Carbon Isotopic Studies of Organic Matter in Precambrian Rocks

Abstract. *Reduced carbon in early Precambrian cherts of the Fig Tree and upper and middle Onverwacht groups of South Africa is isotopically similar (the average value of $\delta^{13}C_{PDB}$ is −28.7 per mil) to photosynthetically produced organic matter of younger geological age. Reduced carbon in lower Onverwacht cherts (Theespruit formation) is anomalously heavy (the average value of $\delta^{13}C_{PDB}$ is −16.5 per mil). This discontinuity may reflect a major event in biological evolution.*

Since 1967, an impressive array of chemical fossils (such as *n*-alkanes, isoprenoids, porphyrins, and isotopically light carbonaceous material) has been detected in early Precambrian sediments (*1*). These reported occurrences, although subject to question because of possible contamination by younger organic matter (*1, 2*), are consistent with morphological fossil evidence indicating that photoautotrophs probably were extant more than 2.8 × 10^9 years ago (*1, 3*). Among the chemical evidences suggestive of early autotrophic activity, the carbon isotopic composition of particulate, insoluble organic matter (kerogen) appears to have been least subject to postdepositional contamination (*4–6*). However, $^{13}C/^{12}C$ ratios have been reported for organic components of fewer than three dozen Precambrian deposits (*1–5, 7, 8*). We have therefore undertaken a survey of the carbon isotopic composition of the total organic fraction (composed of more than 95 percent kerogen) of a suite of Precambrian sediments to detect isotopic trends possibly correlative with early evolutionary events (*9*).

Park and Epstein (*10*) have shown that photosynthetic fixation of carbon results in fractionation of the two stable carbon isotopes, ^{12}C and ^{13}C; the organic matter produced is enriched in the lighter isotope relative to the inorganic source carbon. Biogenic organic matter isolated from sedimentary rocks has $\delta^{13}C_{PDB}$ values (*11*) ranging from about −20 to −40 per mil (*12*); $\delta^{13}C_{PDB}$ values of limestones range from about +10 to −14 per mil (*13*). Because of possible variations in the carbon cycle over geologic time and the complexities introduced by diagenetic and metamorphic factors, interpretation of Precambrian carbon isotopic data is difficult. Nevertheless, biological fixation of inorganic carbon is the major process on Earth that produces an isotopic fractionation between inorganic and organic carbon of the magnitude and direction found in ancient sediments. Since the isotopic content of marine carbonates of all ages appears to vary little (*3, 12*), fossil organic matter having $\delta^{13}C_{PDB}$ values between −20 and −40 per mil seems reasonably interpreted as being of photosynthetic (*1, 3, 4, 7, 8*) or chemosynthetic (*12*) autotrophic derivation.

The $^{13}C/^{12}C$ ratios of reduced carbon were determined according to a method adapted from Craig (*13*). Each sample was ground to a powder and refluxed with redistilled benezene : methanol (70 : 30 by volume) for 24 hours. Fifteen grams of each sample were then reacted with hydrochloric acid to remove carbonates and with hydrofluoric acid to remove silicates. The resulting acid-resistant organic residue, the "total organic fraction," was combusted in oxygen at 1000°C; the carbon dioxide generated was analyzed on a dual-collecting mass spectrometer. The total organic fractions of three samples were further extracted by refluxing for 24 hours with the benzene : methanol solution; the extracted "soluble organic fraction" and the insoluble "kerogen fraction" were analyzed.

