Hot spring sinters: keys to understanding Earth's earliest life forms[1]

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Abstract: The question of what composed the Earth's oldest fossils is the subject of current debate. At present, taphonomical determination of Archean silicified microfossils is largely based on morphological comparisons with extant microorganisms. This method has significant shortcomings because little is known about which types of bacteria silicify, what physical changes are induced on those species during mineralization, and, most importantly, what their preservation potential is. Terrestrial hot springs may help resolve these uncertainties because the silica-supersaturated geothermal fluids mineralize a wide variety of natural microbial communities and thus lead to the formation of numerous distinct biofacies. Some of these biofacies are reminiscent of Archean siliceous stromatolites from which the oldest microfossils were recovered. We suggest that by integrating molecular techniques that characterize the indigenous microbial populations growing in different biofacies with electron microscopy, we may be able to assess better what types of ancient microbes could have become fossilized.

Résumé : La question à savoir quels sont les plus vieux fossiles de la Terre est un sujet de débat actuel. Pour le moment, la détermination taphonomique des microfossiles silicifiés de l'Archéen est grandement basée sur des comparaisons morphologiques avec des micro-organismes existants. Cette méthode comporte des lacunes importantes car l'on en connaît peu sur les bactéries susceptibles de silicification, quels changements physiques sont induits sur ces espèces durant la minéralisation et, encore plus important, quel est leur potentiel de préservation. Les sources thermales terrestres peuvent aider à résoudre ces incertitudes car les fluides géothermiques sursaturés en silice minéralisent une grande variété de communautés microbiennes naturelles et forment ainsi de nombreux biofaciès distincts. Quelques-uns de ces biofaciès font penser aux stromatolites siliceux datant de l'Archéen desquels les plus anciens microfossiles ont été récupérés. Nous suggérons qu'en intégrant des techniques moléculaires, qui caractérisent les populations microbiennes indigènes croissant dans divers biofaciès, à la microscopie électronique, nous serons en mesure de mieux évaluer quels types d'anciens microbes pourraient avoir été fossilisés.

[Traduit par la Rédaction]

Introduction

Much of what we understand about the existence of early life forms comes from the examination of siliceous microfossils that have been recovered from Archean strata. Structures resembling bacteria from 3.5-billion-year-old Apex cherts of the Warrawoona Group in Western Australia have, until most recently, been deemed the oldest morphological evidence for life on Earth (Schopf 1993). Their biological origin was inferred from their carbonaceous (kerogenous) composition, by the degree of regularity of cell shape and dimensions, and by their morphological similarity to extant filamentous prokaryotes (Schopf 1994; Schopf et al. 2002). Some of the Apex specimens exhibit features reminiscent of unbranched, partitioned trichomes, which not only implied that the Archean "microbes" were capable of gliding and possibly phototactic motility,

but that cyanobacteria may already have been in existence at that time (e.g., Awramik 1992; Schopf 1993). Further support for this conclusion came with the discovery of large spheroidal, sheath-like structures (up to 20 mm in diameter) in cherts from the underlying Towers Formation (Schopf and Packer 1987). An alternate view is provided by Walter et al. (1972) who suggested that some Archean microfossils may instead be likened to filamentous, anoxygenic photosynthetic bacteria, e.g., *Chloroflexus*.

A reexamination of the Apex chert by Brasier et al. (2002) has, however, called into question the biogenicity of the filamentous structures and the sedimentary origins of the earliest "fossiliferous" deposits. Instead, they suggested that the structures are probably secondary artefacts formed from Fischer-Tropsch-type reactions associated with sea-floor hydrothermal systems. Nonetheless, they did not discount

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[1]This article is one of a selection of papers published in this Special Issue on *Sedimentology of hot spring systems*.
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the possibility that the structures could be the remains of poorly preserved thermophilic bacteria. The discovery of pyritic, thread-like filaments in 3.2 Ga volcanogenic massive sulphides from the Pilbara Craton of Australia may corroborate the latter view because they seemingly provide evidence that chemolithoautotrophic thermophiles lived in or around hydrothermal systems at that time (Rasmussen 2000).

