19.0$ per mil (Table 1)]. This signifies the existence of a kinetic isotope effect in the reaction that produced the sulfur.

The increase in $\delta^{34}\text{S}$ values of bitumen with thermal alteration is similar to that observed for sulfur in oils and gas in some petroleum basins (8, 14). Orr suggested (8) that sulfate reacts with H_2S at temperatures above 75°C to produce elemental sulfur, which in turn reacts with organic matter to produce H_2S as follows:



These reactions have been demonstrated experimentally at a temperature of 175°C (8). Extrapolation of the kinetic data indicates their feasibility in geological systems at lower temperatures ($\sim 100^\circ\text{C}$) (8).

Equation 3 represents complete oxidation of organic matter, whereas at Pine Point (Eq. 1) oxidation of the bitumen was incomplete and was accompanied by extensive sulfurization. This reflects the mild reaction conditions (75° to 100°C), which were maintained only for the relatively short period of dolomitization and mineralization and is in contrast to the higher temperatures and millions of years available in deep petroleum basins. The reactions at Pine Point are best represented by Eqs. 1 and 2.

Hydrogen sulfide is required to initiate the reaction. At Pine Point, the initial reactant H_2S was probably formed by cracking of organic matter. Since this process occurs with negligible isotopic fractionation (8), its isotopic composition would be that of the parent bitumen ($\delta^{34}\text{S} = +3.15$ per mil). If this H_2S reacted with coeval sulfate ($\delta^{34}\text{S} = +19$ per mil), then from Eq. 2 the $\delta^{34}\text{S}$ value of the first-formed sulfur and H_2S was +7.1 per mil. If this H_2S reacted with more sulfate and so on, about ten iterations would be required for the reactant sulfur to reach a $\delta^{34}\text{S}$ value of +18.0, the value required to satisfy the isotopic relation of the transformation A to B (Fig. 2 and Eq. 1). In this process, isotopically light sulfur became incorporated into the organic matter (for example, transformations A to C and A to E in Fig. 2). This process cannot account for sulfur with $\delta^{34}\text{S}$ val-

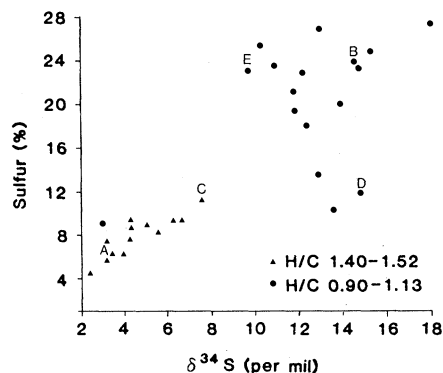


Fig. 2. Relation between sulfur isotopic composition and sulfur content in the Pine Point bitumens. The $\delta^{34}\text{S}$ values are relative to Cañon Diablo troilite. Letters refer to samples mentioned in the text.

ues in excess of that for coeval sulfate (transformation A to D in Fig. 2 and Table 1). This reaction could only occur if isotopic fractionation occurred in Eq. 2 and is in contrast to earlier suggestions that it proceeded with no isotopic fractionation (8).

The kinetic isotope effect for S-O bond rupture is of the order of 20 per mil (15). However, this fractionation effect will only be seen at places like Pine Point where intermediate reaction products in

the form of sulfur and altered bitumens with variable $\delta^{34}\text{S}$ values are preserved. The overall effect of continuing reactions is to homogenize the isotopic composition of the sulfide about the $\delta^{34}\text{S}$ value of the parent sulfate, as occurs at Pine Point (Table 1). The mechanism also explains the few low $\delta^{34}\text{S}$ values found in metallic sulfides at Pine Point [for example, +12.2 per mil for galena (16)]. The existence of isotopically heavy sulfur also requires that the sulfate reservoir be at least a partially closed system. This could occur if the rate of migration of soluble sulfate into the reaction zone, represented by the thermal anomaly, is slower than the reaction rate.

