EARTH'S OLDEST (~ 3.5 Ga) FOSSILS AND THE 'EARLY EDEN HYPOTHESIS': QUESTIONING THE EVIDENCE

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Abstract. We question the biogenicity of putative bacterial and cyanobacterial 'microfossils' from 3465 Ma Apex cherts of the Warrawoona Group in Western Australia. They are challenged on the basis of integrated multidisciplinary evidence obtained from field and fabric mapping plus new high-resolution research into their context, sedimentology, filament morphology, 'septation' and arrangement. They cannot be distinguished from (and are reinterpreted as) secondary artefacts of amorphous carbon that formed during devitrification of successive generations of carbonaceous hydrothermal dyke vein quartz. Similar structures occur within associated carbonaceous volcanic glass. The null hypothesis of an abiotic or prebiotic origin for such ancient carbonaceous matter is sustained until mutually supporting contextural, morphological and geochemical evidence for a bacterial rather than abiotic origin is forthcoming.

Keywords: Archaean, carbon isotopes, early Earth, fossils, hydrothermal, photosynthesis

1. The Early Eden Hypothesis

For more than two decades, it has been widely assumed that life arose extremely early on the surface of planet Earth, and not later than 3.5 Ga. This line of reasoning, here called the 'Early Eden hypothesis', has been based upon three main lines of argument from the rock record: stromatolites, morphological microfossils, and carbon isotopes (e.g., Hofmann *et al.*, 1999; Schopf and Packer, 1987; Schopf, 1992; Mojsis *et al.*, 1996). This Early Eden hypothesis has provided a stimulating launch pad for astrobiology, encouraging the search for early life on Mars (e.g. McKay *et al.*, 1996) and beyond. With several missions now poised to explore the prebiotic-biotic boundary across the solar system, however, it becomes important to scrutinise the status of the hypothesis.

The keystone of the Early Eden hypothesis centres upon what has, until recently, been accepted as the earliest reliable remains of fossils on Earth, preserved within the ~3460 million year old Apex chert of Western Australia. Schopf (1999) compared some of these putative fossils with a bacterial group still alive today, known as the cyanobacteria. The first appearance of cyanobacteria in the fossil record is a matter of great scientific interest because they gave rise to the oxygen-

rich atmosphere that allowed the evolution of higher life forms, including man. Such diversity is not matched in the fossil record for another 1600 Ma. The Apex microfossil-like structures have been classified into eleven different types of fossil bacteria (Schopf, 1992, 1993). This work came to great prominence during the debate over the so-called microfossils from Mars in 1996 (McKay *et al.*, 1996), when Schopf was among the first to question their authenticity, contrasting them with his own supposedly reliable microfossils from Australia (Schopf, 1999).

But is it possible that the evidence provides an alternative interpretation? To test this, an international group of scientists have been undertaking a study on the early geological record of life to explore key assumptions of the Early Eden hypothesis using traditional and advanced techniques at a range of scales from field mapping to high-resolution analytical microscopy and geochemical analysis. For our first report (Brasier *et al.*, 2002) we have re-examined the now famous Schopf fossil site and his prepared materials (15 sections ranging in thickness from 193–380 μm) and have come to some surprising conclusions. We find that the supposed beach-like deposit from which his earliest fossils come have features consistent with a submarine volcanic fissure and hot spring. We observed that the fossil-like structures are present in late stage rock fabrics and volcanic glass, calling their biological origin into question. New, computer-assisted imaging techniques show that many of the Apex chert 'holotypes' are considerably more chaotic than shown in publications and that they intergraded with abiogenic morphologies. We therefore propose that 'Earth's oldest fossils' are artefacts (pseudofossils) formed when carbonaceous glass associated with one of Earth's oldest hot springs recrystallized into spherulitic silica (Brasier *et al.*, 2002). These conclusions have been followed by a flurry of important papers, published during the 'annus mirabilis' of astrobiology (Dalton, 2002; Fedo and Whitehouse, 2002; van Zuilen *et al.*, 2002; Shoenberg *et al.*, 2002). Each of these raises serious questions about hidden assumptions embedded within the Early Eden paradigm, as we discuss below.