Irrespective of whether or not the Apex cherts actually contain fossils, the reported observation of putative filamentous and coccoid microstructures in a number of other rocks of relatively similar age (e.g., the 3.3 Ga old Kromberg Formation, South Africa) that are distinctively laminated and probably of stromatolitic origin (Walsh and Lowe 1985; Walsh 1992; Westall et al. 2001), along with geochemical analyses of carbonaceous residues and biogenic minerals (e.g., Rosing 1999; Shen et al. 2001), indicate that life was present at that time. What type of prokaryote those fossils represent remains unanswered. Metamorphism and deformation of the host rocks have obscured most of their microstructural features and modern analytical techniques used to examine biochemical features cannot differentiate among a diverse suite of micro-organisms. For example, laser-Raman spectroscopic imagery has recently been used to determine the presence of kerogen in samples as small as 1 mm, but it could not ascribe the signal to a particular species, nor could it verify that the organic carbon was of primary origin (e.g., Kudryavtsev et al. 2001). Similarly, ion microprobe analyses can yield elemental and isotopic ratios for microscopic samples (e.g., House et al. 2000), but the $\delta^{13}C$ values of biological materials overlap for a number of dissimilar genera (i.e., *Chloroflexus* versus cyanobacteria, Schidlowski et al. 1983). Therefore, even if the biogenicity of a "fossiliferous" structure can be reasonably ascertained, its taxonomical identification still relies on comparison of the morphological characteristics of the Precambrian microfossil with extant specimens (Buick 1990).

Unfortunately, our current approach to comparing extant microorganisms with Archean microfossils on morphological grounds has significant shortcomings. At present, there are insufficient data on which types of bacteria silicify, what physical changes are induced on those species during mineralization, and, most importantly, what their preservation potential is. These questions are directly relevant to Archean microfossil taphonomy because the ancient rock record is biased towards simple filamentous or coccoid morphologies that appear to have grown as microbial mats in solute-rich waters (Horodyski et al. 1992; Walter et al. 1992). This implies that entire populations of species with different morphologies and growth strategies, or those mats formed in dilute waters, had little chance of being preserved. The lack of such examples as microfossils could be due to differences in the susceptibility of various types of microorganisms to preservation, in the mechanisms of preservation, and (or) in the post-fossilization alteration processes (Reysenbach and Cady 2001). Furthermore, the microfossils typically represent the remains of cell sheath and wall material preserved in chert (Knoll 1985). The finer details of the cytoplasmic components have long been degraded and lost from the fossil record, and, as such, definitive comparisons with extant species invariably leads to controversy. Schultze-Lam et al. (1995), for instance, examined extant microbial mats formed by *Chloroflexus* that had grown around some Icelandic hot springs. As silicification proceeded, details

of the cytoplasm and the cell wall structure were progressively destroyed. The loss of those features meant that the modern silicified microbe could easily be identified as a cyanobacterium.

A number of experimental studies have tried to determine the physical changes induced on various bacteria during silicification (e.g., Oehler and Schopf 1971; Oehler 1976; Francis et al. 1978; Ferris et al. 1988; Birnbaum et al. 1989; Westall et al. 1995; Westall 1997; Toporski et al. 2002). Those studies showed that species-specific patterns of silicification exist and that different microbes are capable of being silicified with different degrees of fidelity. Nonetheless, only a few bacteria have been analysed, and in each study, different experimental conditions were used. As a consequence not only do the different studies yield conflicting results regarding the levels of silicification (e.g., Toporski et al. 2002), but no comprehensive database is presently available with which to confidently assess the preservation potential of a wide range of taxa.

To proceed further, two different approaches need to be taken. First, a systematic study could be employed whereby a large number of bacterial species are silicified and artificially aged under identical experimental conditions. Although such a study would provide general insights regarding the types of microbes that might fossilize, it would suffer from a lack of context because the results would not take into account the dynamics and interactions of mixed species growing in a mat community. Furthermore, such an approach cannot exactly replicate all of the variability that exists in natural environments where microbial silicification takes place.

A second approach, to which we subscribe here, is to study the silicification of natural microbial mat communities in a modern environment that is in some ways analogous to the Archean environment in which the ancient microbes grew. Modern terrestrial hot springs are ideal sites for studying biomineralization processes because the hot, reducing, and often silica-supersaturated, geothermal fluids provide conditions that are considered reasonably similar to those that existed in Archean oceans (Siever 1992; Nisbet and Sleep 2001). Recent ribosomal RNA (rRNA) analyses of nonmineralized mats also indicate that there is tremendous metabolic diversity in geothermal settings, from thermophiles to anoxygenic photosynthesizers to cyanobacteria (e.g., Pace 1997). This means that all indigenous microbes are subject to the same preservational conditions. In contrast, artificial fossilization studies are limited by the prescribed experimental parameters. Furthermore, many hot spring taxa compensate for mineral encrustation and move (or grow) in the direction of accretion faster than the rate of sedimentation. That, in turn, leads to characteristic biosedimentary facies, some of which have analogues in Precambrian fossil-bearing, siliceous stromatolites (Walter 1994). Therefore, in this paper, we outline an alternative approach to studying hot spring silicification that may eventually provide a better framework with which to critically determine the relevance of some ancient biological signatures.

