Antiquity of the biological sulphur cycle: evidence from sulphur and carbon isotopes in 2700 million-year-old rocks of the Belingwe Belt, Zimbabwe

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Sulphur and carbon isotopic analyses on small samples of kerogens and sulphide minerals from biogenic and non-biogenic sediments of the 2.7×10^9 years (Ga)-old Belingwe Greenstone Belt (Zimbabwe) imply that a complex biological sulphur cycle was in operation. Sulphur isotopic compositions display a wider range of biological fractionation than hitherto reported from the Archaean. Carbon isotopic values in kerogen record fractionations characteristic of rubisco activity, methanogenesis and methylotrophy, and possibly anoxygenic photosynthesis. Carbon and sulphur isotopic fractionations have been interpreted in terms of metabolic processes in 2.7 Ga prokaryote mat communities, and indicate the operation of a diverse array of metabolic processes. The results are consistent with models of early molecular evolution derived from ribosomal RNA.

Keywords: Archaean; sulphur isotopes; carbon isotopes; photosynthesis; sulphur cycle; Belingwe Greenstone Belt

1. INTRODUCTION

Models of microbial evolution based on ribosomal RNA (rRNA) (Woese 1987; Pace 1997) imply that microbial metabolic diversity is of great antiquity. Recently, this so-called 'standard' model has been challenged, reinterpreting various aspects of the molecular evidence (Doolittle 1999, 2000). There is a need for an independent geological test. However, geological evidence for the early microbial record is fragmentary. Morphological remains are difficult to interpret. To resolve this, a fruitful line of enquiry is the investigation of the history of metabolism using isotopic and chemical tracers.

Geological evidence for the antiquity of major biochemical cycles gives varying results. For the sulphur cycle (S-cycle), constraints are few. Most importantly, the restricted range of $\delta^{34}\text{S}$ found in old rocks has been interpreted as evidence that the full S-cycle, with both sulphur-reducing and sulphur-oxidizing metabolic skills, did not evolve until relatively late in Earth history. Canfield & Teske (1996) suggested that evolution of non-photosynthetic sulphide-oxidizing bacteria took place after about 1.05×10^9 years (Ga) ago. Habicht & Canfield (1996) concluded that high sulphate concentrations give rise to highly S^{34} -depleted sulphides in modern deposits, and thus inferred that appreciable concentrations of seawater sulphate did not accumulate until atmospheric oxygen increased in post-Archaean times. However, it is possible that the evolution of metabolic innovations was long separated in time from their geochemical expression (Canfield *et al.* 2000). This geologically shaped 'late' view of the S-cycle is hard to reconcile with 'standard' models of microbial evolution

derived from the rRNA molecular biology of extant organisms (Woese 1987; Pace 1997; but see criticism by Doolittle 1999, 2000). These rRNA models show early branching leading to extant forms capable of a wide range of S oxidation and reduction processes. If this is correct, 'sulphur-etum' systems may be very old. In these systems, microbial communities exploit the full range of the S-redox spectrum, with some organisms reducing sulphate while others reoxidize the reduced sulphur species. The fundamental thermodynamic drive comes either from (i) reacting a more oxidized atmosphere against a more reduced subsurface-derived chemical species that has been exposed to mantle-derived igneous melts; or (ii) from photosynthetic production of reduced carbon, which is then recycled by reaction against the resulting enhanced oxidation power of the air–water system.

Evidence for the great antiquity of biological carbon management (Mojzsis *et al.* 1996) is widespread (e.g. Nisbet & Fowler 1996a,b; Nisbet 2000). Since 3.5 Ga ago or earlier, a substantial fraction (15–25%) of the carbon degassed from the Earth's interior has been segregated as isotopically depleted biological carbon, leaving an inorganic reservoir (75–85%) enriched in ^{13}C (e.g. Schidlowski *et al.* 1975; Abell *et al.* 1985). The magnitude of the isotopic fractionation is unambiguous evidence that the enzyme involved in capturing the carbon was rubisco. The widespread presence in Archaean rocks of carbonate with $\delta^{13}\text{C}$ of 0‰ implies that carbon management by rubisco was on a global scale. The only rubisco-using organisms capable of handling carbon on this scale are the oxygenic photosynthesizers. Thus it is thought that large-scale carbon management must have begun with the evolution of the cyanobacteria: the first organisms capable of managing the global C budget. Non-photosynthetic bacteria do use rubisco, but in the absence

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of oxygenic photosynthesis it is improbable that their productivity would have been large enough to manage the global carbon budget of the atmosphere–ocean system.