2. What lines of reasoning?

The Early Eden hypothesis is largely built upon a kind of logic known to philosophers of science as 'deductive reasoning'. The deductive argument runs as follows: if it looks like a cyanobacterium, is septate like a cyanobacterium, and is made of the same material as a cyanobacterium, then the most parsimonious explanation is that it is a cyanobacterium (cf. Schopf, 1999). In other words, we should look for something 'complex' in the ancient rock record and then find something comparable in the later rock record or living world with which it can be grouped by a series of simple descriptive attributes. Outwardly, this seems reasonable. It is a line of reasoning well trammelled by naturalists since Sir Francis Bacon in the early seventeenth century, and has been used without question and with considerable success in relation to the last 500 Ma of Earth history. But it can also lead to

embarrassing mistakes, particularly in the developmental stages of a discipline, when the bounds of our understanding are few: for example, the claims for canal systems ('Lowellian Mars') or human faces on Mars (see Chambers, 1999) and (one of our favourites) for a planet Earth entirely constructed of giant protozoan foraminiferid tests (the 'nummulosphere' of Kirkpatrick (1916)).

Palaeobiologists have been well aware of these problems, of course. Quite often, there has simply not been enough convincing morphological evidence to allow attribution of a structure to a particular class of living or fossil organism. One way round this, therefore, has been to establish a series of biogenicity criteria and to rank the structures as fossils, dubiofossils or pseudofossils according to their score (e.g., Hofmann, 1972; Buick, 1990). This 'dubiofossil' approach has been widely followed over the last three decades, perhaps because it seems to have all the virtues of a democratic compromise: middle of the road, tolerant, cautious in decision-making. But we are concerned that the dubiofossil concept may encourage fuzzy thinking in an area (the prebiotic-biotic boundary) where this now urgently needs to be avoided.

Such deductive reasoning is a risky form of logic when looking for the earliest signs of the simplest kinds of life. We tend to agree with Karl Popper (see Medawar, 1982) that scientific hypothesis may better be tested by means of falsification rather than by justification. To that end, we suggest that Earth's early fossil record should be treated with the caution appropriate to Mars, and urge that future studies explore the following null hypothesis:

> that very ancient/alien microfossil-like structures (or stromatolites or geo-chemical and isotopic signals older than c. 3.0 Ga) should not be accepted as being of biological origin until possibilities of their non-biological original have been tested and can be falsified (see Brasier *et al.*, 2002).

This null hypothesis is germane if, as we suspect, current approaches may be inadequate to resolve the prebiotic-biotic boundary without dispute in the early geological record. New data and techniques (Brasier *et al.*, 2002) are now calling into question many assumptions embedded within the previous 'Apex microfossil' hypothesis of Schopf and Packer (1987) and Schopf (1992, 1993, 1999). Sophistic-ated research tools (such as trace elements, isotopes, molecular organic chemistry) will be needed to map out the context and syngenicity of supposedly 'biogenic' Archaean structures, with due regard to the ambiguity of interpretation from a single technique (cf. Schopf *et al.*, 2002a), and to the need for multiple lines of evidence.

2.1. WHAT IS THE CONTEXT?

Context is the key to decoding the signals for early life on Earth or Mars. Does the context point to a potentially viable habitat for life? For the 3.5 to 3.4 Ga old Warrawoona Group of Western Australia, for example, it has for long been accepted that the layered siliceous cherts were comparable with in part modern

evaporitic and exposed lagoonal environments (Dunlop *et al.*, 1978; Walter *et al.*, 1980; Lowe, 1983; Awramik *et al.*, 1983). While Lowe (2003, and in Tenenbaum, 2003) has argued that some South African dykes are neptunean in origin, our research does not support this interpretation for the Apex dykes of Australia. Several programs of careful mapping have revealed that the environmental setting of these rocks was largely volcanic, volcaniclastic and hydrothermal in nature (e.g., Nijman *et al.*, 1998; van Kranendonk, 2001; Nakashima *et al.*, 2002), a conclusion wholly supported by our mapping, fabric analysis and geochemical studies (Brasier *et al.*, 2002). This raises the question as to whether any of the Warrawoona sediments were truly normal marine sediments and precipitates? Could it be that the so-called 'evaporites' were actually formed in relation to supersaturated situations around hydrothermal vents? If so, then it is possible that the subsurface Schopf site may not have been viable for cyanobacterial life (Brasier *et al.*, 2002).