Microbial silicification

Extensive deposits of opaline silica and (or) $CaCO_3$ precipitates commonly develop on the discharge aprons around hot springs and geysers. In neutral and alkaline waters, the

precipitation of opaline silica typically leads to the development of thick successions of sinters that contain a broad array of well-preserved, silicified microbes (e.g., Cady and Farmer 1996; Jones et al. 1997, 1998, 2001; Konhauser and Ferris 1996; Konhauser et al. 2001). Calcareous deposits, such as those found around many hot springs in the Kenya Rift Valley (Jones and Renaut 1995, 1996; Renaut et al. 1999) and New Zealand (e.g., Jones et al. 1996, 2000) are commonly formed of complex calcite and aragonite crystals that are largely devoid of preserved microbes. Similarly, iron biomineralization of microbial mats at Lýsuhóll, Iceland (Konhauser and Ferris 1996) and Calcite Springs, Yellowstone (Reysenbach et al. 1999) showed that much of the cellular details are obscured by iron precipitates (Fig. 1). In general, microbes are poorly preserved in the $CaCO_3$ and (or) iron precipitates but well preserved in silica.

The mineralization of hot spring microbial communities by silica is not limited to any particular taxa. Most microbial cells are generally silicified through the growth of spheroidal grains (tens of nanometres to 2 mm in diameter) extracellularly on the outer surfaces of living cells and intracellularly in the cytoplasm of lysed cells. If silicification is sustained, the silica particles coalesce until the individual grains are no longer distinguishable; thus, entire colonies can become cemented together in a siliceous matrix several micrometres thick.

Based on the assumption that Precambrian microfossils were cyanobacteria, Oehler and Schopf (1971) and Oehler (1976) experimentally subjected various cyanobacterial genera to colloidal silica solutions over different lengths of time. At temperatures of ~ 100 °C, several months were required for complete mineralization, and only slight alteration to the cells occurred. At higher temperatures (165 °C), the cells mineralized quickly, but the filaments fragmented, the trichomes coalesced, intracellular components were destroyed, and there was a preferential preservation of the sheath and wall material. Later, Konhauser et al. (1999) and Phoenix et al. (1999) showed that mineralization of the cyanobacterium, *Calothrix* sp., took place exclusively on the outer sheath surfaces. This contrasts with observations made on killed cells where mineralization of the cell wall and cytoplasm had taken place. Phoenix et al. (2000) subsequently suggested that the sheath might be necessary for cyanobacteria to survive mineralization, by acting as an alternative mineral nucleation site (preventing cell wall and (or) cytoplasmic mineralization) and by providing a physical barrier against colloidal silica, thereby restricting mineralization to its outer surface. This correlates well with other experimental work that has shown that incubation in saline-enriched media promotes sheath growth on microorganisms, such as *Calothrix* sp. (Padhi et al. 1998). Thus, it appears that some cyanobacteria, such as *Calothrix*, can thrive in silica-rich environments because they form a protective layer that isolates the cells from the damaging effects of silicification. Invariably, that silicification may also lead to their preferential preservation and the retention of morphological features that allow their identification (Fig. 2).

Sheaths are not, however, a prerequisite for survival in silica-saturated geothermal waters. *Oscillatoria*, for example, is a cyanobacterium that is either not ensheathed or thinly sheathed (Rippka et al. 1979), yet it has been isolated from siliceous Icelandic hot springs. Furthermore, various experimental studies have focused on the silicification of unsheathed bacteria. Westall (1997) subjected four bacterial species to highly silicifying solutions and showed that the Gram positive *Bacillus laterosporus* produced a robust and durable crust after a week of silicification, whereas the Gram negative *Pseudomonas fluorescens*, *P. vesicularis*, and *P. acidovorans* maintained delicately preserved walls that were lightly mineralized. Toporski et al. (2002) showed that *P. fluorescens* silicified to a higher extent than *Desulfovibrio indonensis* after 24 h in 1000 ppm silica solutions. With increasing levels of silicification, both bacteria suffered significant loss of shape and cellular detail.

The actual mechanisms of silicification rely, in part, on the microorganisms providing reactive surface ligands that adsorb silica from solution, and thus, reduce the activation energy barriers to heterogeneous nucleation. This means that cell surface charge may have a fundamental control on the initial silicification process and that the cells simply function as reactive interfaces, or templates, for silicification. Phoenix et al. (2002), for example, showed that the sheath of *Calothrix* is electrically neutral at pH 7, comprising predominantly neutral sugars, along with smaller amounts of negatively charged carboxyl groups and positively charged amine groups, in approximately equal proportions. On the one hand, the low reactivity of *Calothrix's* sheath gives the cells hydrophobic characteristics that facilitate their attachment to solid submerged substrates, i.e., siliceous sinters. On the other hand, this same property makes the sheath material less inhibitive to interaction with the anionic colloidal silica fraction in solution (Yee et al. 2003). Silicification subsequently occurs through hydrogen bonding between the hydroxy groups associated with the sugars and the hydroxyl ions of the silica. In contrast, the highly anionic nature of *Bacillus subtilis* limits silicification from occurring on its cell wall, likely as a result of charge repulsion between the deprotonated organic ligands and soluble silica colloids (Phoenix et al. 2003). For silicification to proceed, cation bridging (e.g., Fe^{3+}) is required.