Using Eq. 1, we find that the amount of organic matter required to generate the 8.1×10^6 metric tons of reduced sulfur (17) present in the ore bodies on the Pine Point property is 16.8×10^6 metric tons, which is equivalent to 110×10^6 barrels of bitumen-heavy oil. The amount of bitumen occurring at Pine Point is not known, but 110×10^6 barrels is not unreasonable when compared with the in-place reserves of oil in Middle Devonian fields downdip from Pine Point (3929×10^6 barrels) (18). The reaction of organic matter with sulfur is not necessarily confined to bitumen. The

Table 1. Atomic H/C ratios of bitumens; oxygen, nitrogen, and sulfur contents of bitumens; and $\delta^{34}\text{S}$ values for the sulfur species at Pine Point.

Sample	Mean and standard error of mean	Range		N
<i>Atomic H/C ratio of bitumens</i>				
Unaltered	1.44 ± 0.01	1.39 to	1.52	28
Altered	1.02 ± 0.01	0.90 to	1.10	15
Partly altered	1.16 ± 0.02	1.11 to	1.28	7
<i>Oxygen contents of bitumens (%)</i>				
Unaltered	2.91 ± 0.32	0.65 to	5.76	20
Altered	1.51 ± 0.20	0.68 to	2.85	12
Partly altered	3.47 ± 0.53	2.09 to	4.77	6
<i>Nitrogen contents of bitumens (%)</i>				
Unaltered	0.45 ± 0.02	0.33 to	0.94	28
Altered	0.34 ± 0.02	0.25 to	0.49	13
Partly altered	0.51 ± 0.08	0.35 to	0.94	6
<i>Sulfur contents of bitumens (%)</i>				
Unaltered	7.80 ± 0.32	3.56 to	11.57	28
Altered	22.04 ± 1.23	9.23 to	27.63	15
Partly altered	11.94 ± 1.95	5.22 to	20.09	7
<i>δ³⁴S values of sulfur species (per mil)*</i>				
Unaltered bitumen†	+4.6 ± 0.40	+2.3 to	+7.5	13
Altered and partially altered bitumen†	+12.44 ± 0.74	+3.0 to	+17.9	17
Elemental sulfur (6, 16)	+19.02 ± 1.32	+13.3 to	+25.1	8
Sphalerite (6, 16)	+21.6 ± 0.22	+17.7 to	+24.1	55
Galena (16)	+18.4 ± 0.45	+12.2 to	+22.4	38
Pyrite (16)	+19.7 ± 0.83	+15.5 to	+23.0	9
Marcasite (16)	+19.3 ± 0.45	+15.5 to	+22.2	8
Anhydrite (SO ₄ ²⁻) (16)	+19.0 ± 0.26	+18.4 to	+19.8	4

*The sulfur isotopic ratio is given by

$$\delta^{34}\text{S} = \left[\frac{(^{34}\text{S}/^{32}\text{S})_{\text{sample}}}{(^{34}\text{S}/^{32}\text{S})_{\text{standard}}} - 1 \right] \times 10^3$$

where the standard is Cañon Diablo troilite. Enterprises Inc., Cambridge, Massachusetts.

†Analyses by Geochron Laboratories Division, Krueger

coarsely crystalline dolomite is more widespread than the ore (4). Any organic-rich rock affected by this dolomitization (and hence temperatures of 75° to 100°C) will also generate H₂S. Facies F of the barrier complex contains an average of 3.75 percent organic carbon (6). Approximately 0.13 km³ of facies F would contain the 16.8 × 10⁶ metric tons of organic matter required for the generation of sulfide at Pine Point by Eq. 1. The total volume of facies F is about 5 km³ (4), and thus only 2.6 percent would need to be fully altered for the generation of sufficient sulfide to account for the ore bodies. From these considerations, there appears to be no constraint regarding the availability of organic matter for sulfide generation.

The precipitation of metals by H₂S, formed by in situ thermochemical reduction of sulfate at Pine Point, is consistent with the temperature of formation of the ore bodies; the amounts, alteration, and composition of the bitumen; the presence of native sulfur; and the isotopic composition of the various sulfur species. Such reactions may have provided an important means of generating the large volumes of sulfide necessary to precipitate sulfide ore bodies in sedimentary carbonate rocks.