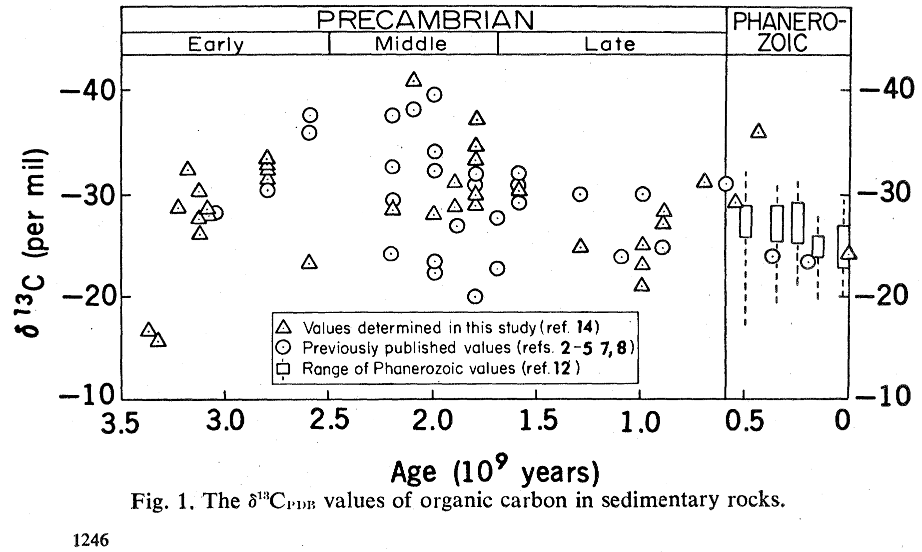

Fig. 1. The $\delta^{13}C_{PDB}$ values of organic carbon in sedimentary rocks.

The $\delta^{13}C_{PDB}$ values of the total organic fractions of 39 Precambrian cherts and limestones representing 32 localities have been determined (*14*). These data (Fig. 1) substantially increase the total number of reported analyses of Precambrian organic matter.

Reduced carbon in Phanerozoic sediments tends to exhibit increasing ^{12}C content with increasing geological age (*12*). In the Precambrian, this trend is not marked (Fig. 1), although ^{12}C content does appear to approach a maximum in sediments about 2.1 × 10^9 years old. With the exception of the very oldest samples analyzed, Precambrian organic carbon generally has $\delta^{13}C_{PDB}$ values between −25 and −35 per mil, well within the isotopic range typical of preserved biogenic organic matter.

Anomalously heavy organic carbon (having a $\delta^{13}C_{PDB}$ value greater than −20 per mil) was detected only in cherts from the lower Swaziland sequence of South Africa; these units are among the oldest sedimentary horizons known. To investigate the discrepancy between these samples and all other cherts, isotopic analyses were made of carbonaceous matter in 16 cherts representing nine stratigraphic horizons in the sequence (*15*).

The Swaziland sequence (Fig. 2), 64,000 feet (19.5 km) thick and well exposed in the Barberton Mountain Land of the southeastern Transvaal, is divided into three units (*16*). The oldest unit, the Onverwacht group, consists of lavas, cherts, and sedimentary carbonates. The Fig Tree group, conformably overlying the Onverwacht, includes shales, graywackes, banded cherts, and lavas. The Moodies group overlies the Fig Tree. The Onverwacht and Fig Tree cherts are thought to have been deposited in a subaqueous, basin-like environment and to have experienced little subsequent alteration (*16*). Rubidium and strontium measurements on intrusives in the Fig Tree group indicate an

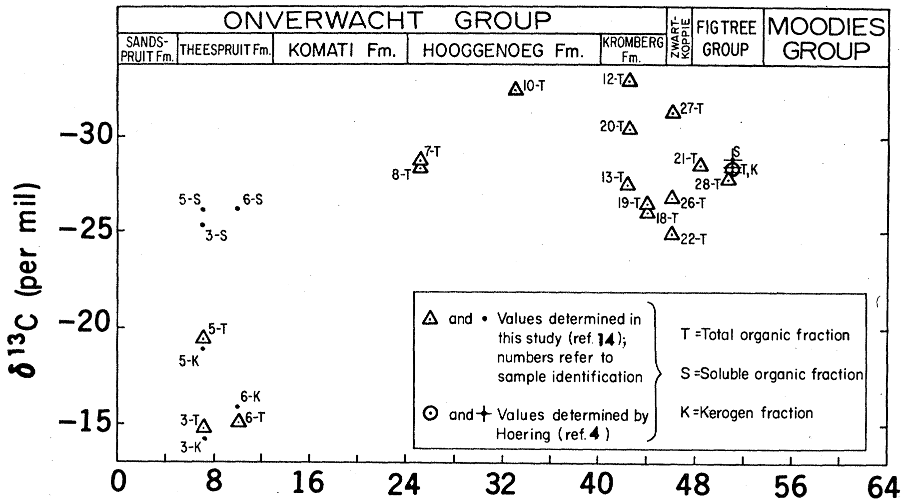

Fig. 2. The $\delta^{13}C_{PDB}$ values of organic carbon in sediments of the Swaziland sequence.