For those bacteria that silicify, continued growth of the silica precipitates presumably occurs autocatalytically and abiogenically because of the increased surface area generated by the small silica phases. This implies that inorganic silicification is extremely important where cooling, evaporation, and (or) steam loss, mixing, and changes in effluent pH following discharge are rapid (Fournier 1985). In other words, silicification continues without any obvious control of the microorganisms simply because the cells grow in a polymerizing solution where silicification is inevitable. This notion is supported by microscopic examination of hot spring sinters where it has been shown that the silica precipitated in the porous spaces between filaments had the same basic motif and morphology as the silica precipitated on the original filaments (e.g., Ferris et al. 1986; Cady and Farmer 1995; Jones and Renaut 1996; Jones et al. 1998). Similarly, Westall et al. (1995) experimentally demonstrated that some bacteria silicified for up to four months became encrusted in very thick and dense mineral matrices that completely enshrouded the underlying cells.

From what we presently know about silicification, there are at least three main factors that lead to the preservation of intact cell structures.

(1) The timing and rate of silicification relative to death of
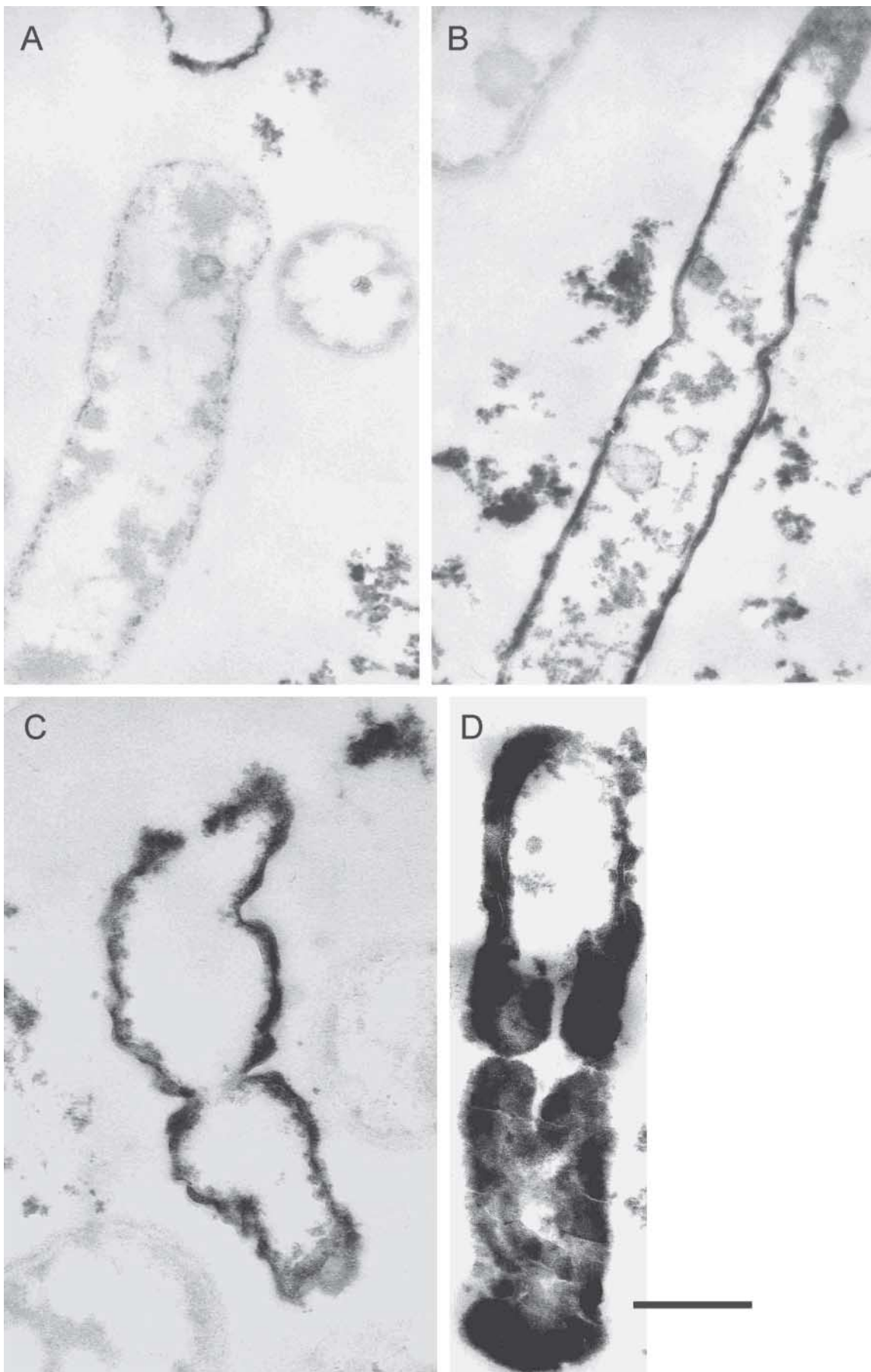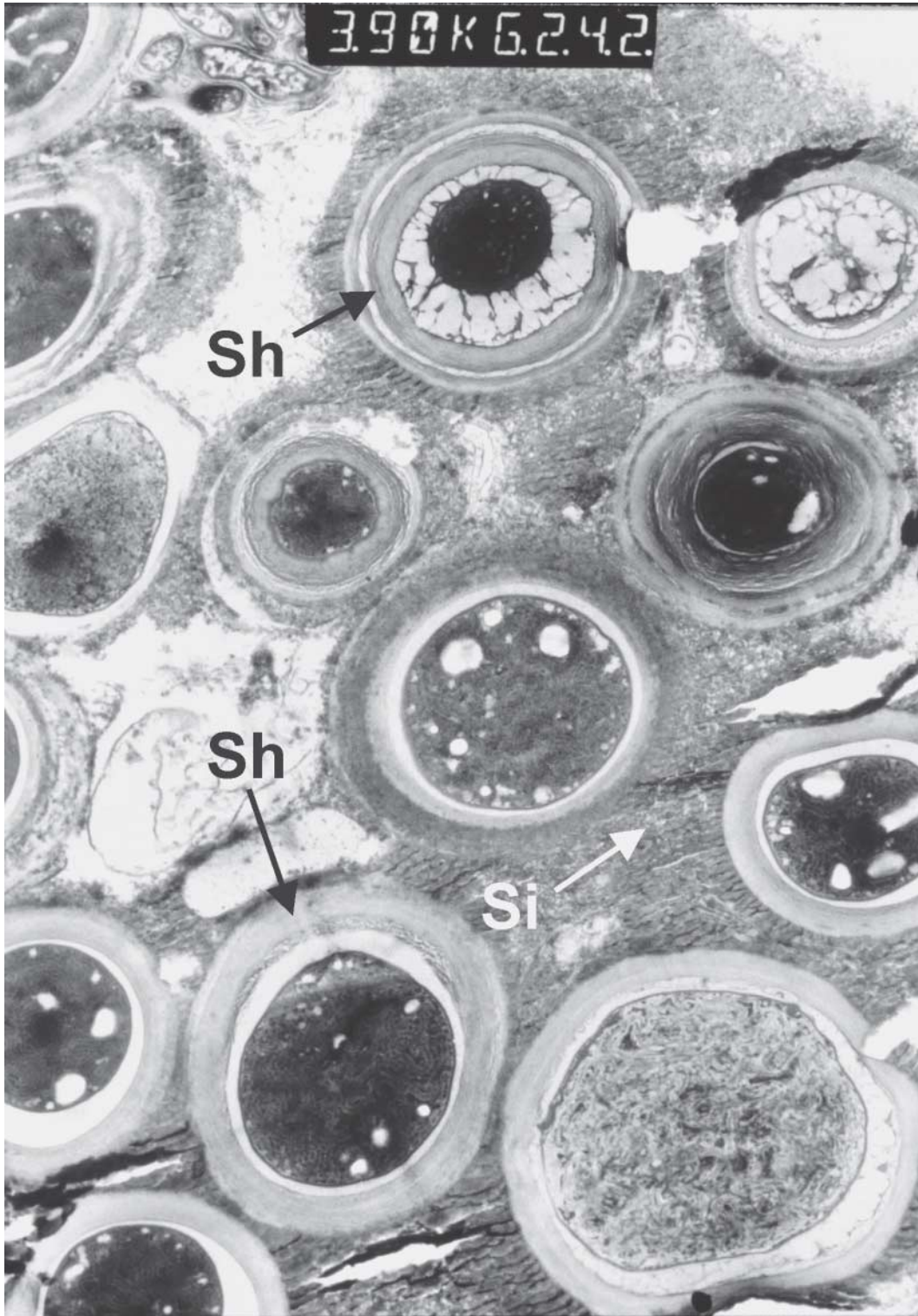
Fig. 1. Transmission electron micrographs of unstained thin sections of a filamentous mat sample collected from Calcite Springs (74 °C), Yellowstone National Park. A–D show increasing amount of iron (from energy dispersive spectroscopy analysis) accumulation in the periplasmic space of the microorganisms inhabiting this environment.These different levels of biomineralization were associated with the same sample, and from fluorescent in situ hybridization analysis (FISH), over 95% of the community were related to a separate lineage in the Aquificales (Reysenbach et al. 2000). Scale bar ≈ 0.5 μm.