Morphological evidence from stromatolites implies (though does not prove) that cyanobacteria may have been present since 2.7 Ga ago and possibly since at least 3 Ga ago (summarized by Lowe 1994), supporting the isotopic deduction that they have been present since before 3.5 Ga ago (Schidlowski & Aharon 1992). Molecular evidence confirms the presence of cyanobacteria in 2.7 Ga rocks (Brocks *et al.* 1999; Summons *et al.* 1999). The ability to manage the oxidation state of varied S species is possessed by many deeply rooted bacteria and archaea. The metabolic implications of so-called (Doolittle 2000) ‘standard’ rRNA models (Woese 1987; Pace 1997) would suggest that the full S-cycle predates oxygenic photosynthesis, and is likely to have been in operation before the time that cyanobacteria had evolved and were building diverse stromatolites. Whether or not the ‘standard’ models are correct, there are strong grounds to infer that the S-cycle is of the greatest antiquity. There is thus an inherent conflict between the supposed antiquity of the rubisco-managed carbon cycle and the apparent youth (Habicht & Canfield 1996) of the geological evidence for the modern S-cycle. Possibly the distribution of oxidant in the atmosphere–ocean–mud system may have been significant. More evidence is needed.

In contrast to the apparent antiquity of the redox metabolic skills of bacteria and archaea implied by ‘standard’ rRNA-based trees, the ‘standard’ model of atmospheric evolution (Holland 1999) is that the atmosphere took on its modern oxic nature only halfway through the planet’s history, in a ‘great oxidation event’ about 2.05–2.25 Ga ago. This model, too, is currently in debate (Ohmoto 1997). Note that the standing amount of oxygen in the air is not directly related to the productivity of photosynthesis, but to the controls on the way oxidant is fluxed through the ocean–atmosphere system (Veizer *et al.* 1982). Consider the height of water in a bathtub. It is not directly linked to the input from a tap, but rather to whether the plug is in or out. In one case, there will be no water in the bath even if the tap is full on; in the other case, the bath will fill and spill even if the tap is only dripping. However, if the hypothesis of a ‘great oxidation event’ is correct, the standard atmospheric model would suggest that prior to the event, even if bacteria capable of living on redox contrast were present, they would have been broadly dependent on SO_x or NO_y, not directly on O₂, except locally, close to cyanobacteria.

2. SAMPLING AND ISOTOPIC METHODOLOGY

The geological study of the evolution of metabolic processes in Archaean (from about 4–2.5 Ga ago) depends largely on isotopic and molecular fingerprinting, as prokaryotes leave few visible remains. Given the absence of bioturbation by metazoa, the discrete metabolic layers in Archaean microbial mat communities may not have been more than a few millimetres thick. Conventional isotopic techniques, with large samples and few analyses, may not have adequate resolution to study the fine metabolic layering of mats and biofilms, especially after the vagaries of burial, deformation and metamorphism (Ohmoto 1992).

In this study, using material from the 2.7 Ga Belingwe Greenstone Belt, Zimbabwe, a large number of samples has been analysed using a rapid high-resolution technique. Some rocks from the Belingwe Belt are among the best preserved of all Archaean remains, having undergone very low metamorphic temperatures and very low strain. As an example of the low metamorphic grade, komatiites from the Reliance Formation (Fm.) stratigraphically close to some rocks studied here, contain the only relict glass in the Archaean (Nisbet *et al.* 1987). Other Belingwe rocks are more strained, but this remains one of the few Archaean sites known where the exceptional quality of preservation justifies very high-resolution study of biochemical relicts.