age in excess of 3 × 10^9 years (*17*); a maximum age for the sequence of about 3.4 × 10^9 years is suggested by rubidium-strontium and lead-lead analyses (*18*).

In Fig. 2 the $\delta^{13}C_{PDB}$ values of the analyzed Swaziland samples are shown. The values fall into two distinct categories. One category is comprised of 13 cherts stratigraphically ranging from the middle third of the Onverwacht group (Hooggenoeg formation) to the middle of the Fig Tree group; the $\delta^{13}C_{PDB}$ values range from −25.0 to −33.0 per mil with an average of −28.7 per mil. Hoering's $\delta^{13}C_{PDB}$ value of −28.3 per mil (*4*) for organic matter in a Fig Tree shale falls within this range. The second category is composed of three samples from the lowest third of the Onverwacht group (Theespruit formation); the $\delta^{13}C_{PDB}$ values of these samples range from −14.7 to −19.5 per mil with an average of −16.5 per mil. The $\delta^{13}C_{PDB}$ values of the soluble organic fractions of these three samples range from −25.5 to −26.2 per mil, presumably reflecting a relatively recent origin [see (*4, 5*)]. In contrast, values of the kerogen fractions, apparently syngenetic with Theespruit deposition, range from −14.3 to −18.9 per mil.

Thus, there is an isotopic discontinuity of about 12 parts per thousand between Theespruit and younger organic matter in the Swaziland sequence. Since all Swaziland cherts were analyzed similarly and since triplicate analyses of one Theespruit sample gave highly consistent results (sample 3-T in Fig. 2; $\delta^{13}C_{PDB} = -15.5, -14.8$, and −14.7 per mil), it is unlikely that the unusually heavy isotopic composition of Theespruit carbon is an artifact of experimental procedure.

It might be suggested that the isotopically heavy nature of Theespruit reduced carbon has resulted from metamorphic loss of light carbon species (*19*). In this regard, chert from the middle Hooggenoeg formation, collected immediately adjacent to an intrusive body, provides an indication of possible isotopic effects of contact metamorphism on Swaziland organic carbon. The $\delta^{13}C_{PDB}$ value of reduced carbon in this chert (sample 10-T, −32.5 ± 0.4 per mil) does not differ significantly from values typical of organic matter in other cherts of similar stratigraphic position (Fig. 2), most of which are far removed from zones of contact metamorphism. Since the Theespruit samples were collected more than 0.4 km away from known intrusives, it seems unlikely that their atypical isotopic composition has resulted from contact metamorphism. Moreover, the Swaziland samples analyzed are from a conformable succession and have been subject to similar depositional and tectonic environments, so that isotopic alteration by diagenesis or regional metamorphism would be expected to have occurred similarly throughout the sequence. The difference in isotopic composition between Theespruit and younger Swaziland carbon is probably not, therefore, a result of postdepositional alteration, a conclusion consistent with the results of other studies (*8, 12, 20*). Thus, the two categories of reduced carbon detected in Swaziland sediments may be of differing origins.