Fig. 2. Transmission electron micrograph of a colony of experimentally silicified *Calothrix* sp. cells. Note the presence of intact sheaths (Sh) completely encrusted in amorphous silica precipitate (Si).

the microbes is of paramount importance (Jones et al. 2001). When silicification is rapid, both live and recently lysed cells may resist decay, thereby retaining intact morphologies (e.g., Konhauser et al. 2001). Silicification also prevents heterotrophic microbes from completely degrading the cells prior to their incorporation into the sedimentary record and for maintaining intact organic residues in a relatively impermeable matrix (Oehler and Schopf 1971). In contrast, nonmineralized cells degrade only a few days after death (Bartley 1996), and, as a consequence, their remains in the sinter may become progressively diminished, until eventually only the silica matrix remains.

(2) In Precambrian cherts, there is a preservational bias towards cells that had degradation-resistant cell walls and sheaths (Knoll 1985), while the cytoplasm and the other cellular contents may either have formed dense granules or completely degraded (Awramik et al. 1972). This is not unlike many modern environments, where species with thick sheaths are generally more resistant to degradation than those with thinner sheaths or those lacking sheaths altogether (Horodyski et al. 1977). Therefore, in terms of preservation potential, microfossil assemblages may be biased towards microorganisms such as some cyanobacteria simply because they possessed ultrastructures that proved amenable to mineralization. Conversely, other cells with different cellular features likely decomposed and left little evidence of their original organic framework (Walter et al. 1992).

(3) Ferris et al. (1988) showed that the binding of metallic ions to bacterial cell surfaces, in particular the retention of iron, was an important contributing factor to the silicification of *Bacillus subtilis*: cells not pre-mineralized by iron suffered extensive lysis after several days of ageing. This inhibition of cell degradation appears to lie with the ability of metals to deactivate the cells' own autolytic enzymes. Correspondingly, Horodyski et al. (1992) suggested that the fossil record is biased towards those cells that tolerate elevated salinities.

The subtleties of the silicification process are critical because they may control the appearance of the preserved microbe and the features that are needed to identify the microbes in terms of extant taxa (Jones et al. 2001). Many of the taxonomically critical features of microbes are lost during silicification or are concealed by mineral precipitate. Thus, a silicified microbe analyzed under scanning electron microscopy (SEM) and (or) transmission electron microscopy (TEM) may only display a few distinct features (i.e., size, general morphology, presence/absence of sheath, septa; and possibly cytoplasmic components) that can be used for identification purposes (e.g., Fig. 3). Therein lies the problem for microbial identification. For instance, Castenholz and Waterbury (1989) listed 37 characteristics that have been used in the identification of cyanobacteria. Those criteria include cell morphology (i.e., shape, size, planes of fusion), ultrastructure (i.e., cell wall), colony or filament morphology (i.e., colony shape, filament shape), genetic characteristics (i.e., DNA), culture conditions, and habitat. Unfortunately, as demonstrated by Jones et al. (2001), silicification may selectively mask and (or) destroy some features, while preserving others. Subsequently, a silicified microbe may fail to display key features