A comparable lack of contextural analyses has unseated what may be premature claims for earlier life signatures in Greenland. Here, the ∼3.85 Ga old Akilia rocks have been held to reveal carbon isotopic signatures consistent with life, in rocks widely assumed to have been banded iron formation (BIF) sediments (Mojsis *et al.*, 1996). Recent mapping and fabric analysis reveal, however, that the rock may not be BIFs at all but highly metamorphosed ultrabasic igneous rocks, seriously questioning the validity of those claims (Fedo and Whitehouse, 2002). At Isua, negative carbon isotopic signatures in graphite of presumed biogenic origin (Mojsis *et al.*, 1996) is now argued to originate from the reduction of metasomatic iron carbonate and they are unlikely, therefore, to be biogenic (van Zuilen, 2002). In the western part of the Isua belt, graphitic grains with negative carbon isotopic values occur within putative sediments that have been explained as due to the activities of photoautotrophic phytoplankton (Rosing, 1999). It emerges, however, that the same sediments contain tungsten isotopic values consistent with a major, early, meteoritic contribution to the sediments, which means that an extraterrestrial, carbonaceous chondrite source for the negative isotopes cannot yet be rejected (Schoenberg, 2002).

2.2. ROUNDED CLASTS?

It has been claimed that the 'Microfossils of the Apex Basalt occur within clasts that were deposited in this unit prior its lithification' (Schopf and Packer, 1987). More specifically, it has been argued that the presence of 'microfossils in a petrographically distinctive population of clasts and their absence from all other clasts or the surrounding matrix indicate that the filaments predate deposition of the chert unit and were initially preserved in older rocks, some part of which was eroded, transported and redeposited as a detrital component of the bedded chert' (Schopf, 1993). Careful study of the generational histories of the rock, however, has enabled us to falsify this claim. We have found that the majority does not occur in first generation fabrics at all, nor in rounded clasts, but in mainly subangular

breccia fragments of second and third generation origin, as well as in cements after hydrothermal microquartz chalcedony (Brasier *et al.*, 2002). We can also confirm that pseudoseptate microfossil-like structures of carbonaceous composition (in Raman) occur, without question, in associated, devitrified volcanic glass shards and cements (Brasier *et al.*, 2002, Figures 2e, k, l), i.e., in settings that are arguably incompatible with the preservation of primary biological morphologies.

2.3. STROMATOLITE-LIKE CLASTS?

Are 'stromatolite-like clasts' present at the Schopf site and could they be of biogenic origin? Such association of putative Archaean microfossils with stromatolitic laminae has sometimes been regarded as a criterion for biogenicity, and 'stromatolite-like' laminae have indeed been reported from the Schopf site (Schopf, 1993). The biogenicity of stromatolitic structures cannot be assumed, however (Buick *et al.*, 1981; Buick, 1990; Grotzinger and Rothman, 1996) and abiogenic origins will need to be falsified. Our observations indicate that the so-called 'stromatolite-like' structure from the Apex chert (Schopf, 1993, Figure 3c) is intimately intergrown with fissure-filling fabrics of a second generation (A2) and indistinguishable from the associated layered microquartz void fills (cf. geyerite silica) of generation B2. Hence the biogenicity of this 'stromataloid' structure cannot be assumed and from our interpretation seems questionable.

2.4. BACTERIAL BEHAVIOUR?

It is interesting to note that the Apex chert microfossil-like structures 'are irregularly distributed and randomly orientated solitary filaments' (Schopf, 1993), since this is one observation confirmed by us during the study. This random pattern contrasts with the orientated and clumped distributions of the oldest *bona fide* Precambrian (Gunflint chert) microfossil assemblages. Of course, bacteria can grow without forming patterns, but it means that the Apex chert structures fail another of the biogenicity criteria put forward by Schopf and Walter (1983).

2.5. SIMPLE AND UNBRANCHED FILAMENTS?

Are the filaments really simple and unbranched, as claimed (Schopf and Packer, 1987; Schopf, 1992, 1993, 1999)? Here again, our answer is negative. Many of the holotypes are shown to have branches not illustrated or mentioned in the type descriptions (see Brasier *et al.* (2002), in which the term 'branched' is here used to describe any feature of variable diameter, visibly connected to the main structure and arising in some way from its main axis. Our recognition of 'branching' is based on light microscope observations supplemented with montage analysis. The Automontage technique used by us to illustrate this branching (Brasier *et al.*, 2002) is a sophisticated computer merging of digital images, upgrading the traditional

(cut and paste of single focal plane monochrome images) manual montage technique used by Schopf (1992, 1993) to display 3D structures in 2D format. We note that Bonnie Packer (of Schopf and Packer, 1987) concurs with our use of the term 'branching' and that Schopf may originally have used this term himself (Dalton, 2002). Schopf has stated, however, that some of the structures are not actually branched but 'folded' or 'overlapping' (Dalton, 2002). If this is accepted, it compounds the problem – for what once had seemed to be the 'end' of a filament must be reinterpreted either as something disrupted or from the middle. This then calls into question the original criterion of 'terminal cell shape' required for the diagnosis of the eleven taxa of microfossils (Schopf, 1992, 1993). We do not accept that they are 'folded', however, since the branches typically show dramatic changes in diameter when the plane of focus is changed.