TREVOR G. POWELL*

Institute of Sedimentary and Petroleum Geology, Geological Survey of Canada, Calgary, Alberta T2L 2A7

ROGER W. MACQUEEN

Department of Earth Sciences, University of Waterloo, Waterloo, Ontario, Canada N2L 3G1

References and Notes

- G. M. Anderson and R. W. Macqueen, *Geosci. Can.* **9**, 108 (1982); L. B. Gustafson and N. Williams, in *Economic Geology, Seventy-Fifth Anniversary Volume*, B. J. Skinner, Ed. (Economic Geology, New Haven, 1981), p. 139.
- G. M. Anderson, *Econ. Geol.* **70**, 937 (1975); in *Proceedings volume, International Conference on Mississippi Valley Type Lead-Zinc Deposits*, G. Kisvarsanyi, S. K. Grant, W. P. Pratt, J. W. Koenig, Eds. (University of Missouri, Rolla, 1983), p. 61; E. L. Ohle, *Econ. Geol.* **75**, 161 (1980).
- H. E. Dunsmore, thesis, Royal School of Mines, London (1975).
- H. Skall, *Econ. Geol.* **75**, 161 (1980).
- S. A. Jackson and F. W. Beales, *Bull. Can. Pet. Geol.* **15**, 393 (1967).
- R. W. Macqueen and T. G. Powell, *Econ. Geol.* **78**, 1 (1983).
- E. Roedder, *ibid.* **63**, 439 (1968); P. Fritz and S. A. Jackson, *Proc. Int. Geol. Congr. 24th, Montreal, Sect. 6* (1972), p. 230.
- W. L. Orr, *Bull. Am. Assoc. Pet. Geol.* **58**, 2295 (1974); in *Advances in Organic Chemistry 1975*, R. Campos and J. Goni, Eds. (Enadisma, Madrid, 1977), p. 571; abstracts, Geological Society of America annual meeting, New Orleans (1982), p. 360; personal communication.
- Analyses for carbon, hydrogen, and nitrogen were performed on a Perkin-Elmer 240 Microanalyzer modified for automatic operation by Control Equipment Corporation. In the analysis for sulfur the samples were subjected to combustion and then to automatic titration (Leco) with potassium iodate-iodide solution [A. E. Foscolos and R. R. Barefoot, *Geol. Surv. Can. Pap.* **70-11** (1970)].

- Saddle dolomite of B. M. Radke and R. L. Mathis, *J. Sediment. Petrol.* **50**, 1149 (1980).
- B. P. Tissot and D. H. Welte, *Petroleum Formation and Occurrence* (Springer, Berlin, 1978).
- V. H. W. Prinzler and D. Pape, *Erdoel Kohle* **17**, 539 (1964); Y. Mehmet, *Alberta Sulfur Res. Ltd. Q. Bull.* **18** (No. 1), 1 (1971); M. Bestougeff and A. Combaz, in *Advances in Organic Geochemistry*, 1973, B. Tissot and F. Biennier, Eds. (Editions Technip, Paris, 1974), p. 747.
- B. Durand and J. C. Monin, in *Kerogen*, B. Durand, Ed. (Editions Technip, Paris, 1980), p. 113.
- H. R. Krouse, *J. Geochem. Explor.* **7**, 189 (1977).
- A. G. Harrison and H. G. Thode, *Trans. Faraday Soc.* **54**, 84 (1958).
- A. Sasaki and H. R. Krouse, *Econ. Geol.* **64**, 718 (1969).
- Ore reserves at Pine Point consist of 75.2 × 10⁶ metric tons, containing 1.9 percent lead (galena) and 5.0 percent zinc (sphalerite). The iron content (marcasite) is estimated to be 7 percent of the ore. D. W. Aldrick *et al.*, in *Field Guides to Geology and Mineral Deposits* (Calgary 1981