of the original microbe (Fig. 4). At other times, the silicification process may generate artefacts that actually appear biological in nature. Even in the most well-preserved silicified microbes only a few of the taxonomically important characteristics can be determined (e.g., Jones et al. 1999, 2001). It is, therefore, not surprising that the silicified biotas found in hot spring sinters typically contain less than 10 morphologically defined taxa despite the fact that the microbes seem to be so well preserved (Jones et al. 1998); the most taxa yet recognized from a silicified biota are nineteen (Jones et al. 2003).
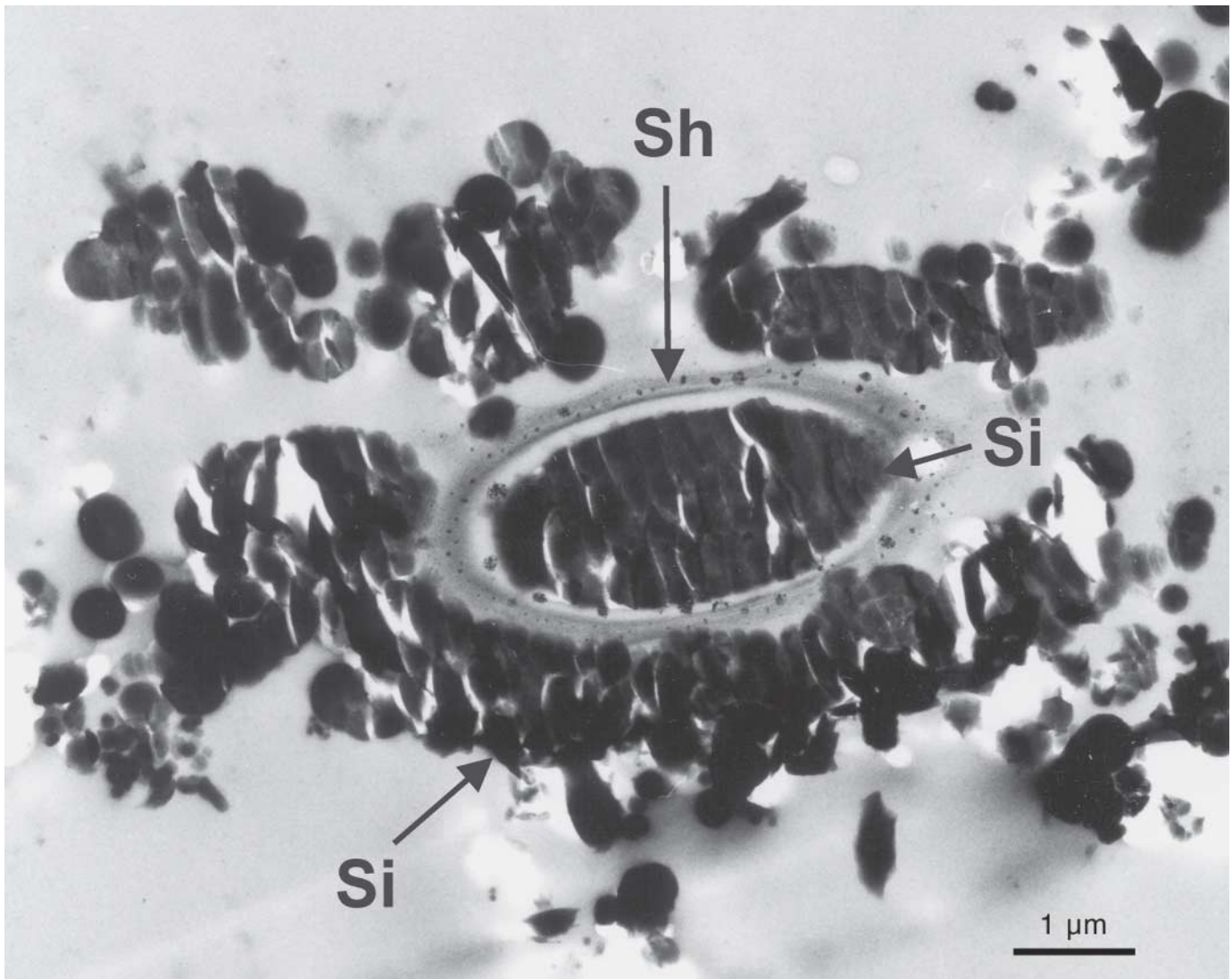
Microbial metabolic diversity

The use of rRNA sequence-based analyses to characterize modern microbial populations has increasingly been used to provide a much more comprehensive view of how life evolved on Earth. Although DNA is easily degraded, and consequently, it cannot be used to directly infer what taxa composed the earliest microfossils, molecular studies of hot spring microbial mats have led to numerous discoveries directly relevant to what might have constituted Archean life. The first is the recognition that the microbial world is much more diverse than previously imagined. Many new types of microorganisms have recently been identified, some of which represent major new lineages only distantly related to known ones (e.g., Barns et al. 1994; Hugenholtz et al. 1998). Recently, a very unusual member of the Archea, "*Nanoarchaeum equitans*," was identified and proposed as forming a new phylum in the Archea, the Nanoarchaeota (Huber et al. 2002). Meanwhile, viruses showing a wide range of morphology have been found in nearly boiling waters at Yellowstone, hosted by hyperthermophilic Archea, further highlighting our very limited view of microbial diversity (Rice et al. 2001; Rachel et al. 2002). The implications of these studies are clear. We have been limited in our comparisons between extant microbes and Archean microfossils simply because we have not been able to identify the full microbial consortia found in modern hot spring mats. Therefore, the Apex chert "microfossils" may indeed be something other than cyanobacteria or *Chloroflexus*.

A second point is that the deeply rooted lineages within the small subunit rRNA universal tree of life are all represented by thermophilic Archea and Bacteria (Stetter 1996), although other gene trees do not always support this observation (e.g., Klenk et al. 1999). Nevertheless, the former does imply that the earliest ecosystems on Earth were hydrothermal systems, and perhaps the Apex microfossils were originally thermophiles, as suggested by Brasier et al. (2002).