This study is based on the Manjeri and Cheshire Fms (see maps in Bickle & Nisbet (1993)). Beach and shallow-water sediments of the Spring Valley Member, the base of the Manjeri Fm., rest unconformably on older (3.5 Ga) gneissic crust. The Spring Valley sediments include silts, sands, oxide ironstones, stromatolitic limestones (Martin *et al.* 1980; Abell *et al.* 1985) and some iron sulphide stringers. The Manjeri Fm. succession continues with the clastic Rubweruchena Member (Hunter *et al.* 1998), followed by a sulphide–carbon–chert horizon, the Jimmy Member, that has fine laminations, which may have been laid down in somewhat deeper water, below the wave base. The Jimmy Member is sheared and its origin is disputed (Kusky & Winsky 1995), but we consider the sequence to be *in situ* and coherent (Bickle & Nisbet 1993). This Member is extensive and stratigraphic evidence suggests that it may originally have been widespread over a few square kilometres. This precludes origin as a localized volcanic-exhalative mat, although rare-earth element (REE) data (Hunter *et al.* 1998) indicate that distal volcanism may have enriched the water in metals and sulphur species. Komatiite lavas and basalts overlie the Manjeri Fm. The Cheshire Fm., much higher in the stratigraphy above the basalts, includes shales and extensive stromatolitic limestones (Martin *et al.* 1980; Abell *et al.* 1985).

Samples studied from the Manjeri Fm. include (i) stromatolite samples; (ii) drill core of black shales and cherts containing sulphides and organic carbon (Spring Valley Member); and (iii) drill core from the sulphur–carbon–chert-shales section (Jimmy Member). In addition, (iv) carbon isotopic compositions of total organic carbon and carbonates were analysed on material from the Cheshire Fm. shales and stromatolites to complement previous work (Abell *et al.* 1985). The samples were hand-picked from material that was well preserved, typically with very fine sub-millimetre-scale lamination and textures that appeared to be primary.

Sulphur and carbon isotopes were analysed using a CF-IRMS technique (continuous flow-elemental analyser system, Fisons Carlo Erba NA1500, on-line to a VG Optima (Micro-mass, UK) isotope ratio mass spectrometer). This method has been recently described by Grassineau *et al.* (2000). The weights used vary from 0.8 to 3.5 mg for analysis of sulphide minerals containing 53–13 wt% of S, and from 0.5 to 25 mg for samples containing 20–0.5 wt% of C. The samples are hand-picked and crimped in a tin capsule. Then they are dropped into the reactor at *ca.* 1030 °C, under O₂ flux producing a flash combustion at 1800 °C. The released gas is carried through an oxidation–reduction column and water trap by a continuous He flow, passing through the chromatographic column, which separates the different gas species. The He flow finally directs the eluting gas pulses to the thermal conductivity detector and the mass spectrometer where the gas under study is isotopically analysed.

Figure 1. Distribution of the carbon isotopic compositions in the Manjeri and Cheshire Fms of the 2.7 Ga Belingwe Greenstone Belt. Asterisks, samples from Abell *et al.* (1985); \$-data cited by Strauss & Moore (1992). Other results from this study. Some of the $\delta^{13}\text{C}_{\text{org}}$ in the Jimmy Member are lighter than the values of the Spring Valley Member, indicating likely methanogenic and methanotrophic processes in addition to photosynthesis. This process is dominant in the Cheshire shales.

SO_2 and CO_2 are analysed separately because their extraction requires different oxidation–reduction reactors and gas chromatography (GC) columns. The technique has many improvements over conventional methods for analysing these two isotopes, in particular for $\delta^{34}\text{S}$. Whereas conventional methods need long analysis times and > 10 mg of sample, this technique for $\delta^{34}\text{S}$ has rapid analysis times (7 min 30 s), and uses small samples (pyrite 0.8–1.2 mg; pyrrhotite 1.4–1.6 mg), allowing the resolution needed for study of fractionation in biofilms in Archaean sulphide-rich organic sediments. The average reproducibility for homogeneous natural minerals is $\pm 0.1\text{‰}$ (1 σ). The rapidity of the technique allows 70–80 analyses per day, after 24 h for system set-up and stabilization.

Analytical error, derived from repeated calibration experiments, is better than conventional techniques, and with its high precision, small sample size and large number of samples analysed, this technique offers far better insight into the relicts of a fine-scale microbial sedimentary community than has hitherto been possible.

3. ISOTOPIC RESULTS

There is wide evidence for the presence of organic carbon in the Manjeri Fm.: the $\delta^{13}\text{C}$ results from this and earlier works on Belingwe (Abell *et al.* 1985) are summarized in figure 1. Some small samples are very rich in what appears to be organic carbon (*ca.* 10 wt% C_{org}). Diagenetic and metamorphic processes tend to shift $\delta^{13}\text{C}$ of organic carbon to less negative values, but such shifts are small for the temperatures experienced by the Manjeri sediments (Abell *et al.* 1985), which are of low metamorphic grade (see descriptions in Bickle & Nisbet 1993).