2.6. SEPTATION, BIFURCATION AND 'COMPLEXITY'?

Both 'septation' and 'bifurcated cells' have been advanced as evidence in favour of the biogenicity of the Apex filaments (Schopf, 1992, 1993). We find, however, that these features are not readily distinguishable from similar features found in associated spherulitic artefacts and crystal rims (Brasier *et al.*, 2002). Nor does the septation result in hollow structures, taken by Buick as a key criterion for biogenicity (1990; see below; the structures are typically solid). Hence, a mineralised origin for the supposedly biological septation and bifurcation cannot be falsified using the present evidence.

In isolation, complex carbonaceous filaments cannot be taken, as an indicator of biogenic processes – there is a very extensive literature on this. Nanotubes are produced abiotically by vapour deposition from Ni/Co enriched carbon (Rao *et al.*, 1997; see also Harris, 2001), while complex organic microfilaments have been formed by co-precipitation of barium salts and silica (Hyde *et al.*, 2002); by Miller-Urey discharge (Fulsome *et al.*, 1975), by Fischer-Tropsch-like synthesis on metallic grains (Baker and Harris, 1978), by evaporation of sessile droplets (Deegan, 2000) and, of course, by diagenetic and metamorphic formation around spherulites and other crystals (Horodyski, 1981; Brasier *et al.*, 2002; Parnell, 2003). The abiogenic formation of complex structures by simple processes is well attested as a general phenomenon (see Prigogine and Stengers, 1984; Kauffman, 1996). We would argue, therefore, that 'complexity' should no longer be used as a guide to biogenicity without regard to the null hypothesis stated above (*pace* Buick, 1990; Schopf, 1999).

2.7. GRAPHITE OR KEROGEN?

Are the Apex structures composed of graphite (Brasier *et al.*, 2002) or of kerogen (Schopf *et al.*, 2002a, b) according to Raman spectroscopy? Kerogens comprise a mixture of molecules derived from the breakdown of once-living organisms and consist of many different, typically aromatic (i.e., benzene ring-containing)

C-O-H molecules (Wopenka and Pasteris, 1993). While such molecules can be classified as 'organic' in the traditional chemical sense it is important to note that 'organic' chemistry does not imply a biogenic origin (Pasteris and Wopenka, 2002a, b). Raman spectra of kerogens show only the 'generic spectral features' indicative of discontinuous arrays of condensed benzene rings. According to Pasteris and Wopenka (2002a): 'Most prominent are two very wide, first-order Raman bands that peak at ~1360 and ~1600 Dcm-1. These bands reflect only the most fundamental level of the molecular structure of kerogen and are not specific to individual organic molecules. Nor do typical kerogen spectra show any other bands characteristic of additional chemical functional groups. Further confounding the definitive identification of kerogen in geologic materials is the fact that poorly ordered carbonaceous materials also can arise through non-biological processes, such as heating *in situ* of organic or inorganic compounds, metamorphic mobilization of pre-existing carbon compounds, and high-temperature precipitation from hydrothermal solutions. Unfortunately, most of those abiogenetically produced carbonaceous materials have Raman spectra that are indistinguishable from those of kerogens. Thus, a Raman spectrum cannot definitively identify "kerogen", but only "disordered carbonaceous material" '. . . . 'Thus, the recent declaration by Schopf *et al.* (2002), that Raman spectroscopy can identify a kerogenous signature (and, thus, a signature of past life), is erroneous'. Furthermore, Pasteris and Wopenka (2002b) stated: 'we contend that the Raman spectra of Schopf *et al.* (2002) indicate that these are disordered carbonaceous materials of indeterminate origin. We maintain that Raman spectroscopy cannot be used to identify microfossils unambiguously, although it is a useful technique for pinpointing promising microscopic entities for further investigation'.

Our work (Brasier *et al.*, 2002) reveals identical graphitic/kerogen signals within the 'microfossils' and in other spherulitic artefacts, crystal margins and matrix. Raman spectra, when used alone and without control studies (e.g., Schopf *et al.*, 2002a), cannot therefore be taken to imply or exclude biogenicity (Brasier *et al.*, 2002). Further work is needed on the H:C ratio and elemental content within the 'microfossils' and matrix using techniques such as pyrolysis gas chromatography-mass spectrometry and TEM with EDS and EELS (e.g., De Gregorio and Sharp, 2003).