In conclusion, the similarities in the isotopic content of reduced carbon in all but the oldest Precambrian samples analyzed (Fig. 1), together with the essentially constant isotopic composition of oxidized carbon in marine carbonates deposited during the past 2.8 (*3, 12*) or perhaps 3.3 × 10^9 years (*21*), seem consistent with the existence of autotrophic organisms since the deposition of the middle third of the Onverwacht group, approximately 3.3 × 10^9 years ago. Kerogen in lower Onverwacht Theespruit cherts has $\delta^{13}C_{PDB}$ values which fall outside the normal range for preserved reduced carbon of established biological origin but which are comparable to values of about −16 per mil characteristic of primordial organic matter in carbonaceous chondrites (*12, 22*). The isotopic discontinuity between Theespruit and younger cherts is difficult to interpret because of uncertainties regarding the nature of the early carbon cycle and the primitive environment. Possibly this break reflects a geological event that altered the distribution of isotopes in the carbon reservoirs; or, it may mark the time of origin of biochemical mechanisms capable of fractionating carbon isotopes in a manner similar to that of modern autotrophs. The Theespruit organic carbon may even, in part, be a remnant of abiologically produced organic material, isotopically similar to that present in carbonaceous chondrites. Although such speculations are consistent with the known early Precambrian fossil record (*1*) and find marginal support in reported chemical analyses of Onverwacht organic matter (*6, 23*), the data are so few and their potential implication so far-reaching that no firm conclusion should be drawn at the present time.

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References and Notes

1. J. W. Schopf, *Biol. Rev. Cambridge Phil. Soc.* **45**, 319 (1970).
2. J. W. Smith, J. W. Schopf, I. R. Kaplan, *Geochim. Cosmochim. Acta* **34**, 659 (1970).
3. J. W. Schopf, D. Z. Oehler, R. H. Horodyski, K. A. Kvenvolden, *J. Paleontol.* **45**, 477 (1971).
4. T. C. Hoering, in *Researches in Geochemistry*, P. H. Abelson, Ed. (Wiley, New York, 1967), vol. 2, pp. 88–111.
5. ———, *Carnegie Inst. Washington Yearb.* **64**, 215 (1965); *ibid.* **65**, 365 (1967).
6. W. M. Scott, V. E. Modzeleski, B. Nagy, *Nature* **225**, 1129 (1970).
7. M. M. Roblot, M. Chaigneau, M. Majzoub,

C. R. Acad. Sci. **258**, 253 (1964); J. Hoefs and M. Schidlowski, *Science* **155**, 1096 (1967); E. S. Barghoorn and S. A. Tyler, *ibid.* **147**, 563 (1965); E. S. Barghoorn, W. G. Meinschein, J. W. Schopf, *ibid.* **148**, 461 (1965).

8. E. Bondensen, K. R. Pedersen, O. Jørgensen, *Medd. Groenland* **164**, No. 4 (1967).
9. D. Z. Oehler and J. W. Schopf, *Amer. J. Bot.* **58**, 471 (1971).
10. R. Park and S. Epstein, *Geochim. Cosmochim. Acta* **21**, 110 (1960); *Plant Physiol.* **36**, 133 (1961).
11. The $\delta^{13}C_{PDB}$ (per mil) value is defined as

$$\frac{(^{13}C/^{12}C)_{sample} - (^{13}C/^{12}C)_{PDB}}{(^{13}C/^{12}C)_{PDB}} \times 1000$$

where PDB refers to the Pedee belemnite standard from the Pedee formation, Upper Cretaceous of South Carolina.

12. E. T. Degens, in *Organic Geochemistry*, G. Eglinton and M. T. J. Murphy, Eds. (Springer-Verlag, New York, 1969) pp. 304–329.

→ H. Craig, *Geochim. Cosmochim. Acta* **3**, 53 (1953).

14. The following $\delta^{13}C_{PDB}$ values were determined. The abbreviations are T (total organic fraction), S (soluble organic fraction), K (kerogen fraction), BY (approximate age × 10^9 years), sample (sample number shown on Fig. 2).