If metamorphism has shifted isotopic ratios, this reinforces the conclusion of organic origin.

Samples with $\delta^{13}\text{C}$ *ca.* -30 to -25‰ show the isotopic fingerprint of rubisco (*ca.* 29‰ fractionation in an open system), implying the activity of aerobic bacteria using the Calvin–Benson cycle, while isotopically lighter kerogen ($\delta^{13}\text{C}$ *ca.* -40‰) may include carbon further recycled via methanogens to methylotrophs. Samples with $\delta^{13}\text{C}$ *ca.* -15 to -5‰ have various possible origins (e.g. rubisco-mediated capture when the CO_2 supply is inadequate, or mixing of depleted and less-depleted material on a scale smaller than the sample), but perhaps the simplest possibility is that at least some values result from anaerobic carbon capture processes, or alteration. $\delta^{13}\text{C}$ around $0 \pm 0.5\text{‰}$ in carbonate from stromatolites confirms the view (Schidlowski & Aharon 1992) that the modern oxygenic photosynthetic carbon cycle was in operation, controlling the isotopic composition of ambient CO_2 in the water–air system.

S-isotopic compositions for 161 sulphide samples are shown in figure 2. Nearly all analyses are well duplicated, with reproducibility in 80% of samples better than $\pm 0.15\text{‰}$. The exceptions are discussed below. A striking feature of the $\delta^{34}\text{S}$ is the wide range in each Manjeri Fm. unit, with a combined range of about 37‰. Although the sample population is large, this distribution is orders of magnitude greater than analytical error, and clearly real. The range is wider than reported hitherto from the Archaean. In contrast, most conventional $\delta^{34}\text{S}$ studies have typically reported small ranges ($0 \pm 5\text{‰}$; Habicht & Canfield 1996). The large sample population, and the small sample size may both be

Figure 2. Sulphur isotopic compositions from the Spring Valley and Jimmy Members, Manjeri Fm. The bimodal distribution in the Jimmy Member peaks, between -6 and -5 ‰, and around 0 ‰, indicate two different processes. In the Spring Valley Member, the unimodal distribution pattern suggests only one dominant process.

important in detecting a wide isotopic range, as previously only studies with large numbers of samples (Goodwin *et al.* 1976; Hayes *et al.* 1992; see also Strauss & Moore (1992) and works cited within) have recorded wide isotopic ranges.

In modern sediments, the activity of burrowing organisms supplies sulphate to sediments to depths *ca.* -0.5 m below the surface. This is inferred from essentially constant pore-water concentration profiles of dissolved ions in horizons where sulphate reduction is demonstrated by the presence of iron sulphides (Goldhaber & Kaplan 1980). Below this level, sulphate is further reprocessed by microbial organisms, with increasing fractionation downwards. In Archaean sediments, though very simple single-celled eukaryotes were probably present (Summons *et al.* 1999), there is no evidence for assemblies of cells or for larger multicelled eukaryotes (with their internal management of redox). Archaean-living organisms, before the evolution of metazoan bioturbation, presumably did not have the ability to move fluids significantly. Thus diffusion- rather than advection-controlled biological processes in Archaean microbial biofilms probably dominated the sulphate supply, and redox zonation took place on a much finer scale than in equivalent modern settings (Nisbet & Fowler 1999). This limitation on the depth and rate of sulphate supply from the surface downwards would have had isotopic consequences. Isotopic zonation of sulphur would have been on a finer scale than the Precambrian and Phanerozoic sediments laid down after metazoa evolved.

If this is so, detection in Archaean sediments of isotopic stratification will be more difficult than in later sediments. Conventional S-isotopic techniques may lack the resolution to detect primary biogenic heterogeneities. Conventional studies, involving large individual samples would average out the finely fractionated, highly localized relict biological fingerprint of an early microbial community, unless large sample populations are

measured, which accidentally include localized extremely fractionated larger relicts of microbial guilds.