2.8. CARBON ISOTOPES AND PHOTOAUTOTROPHY?

Negative isotopic values between c. –26 and –30‰ appear typical for the Apex chert (Strauss and Moore, 1992; Brasier *et al.*, 2002). These are a little more positive than some values obtained from older and younger cherts of the Pilbara (<-34‰, Strauss and Moore, 1992) and lie (just) within the range for oxygenic photosynthesis (Schopf, 1999). But they also span the range known from methogenic metabolism (cf. Schidlowski and Aharon, 1992). Abiogenic synthesis of isotopically light hydrocarbons is known in the laboratory (Horita and Berndt,

1999), from modern mid-ocean ridge systems (Holm and Charlou, 2001), from Archaean crystalline rocks (Sherwood Lollar *et al.*, 2002; van Zuilen *et al.*, 2002), and from meteorites (Schoenberg *et al.*, 2002). Clearly, therefore, carbon isotopes alone cannot demonstrate biogenicity of the organic matter in the Apex cherts nor can a Fischer-Tropsch type synthesis of carbon compounds yet be rejected (Brasier *et al.*, 2002) as a null hypothesis. There is clearly an urgent need for observational and experimental work to test the potential sources for ^{13}C-depleted carbonareous matter on the early Earth.

2.9. DEGRADED COCCOID COLONIES?

Kazmierczak and Kremer (2002) have argued that structures in Silurian (c. 350 Ma-old) cherts contain filaments interpreted as the degraded remnants of a mono-specific assemblage of cyanobacteria-like coccoidal cell colonies that lived in a hydrothermal setting, and have gone on to speculate that the Apex microfossil-like structures could have formed in a similar way. That thermal alteration of Silurian carbonaceous matter might produce microfossil-like artefacts that are curved seg-mented and branched is interesting. It is, however, a leap of faith to infer that the Apex structures arose from the degradation of large, cyanobacteria-like coccoidal cell colonies for which there is no convincing evidence in the fossil record for a fur-ther 1600 Ma. Nor do we find that the Silurian structures are comparable with the majority of the Apex structures. When carbonaceous reaction rims form along the spherulitic fronts of chalcedonic silica during devitrification they leave a continuum of structures ranging from a central clotted mass often filled with spherulites of radial fibrous origin (Figure 1a), through complex dendritic forms (Figure 1c) to isolated arcuate forms (Figure 1b) of widely varying diameter. This is how the majority of the Apex pseudofossils, including the holotype of *Archaeoscillatoripsis maxima* (Figure 1a, arrowed), can be explained most parsimoniously. It can be contrasted with the more-or-less fixed diameter and presumably non-fibrous origin of degraded coccoid cell colonies. When carbonaceous reaction rims accumulate along the rims of growing hydrothermal barite crystals, they form a galaxy of struc-tures that range from castellated sheets to discrete angular filaments (Figure 1e) that intergraded with the adjacent holotype of *Eoleptonema apex* (Brasier *et al.*, 2002; Figure 3).