1) Swaziland samples (cherts, 3.4 to 3.0 BY). Lower third Theespruit formation, 32 km southwest of Barberton: sample 5, $T = -19.5$, $S = -26.1$, $K = -18.9$ per mil; sample 3, $T = -15.5$, $T = -14.8$, $T = -14.7$, $S = -25.5$, $K = -14.3$ per mil. Upper third Theespruit formation, 32 km southwest of Barberton: sample 6, $T = -15.1$, $S = -26.2$, $K = -15.8$ per mil. Lower third Hooggenoeg formation, 27 km southwest of Barberton: sample 7, $T = -28.8$ per mil; sample 8, $T = -28.4$ per mil. Middle third Hooggenoeg formation, 38 km south-southwest of Barberton: sample 10, $T = -32.1$, $T = -32.9$ per mil. Middle third Kromberg formation, 29 km south-southwest of Barberton: sample 12, $T = -33.0$ per mil; sample 13, $T = -27.5$ per mil; sample 20, $T = -30.6$ per mil. Upper third Kromberg formation, 29 km south of Barberton: sample 18, $T = -26.1$ per mil; sample 19, $T = -26.2$ per mil. Zwartkoppie formation, 11 km northeast of Barberton: sample 22, $T = -24.9$ per mil. Zwartkoppie formation, 24 km east-northeast of Barberton: sample 26, $T = -26.9$ per mil. Zwartkoppie formation, 23 km east-northeast of Barberton: sample 27, $T = -31.4$ per mil. Lower third Fig Tree group, 10 km northeast of Barberton: sample 21, $T = -28.7$ per mil. Middle third Fig Tree group, 13 km south-southeast of Barberton: sample 28, $T = -28.0$ per mil.

2) Other Precambrian samples ($\delta^{13}C_{PDB}$ total organic fractions). Limestone: 3.3 to 2.8 BY, Bulawayan group, Rhodesia (three different samples), −33.5, −32.1, −32.5 per mil. Cherts: 3.3 to 2.8 BY, Bulawayan group, Rhodesia, −31.8 per mil; 2.6 BY, Keewatin, Schreiber, Ontario, −23.2 per mil; 2.2 BY, middle Fortescue group, Western Australia, −28.4 per mil; 2.2 BY, upper Fortescue group, Western Australia, −40.8 per mil; 2.0 BY, Transvaal System, South Africa, −28.0 per mil; 1.9 BY, Wittenoom Dolomite, Hamersley group, Western Australia, −29.0 per mil; 1.9 BY, Wittenoom Dolomite, Hamersley group, Western Australia, −31.2 per mil; 1.9 BY, Wittenoom Dolomite/basal Brockman Iron formation, Hamersley group, Western Australia, −30.2 per mil; 1.8 BY, middle to lower Brockman Iron formation, Hamersley group, Western Australia, −33.4 per mil; 1.8 BY, upper Brockman Iron formation, Hamersley group, Western Australia, −29.3 per mil; 1.8 BY, Gunflint Iron formation, Schreiber Beach, Ontario, −37.2 per mil; 1.8 BY, Gunflint Iron formation, Nolalu, Ontario, −34.1 per mil; 1.6 BY, Koolpin Chert, South Alligator group, Northern Territory, Australia, −31.0 per mil; 1.3 BY, Beck Spring Dolomite, California, −25.0 per mil; 1.0 BY, Skillogalee Dolomite, Yatina, South Australia, −21.0 per mil; 1.0 BY, Skillogalee Dolomite, Mundallio Creek, South Australia, −23.0 per mil; 1.0 BY, Skillogalee Dolomite, Depot Creek, South Australia, −25.2 per mil; 0.9 BY, Bitter Springs formation, Jay Creek, central Australia, −28.4 per mil; 0.9 BY, Bitter Springs formation, Ellery Creek, central Australia, −27.2 per mil; 0.7 BY, Conception group, East Newfoundland, −31.2 per mil.

3) Phanerozoic samples (cherts, $\delta^{13}C_{PDB}$ total organic fractions). Upper Cambrian, Catlin member, Windfall formation, Nevada, −29.4 per mil; Upper Ordovician, Maravillas Chert, W. Texas, −36.1 per mil; Upper Tertiary, Lost Chicken Creek formation, Alaska, −24.2 per mil.