The reproducibility on replicated samples varies from ± 0.0 to ± 1.8 ‰ (average = ± 0.1 ‰) for a population of 161 $\delta^{34}\text{S}$ -values ranging from -19.9 to $+16.8$ ‰ (average = -2.6 ± 5.8 ‰). If three different subgroups are selected among these $\delta^{34}\text{S}$ -values, the range between -6.9 and $+5.4$ ‰ shows a reproducibility of ± 0.1 ‰ ($n=129$). The reproducibility averages ± 0.3 ‰ for the ^{34}S -depleted samples between -19.9 and -7.7 ‰ ($n=22$), whereas those more ^{34}S -enriched, from $+7.4$ to $+16.7$ ‰, show more noticeable heterogeneity in the replicates, averaging ± 0.9 ‰ ($n=10$). This isotopic heterogeneity of the sulphide minerals is probably on a sub-millimetre scale in the samples measured in this study. For example, generally in less C-rich samples and in control standards the replication in isotopic measurements is excellent (± 0.1 ‰). Despite this, there was difficulty in reproducing precisely the $\delta^{34}\text{S}$ of the sulphides in a few specific samples. This was found particularly in samples that contain visible 'blebs' of organic matter. These 'blebs' of organic appearance in some cases contain over 18 wt% C_{org} and have light $\delta^{13}\text{C}$. In these samples, the $\delta^{34}\text{S}$ replicates for *ca.* 1 mg samples may diverge as much as 1.8‰. This difficulty of replication in very organic-rich samples may be an analytical problem, but in general the technique is robust (Grassineau *et al.* 2000), which suggests the heterogeneity is real, caused by the presence of primary biogenic isotopic variation on a very small scale. Such heterogeneity is difficult to resolve even by this high-resolution technique, and is detected here by the systematic study of a large population of small samples, a few of which include very fractionated aggregations of microbial debris.

4. INTERPRETATION

The distribution of $\delta^{34}\text{S}$ -values in the histogram (figure 2) is large. Open-system formation of sulphide by sulphate reduction from seawater should, if it is the only process operating, produce strongly skewed distributions (Ohmoto 1992; Goodwin *et al.* 1976). Two different patterns are seen in the Manjeri Fm. results. There is a broad spread in the Spring Valley Member, with small peaks around -4.5 ‰ and -2.9 ‰, and a separate peak at $+3$ ‰. In the Jimmy Member, two peaks are present, a strong central peak around 0 ‰, and a peak with a possible skewed distribution at -5.9 ‰. If the 0 ‰ peak in the Jimmy Member is excluded, a possible explanation of the pattern is that it reflects sulphate reduction from relatively sulphate-rich seawater (at, say, $+5$ ‰ in the Late Archaean, but this is not well constrained), in a variety of settings from open to nearly closed system (see discussion of fractionation in Ohmoto (1992)). Specific units need to be considered separately, linking the carbon isotope evidence to the sulphur isotopic results.

(a) *Spring Valley Member*

In the Spring Valley Member (lower Manjeri Fm.), sedimentary facies indicate the host silts containing the sulphide-carbon stringers were laid down in shallow water (Hunter *et al.* 1998). The $\delta^{13}\text{C}$ record of rubisco is present (figure 1). Given the shallow-water facies and the

proximity of stromatolites, this strongly suggests local oxygen production by aerobic oxygenic photosynthesis. In the same rock suite, in associated sulphides, $\delta^{34}\text{S}$ shows a wide range from -19.9‰ to $+5.4\text{‰}$ (figure 2). This range probably records sulphate reduction in an open system, as would be expected in shallow-water sediments (see Ohmoto (1992) for a discussion). However, the marked asymmetry in the histogram that would be expected from a simple sulphate-reduction control is not present, so it is likely that other processes were present too. The rRNA phylogeny would imply that photosynthetic S-redox handling would be expected to have evolved prior to oxygenic photosynthesis. Samples with $\delta^{13}\text{C}$ ca. -18‰ are consistent with (though do not prove) the signature of anaerobic photosynthesis in the rock (e.g. green sulphur bacteria; see Pringault *et al.* 1998), and similarly the $\delta^{34}\text{S}$ results can be explained as the product of S-oxidation processes in photosynthetic S bacteria. This is permissive evidence, not proof, that a sulphuratum was present, with sulphate reduction being balanced by reoxidation of S species. The general supply of oxidation power in the Spring Valley facies would have been photosynthetic; both by anoxygenic S bacteria and by the creation of more oxygenated local water by oxygenic photosynthesis in cyanobacteria. Reduction power would have come from recycled organic matter and from microbially excreted products.