The absence of convincing evidence for entire coccoidal cell colonies in the Apex cherts (or from other early Archaean rocks), and the lack of the predictive power provided by the degraded coccoid hypothesis (Kazmierczak and Kremer, 2002), cannot explain parsimoniously (as can our reaction rim hypothesis), the as-sociation of structures with deep hydrothermal dykes. Neither can it explain related structures and fabrics from felsic volcanics and volcanic glass, botryoidal cements, estimated palaeotemperatures of 250–350 °C, intergradations of 'filaments' with spherulitic artefacts (i.e., with radial fibrous origin), and the association with highly reduced (rather than oxidised) mineral species (Brasier *et al.*, 2002). We do not
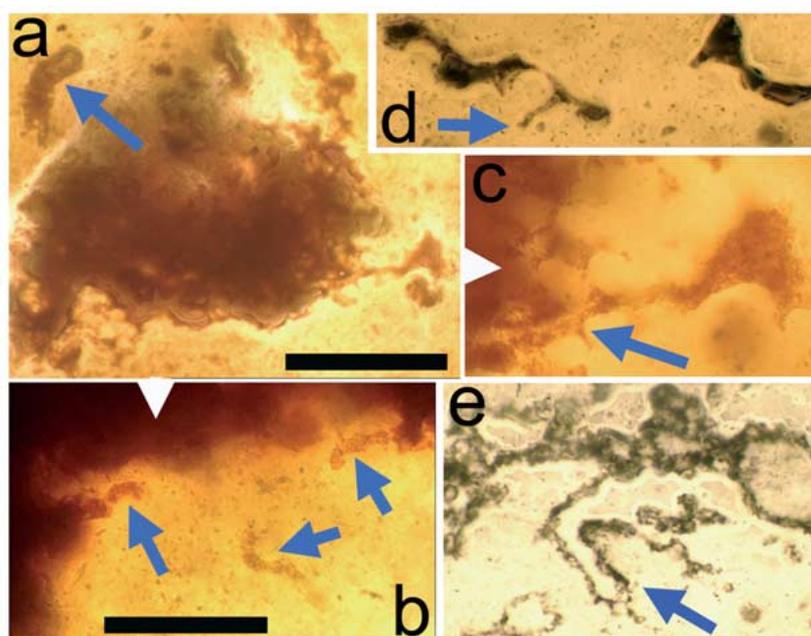
Figure 1. How the Apex chert 'microfossils' were formed. (a–c) Devitrification of hydrothermal carbonaceous glassy silica to spherulitic chalcedonic quartz produces relict clumps of carbon containing clear spherulites (a, c) that typically have fringes of septate dendritic (c) to arcuate (b) filamentous pseudofossils, formed as reaction rims. (d) Abiogenic septate carbonaceous filament shown branching off spherulitic mass. (e) Growth of hydrothermal barite produces castellated pseudofossils. The colours (red, brown, black) here relate to levels of secondary weathering and oxidation of associated iron compounds. ((a–c) NHM V 63164; (d) 63166, (e) 63729). Scale bar: 100 μm (a); 40 μm (b–e).

exclude the exciting possibility that the Apex chert structures could have formed from the degradation of hyperthermorphilic bacterial carbon during devitrification of hydrothermal silica (Brasier *et al.*, 2002). Adherence to the null hypothesis, however, requires that an abiogenic origin be falsified, and this has not yet been achieved.

3. Conclusions

We have found that the evidence for the Apex 'microfossils' (Schopf, 1992, 1993, 1999) can be falsified at a range of scales (Brasier *et al.*, 2002) and believe that the Earth's early fossil record should henceforth be treated with the same care and respect as Martian meteorite samples. In our view, future studies should address the above stated null hypothesis. Attempts to solve the dilemma of the Apex fossils by means of appeals to unconstrained morphological comparisons (Kazmierczak and Kremer, 2002) or to interpretation of data from a single technique such as Raman (Schopf *et al.*, 2002a) are questionable.

Any future attempt at a 'revised Apex Microfossil' hypothesis appears to be left with a series of apparently *ad hoc* reinterpretations of greatly reduced predictive power: 'microfossils' of unknown original morphology, uncertain biology, some unexpectedly large ($<36~\mu$m), no longer meeting established criteria for biogenicity, lying within successive hydrothermal fissure fillings and cements. Key aspects of our 'hydrothermal carbon' hypothesis for the Apex chert 'microfossil' site at Chinaman Creek, hitherto unchallenged, include:

- the subsurface, metalliferous hydrothermal setting (making unrealistic the possibility of a photosynthetic, cyanobacterial origin),
- the association of 'microfossils' with late stage fissure-filling fabrics (meaning that the majority of figured 'microfossils' are not syngenetic with the earliest 'clasts' but with later stages of fissure-filling and quartz intrusion, contravening a basic criterion for biogenicity; see Schopf and Walter, 1983),
- the association of microfossils with evidence for metastable mineral phases such as chalcedonic silica (which can destroy original morphologies and produce new artefacts of similar composition, again contravening a basic criterion for biogenicity; see Schopf and Walter, 1983),
- the lack of 'behavioural' distribution patterns of the microfossils consistent with biogenicity (cf. Schopf and Walter, 1983).

We emphasize that graphitic or simple kerogenous composition cannot be readily distinguished using Raman spectra alone, nor can either be taken to imply or exclude biogenicity. The same is true for carbon isotopes. These observations, as well as new data relating to inferred temperatures and the possibilities of Fischer-Tropsch-type synthesis will be discussed in later papers. We conclude that the null hypothesis of Brasier *et al.* (2002) is sustained and that the supposedly biogenic structures in the Apex chert at Chinaman Creek no longer provide evidence for a morphologically diverse biosphere, including photoautotrophs, as early as c. 3.5 Ga.

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