15. These samples were collected in May 1968 by K.A.K.
16. C. R. Anhaeusser, C. Roering, M. J. Viljoen, R. P. Viljoen, *Information Circular No. 38* (Economic Geology Research Unit, University of Witwatersrand, Johannesburg, 1967); M. J. Viljoen and R. P. Viljoen, *ibid., No. 36* (1967).
17. H. L. Allsopp, T. J. Ulrych, L. O. Nicolaysen, *Can. J. Earth Sci.* **5**, 605 (1968).
18. T. J. Ulrych, A. Burger, L. O. Nicolaysen, *Earth Planet. Sci. Lett.* **2**, 179 (1967).
19. D. R. Baker and G. E. Claypool, *Amer. Ass. Petrol. Geol. Bull.* **54**, 456 (1970); F. Barker and I. Friedman, *Geol. Soc. Amer. Bull.* **80**, 1403 (1969).
20. F. E. Wickman, *Geochim. Cosmochim. Acta* **3**, 244 (1953); *ibid.* **9**, 136 (1956); S. Gavelin, *ibid.* **12**, 297 (1957).
21. J. R. Vail and M. H. Dodson, *Trans. Geol. Soc. S. Afr.* **72**, 79 (1969).
22. J. W. Smith and I. R. Kaplan, *Science* **167**, 1367 (1970).
23. M. Calvin, *Chemical Evolution* (Oxford Univ. Press, New York, 1969), pp. 85–88; W. D. MacLeod, Jr., *J. Gas Chromatogr.* **6**, 591 (1968).

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Decreased Concentration of Phenacetin in Plasma of Cigarette Smokers

Abstract. *The amount of phenacetin in plasma was determined in nine control subjects (nonsmokers) and nine subjects who smoked at least 15 cigarettes per day. The mean plasma concentration of phenacetin at 1, 2, 3½, and 5 hours after its administration was markedly lower in cigarette smokers than in nonsmokers. At 2 hours after the oral administration of 900 milligrams of phenacetin, the plasma concentration (± standard error) of unchanged drug was 2.24 ± 0.73 micrograms per milliliter in the controls and 0.48 ± 0.28 micrograms per milliliter in the smokers. The rate of excretion in urine of the major metabolite of phenacetin,* N-*acetyl-*p-*aminophenol, was the same in both groups. These results indicate for the first time decreased concentrations of a drug in plasma of persons who smoke cigarettes, and the results suggest that the decrease in the amount of phenacetin in plasma may result from increased metabolism of phenacetin in cigarette smokers.*

The administration to rats of many polycyclic aromatic hydrocarbons that are present in cigarette smoke (*1*) (such as 3,4-benzpyrene; 1,2,5,6-dibenzanthracene; and 1,2-benzanthracene) causes an increase in the activity of microsomal enzymes that metabolize certain carcinogens and drugs (*2*). The activities of benzpyrene hydroxylase and aminoazo dye *N*-demethylase, which are enzyme systems inducible by polycyclic hydrocarbons, were greatly increased in term placentas of individuals who smoke cigarettes (*3*). Increased benzpyrene hydroxylase activity was also found in the lung, the liver, the intestine, and the placenta of rats exposed to cigarette smoke (*4*).

We report here, for the first time, an effect of cigarette smoking on the plasma concentration of a drug. Phenacetin, widely used as an analgesic and an antipyretic, was selected for study because the conversion of this drug to its major metabolite, *N*-acetyl-*p*-aminophenol (APAP) (*5*), has been shown to be inducible in animals by polycyclic hydrocarbons (*6*).

Eighteen healthy volunteers, ranging in age from 24 to 35 years, included five female and four male cigarette smokers and five female and four male nonsmokers. The number of cigarettes smoked per day by the smokers varied from 15 to 40. The mean weight of the subjects (± standard error) was 123.2 ± 5.3 pounds (1 pound = 0.45 kg) for female smokers, 119.0 ± 4.9 pounds for female nonsmokers, 154.5 ± 9.8 pounds for male smokers and 159.0 ± 4.8 pounds for male nonsmokers. No drugs were taken by any subject for 2 weeks prior to the study. The ingestion of coffee and alcohol by these subjects was modest, and the amount consumed was the same for smokers and nonsmokers.

The cigarette smokers were not permitted to smoke for 8 hours preceding the administration of phenacetin and for the first 5 hours after drug administration. All subjects fasted overnight, except for water as they desired, and at approximately 8 a.m. on the day of the study they were given 900 mg of phenacetin (as a fine powder)