(b) *Jimmy Member*

Carbon isotopic compositions in the Jimmy Member show the record of rubisco, and possibly other C-fractionating metabolic enzymes. The Jimmy Member as a whole is very heterogeneous in S-isotopic values, even on a small scale. $\delta^{34}\text{S}$ in the drill core varies from -19.9‰ to $+16.7\text{‰}$, a wide range compared to other rock units in the Archaean (see Strauss & Moore 1992; Hayes *et al.* 1992). The $\delta^{13}\text{C}$ evidence for rubisco fractionation implies the working of aerobic or microaerobic bacteria, which in turn implies a supply of oxidation power: seawater sulphate. Reduction power would have come from organic matter and products, but perhaps also from fluids that had equilibrated with the rock in hydrothermal systems, or near volcanic vents, especially as the Jimmy Member is in close physical (and probably stratigraphic) proximity to overlying lavas.

In the Jimmy Member there is a sharp peak of $\delta^{34}\text{S}$ -values around 0‰ . These compositions could be primary depositional values, but could also record late diagenetic or later fluid movement and recrystallization (e.g. in deformation), which would isotopically overprint sulphide deposits. However, if the primary biological heterogeneity to positive and negative values had been on a small scale (greater than millimetres), a process as local as penecontemporaneous partial recrystallization would produce a massive peak near 0‰ . The Jimmy Member is in close spatial and temporal proximity to komatiitic basalt pillow lavas. REE data show strong hydrothermal influence in the Jimmy Member sediment, and hydrothermal influence is also present, though to a lesser extent, in the Spring Valley Member (see also Hunter *et al.* 1998). Ingress of contemporary hydrothermal fluids that contained mantle-derived S, during formation of the Jimmy Member deposit, would produce a homogeneous

and sharp peak around 0‰ . The Jimmy Member ecosystem may have been near a hydrothermal vent, or may have had distal hydrothermal input into a closed basin.

The wide range of the $\delta^{34}\text{S}$ cannot simply be explained by inorganic processes, but may be understood by considering the possible impact of microbial biota. If the sharp 0‰ peak is attributed to hydrothermal fluids ($n=47$, $\pm 0.09\text{‰}$), the histogram of the remaining results is asymmetrical about a peak at -5.9‰ . One possible explanation is that open-system microbial sulphate reduction from seawater sulphate (at, say, $+5\text{‰}$, but not well known) produced the peak at -5.9‰ , with a spread of $\delta^{34}\text{S}$ -values between -6 and $+5\text{‰}$, or possibly wider. However, the very wide range of values past these bounds may need further explanation.

The absence of metazoan disturbance means that redox distribution in the top few centimetres of Archaean sediments forming by deposition below the wave base would have been controlled only by microbial motility and diffusion, not bioturbation. Water flushing may have been less than today. This would lead to a nearly closed-system fractionation of the sulphur isotopic compositions. Downwards in the sediment, each layer would be overlain by more enriched sulphate. Closed-system sulphate reduction could ultimately lead to a strong enrichment in ^{34}S in residual sulphide, which could explain the very heavy $\delta^{34}\text{S}$. It is possible that rare strongly positive $\delta^{34}\text{S}$ in the Jimmy Member may record a nearly closed-system sulphide precipitation by sulphate-reducing bacteria (e.g. Alt & Shanks 1998), using organic residue from dead organisms (for example, from photosynthetic plankton in the overlying water). The spread of values to much lighter than -8‰ may simply be a product of sulphate reducers alone, but suggests a two-stage process, with reoxidation of early formed sulphide, say with $\delta^{34}\text{S}$ around -8‰ , and then further microbial reduction to the very light values. Thus range and symmetry of the distribution suggest that open-system sulphate reduction was not the only process operating: it is possible that the reverse process of sulphide oxidation also occurred. Overall, the wide isotopic range, both in carbon and sulphur, is probably biogenic: there is no plausible inorganic explanation.

The width of the range in sulphur isotopes thus does not prove but strongly implies the existence of a microbial community capable of diverse metabolic processes, cycling sulphur in a variety of fractionation steps. Moreover, this large range, though less than in modern sediment, and the shape of the distribution, strongly imply a complex ecology capable of multiple sulphate reduction and oxidation steps.