- annual meeting, Geological Association of Canada, Mineralogical Association of Canada, Canadian Geophysical Union), R. I. Thompson and D. G. Cook, Eds. (Department of Geology, University of Calgary, Calgary, Alberta, 1981), p. 41; J. A. Collins, personal communication.
- Alberta's Reserves, 19th Edition* (Energy Resources Conservation Board, Calgary, Alberta, 1979).
 - We thank L. R. Snowden and J. F. Barker for comments on the manuscript; Cominco Ltd., Vancouver, for permitting us to sample and describe bitumen occurrences at Pine Point; R. Fanjoy, B. Gorham, and R. A. Davidson for analytical work at the Institute of Sedimentary and Petroleum Geology, Calgary. One of us (R.W.M.) received financial support from the Natural Sciences and Engineering Research Council of Canada and the Department of Energy, Mines, and Resources.
- * Present address: Bureau of Mineral Resources, Geology, and Geophysics, Post Office Box 378, Canberra City, Australia ACT 2601.

1 September 1983; accepted 10 January 1984

Polyene Toxicity in Renal Medulla: Injury Mediated by Transport Activity

Abstract. *Polyene antibiotics such as amphotericin and nystatin increase membrane permeability and thus increase the amount of oxygen consumed in active electrolyte transport. In isolated perfused rat kidneys, the polyenes produced extensive injury to the medullary thick ascending limb, a segment of the nephron with limited oxygen supply. This damage was prevented if reabsorptive transport was inhibited by ouabain. Cell death under these circumstances thus appears to be mediated by increased oxygen demand for transport activity.*

Acute renal failure is a well-known complication of amphotericin therapy, thought to be related at least in part to the increased membrane permeability induced by polyene antibiotics (1), which leads to disruption of the internal electrolyte milieu and disordered volume regulation in cells (2). Which of the many consequences of membrane damage

caused by polyenes is ultimately responsible for cell death is unknown (3). Increased permeability triggers a compensatory increase in the rate of active electrolyte transport, associated with a rise in oxygen demand (4), but the significance of these changes for the generation of injury is not generally recognized. We report that in isolated perfused rat kid-

Table 1. Effects of amphotericin ($3 \times 10^{-5}M$) and nystatin (200 U/ml) on renal function and structure in comparison with regular perfusions (control) and perfusions equilibrated with 95 percent N₂ and 5 percent CO₂ (hypoxia). Functional parameters are whole kidney measurements and include renal perfusion flow, oxygen consumption (QO_2), and tubular reabsorption of sodium (expressed as T_{Na}/QO_2). Quantitation of histological injury (evaluated without knowledge of experimental conditions) is for the mTAL only and is expressed as the proportion of tubules affected by severe damage (illustrated in Fig. 1, A and B). The polyenes led to a recruitment of all tubules by a lesion identical to that seen in control and hypoxic perfusions and shown to be derived from local oxygen deficiency (6) but much more extensive. Treatment with ouabain ($10^{-2}M$) 20 minutes before the addition of the polyenes essentially eliminated polyene-induced damage to the mTAL. The results are expressed as means ± standard errors, for three to seven experiments per group, at 90 minutes of perfusion, and were analyzed by a multiple comparison procedure (Walker-Duncan).

Treatment	Flow (mg/min)	QO_2 (μmole/min)	T_{Na}/QO_2 (μeq/μmole)	mTAL's with severe damage (%)
Control	40.9 ± 2.5	3.8 ± 0.2	18.6 ± 1.2	62 ± 12
Hypoxia	31.9 ± 2.7	0.9 ± 0.1	8.9 ± 1.3	85 ± 6
Amphotericin	15.9 ± 3.3	4.8 ± 1.3	0.5 ± 0.4*	100 ± 0*
Nystatin	26.3 ± 3.8	3.8 ± 0.4	0.7 ± 0.5*	100 ± 0*
Ouabain and amphotericin	17.5 ± 0.6	2.4 ± 0.1	1.7 ± 0.9	6 ± 3†
Ouabain and nystatin	14.8 ± 3.1	2.7 ± 0.2	2.1 ± 0.8	0 ± 0†
Furosemide and amphotericin	26.3 ± 1.0	5.7 ± 0.5	1.8 ± 0.7	100 ± 0

*Significantly different ($P < 0.05$) from control.

†Significantly different ($P < 0.05$) from polyene alone.