The setting of the Jimmy Member may intermittently have been roughly analogous to deposition of sulphides in the modern Black Sea. However, the analogy is not close, as anoxia in the Black Sea sediments is driven by surface productivity and sulphate input from the Mediterranean (Ross & Degens 1974), and volcanic sources are absent. The chemistry of the deep Archaean ocean was probably Fe-rich and therefore H_2S -poor (Drever 1974), but may have contained downwelling SO_4^{2-} (Ohmoto 1992, 1997). Reduced S species may have been supplied in part by hydrothermal fluids (REE evidence, Hunter *et al.* 1998), but a major source of S would have been seawater

sulphate, which was probably abundant in the overlying local near-surface seawater, with oxidation power possibly supplied locally by oxygen emissions from nearby cyanobacterial stromatolites. Cyanobacterial plankton (Nisbet 2000) may have been present in the upper levels of the water, providing both oxidant to the water and also a source of reduced organic matter to the bottom for any non-photosynthetic mat communities. Infall of organic carbon from the overlying ocean plankton possibly as blobs of organic matter settling after blooms would supply reduction power. The evidence for rubisco in the $\delta^{13}\text{C}$ -values of the Jimmy Member (which may have been below the photic zone) could either record unused detrital organic photosynthetic carbon or record the presence of aerobic sulphate-processing bacteria using the Calvin–Benson cycle, including linked oxidation–reduction sulphureta. Given the apparent fine scale of the isotopic heterogeneity, conventional methods, using large samples, would be unlikely to show the full $\delta^{34}\text{S}$ range in the Jimmy Member.

5. ARCHAEOAN ECOLOGY

The results show a much more fractionated $\delta^{34}\text{S}$ record than previously recorded from the Late Archaeon, implying that a diverse S ecology did indeed exist, at least by this stage. The new evidence here is most simply interpreted if the full microbial S-cycle was present. This does not prove or disprove either the ‘standard’ model of molecular evolution or that of the history of atmospheric oxygen. It is, however, compatible with the notion that a flux of oxidized sulphur species (mainly probably sulphate) was available in late Archaeon shallow to medium-depth sea-floor environments.

The results are thus most simply interpreted as evidence for the existence of diverse microbial ecology in the Belingwe Belt, and that by this time a diverse microbial community existed (Nisbet 2000), with the full microbial S-cycle in operation, though on a finer scale than in Phanerozoic muds bioturbated by metazoa. The Belingwe samples are from rocks that are arguably among the best-preserved of any in the Archaeon geological record, with a very low-grade metamorphic and structural history. Thus it is not surprising that they show isotopic preservation that has not been observed in other material. Nevertheless, the range of isotopic heterogeneity that remains in the Belingwe sediments is remarkable—explicable only as a record of biological activity and diversity.

The range of $\delta^{34}\text{S}$ results reported here, implying evolutionary diversity, suggests that a variety of microbial processes occurred in other settings also, in silts and muds of the shallow photic zone, and possibly also in subphotic conditions. Rasmussen (2000) has reported evidence for life around deep-water hydrothermal systems 3.2 Ga ago, in a setting perhaps not greatly different from the Jimmy Member, except perhaps that the Jimmy Member facies may be somewhat shallower. The wide ranges and distributions of carbon and sulphur isotopic compositions reported here from the shallow-water environment (Spring Valley Member) support a model of small-scale but productive microbial mats growing on a shallow sea floor under a comparatively oxidized surface-water body, with local oxygen production from cyano-

bacteria. Iron-rich horizons may have record microbial blooms fed by inflows of more reduced waters. Photosynthetic oxidation may have occurred in this environment. Such complex communities, based on both photosynthesis and sulphur cycling in sulphureta involving both reduction and oxidation, would have been capable of producing both the wide ranges of isotope values reported here. Whether aerobic sulphur oxidation in somewhat deeper water took place also is unproven, but is suggested by the $\delta^{34}\text{S}$ range and $\delta^{13}\text{C}$ evidence for rubisco in the Jimmy Member sulphide–carbon–chert horizon, which was probably deposited close to active hydrothermal vents below the photic zone. Similarly, while not proving or disproving the competing models of atmospheric evolution, our results do imply that oxidation power was plentifully available at least in waters in these settings close to abundant oxygenic photosynthesizers.

We thank J. F. Wilson and T. Blenkinsop for their support. Tony and Charlotte Rauch of Manjeri Ranch and Zimasco Zimbabwe made the work possible. Nick Butterfield is thanked for very useful criticism (and helpful disagreement). Three anonymous reviewers made very helpful comments. This work was supported at varying stages by the University of Zimbabwe, Natural Sciences and Engineering Research Council Canada, Natural Environment Research Council, and particularly by the Leverhulme Trust.

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