Thirdly, these deeply rooted hot spring Archea and Bacteria (for review of diversity see Reysenbach et al. 2002 and references therein) obtain their energy chemolithoautotrophically or chemoheterotrophically, but not photosynthetically (Barns and Nierzwicki-Bauer 1997). Many grow anaerobically by the oxidation of H_2, using sulphur compounds such as elemental sulphur and thiosulphate as electron acceptors. Some deeply rooted chemolithoautotrophs, such as *Aquifex*, can even use O_2 to oxidize H_2. The position of the Aquificales in the phylogenetic tree is particularly interesting because if O_2 utilization is indeed a primitive characteristic, then this suggests that free oxygen must have been locally available even at such early times (Stetter 1994; Towe 1994). However, many

Fig. 3. Transmission electron micrograph of a lysed cell with epicellular and intracellular silica precipitation (Si). Note the selective preservation of cell wall and sheath material (Sh). This sample was collected from the Krisuvik hot spring, Iceland at 30 °C, where *Calothrix* is the dominant microorganism comprising siliceous sinters. Yet, the TEM image yields very little information that can be used to accurately identify the microbial remains even in such modern sediment.
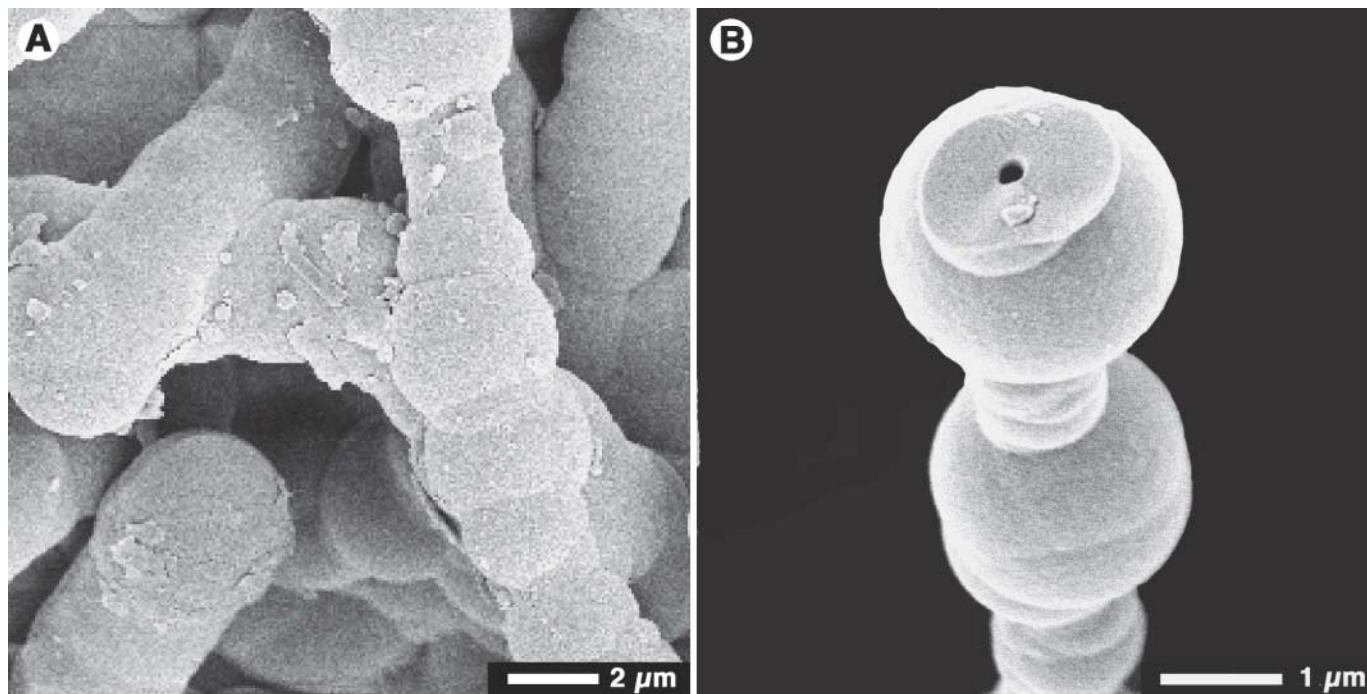

members of this order are also able to use nitrate as an electron acceptor, and some (e.g., *Persephonella marina*) are even able to use sulphur (Gotz et al. 2002). It has been proposed that the ability of this lineage to use a wide range of electron acceptors and donors, coupled with its deeply rooted position, may reflect their evolutionary history in the changing atmosphere of early Earth (Reysenbach and Shock 2002). Based on their placement in the small subunit rRNA tree, photosynthetic microbes evolved later, represented first by the divergence leading to the anoxygenic phototrophs, such as *Chloroflexus* and *Chlorobium*. The only oxygenic phototrophic lineage, represented by the cyanobacteria, diverges near the terminal tips of the bacterial tree.

Although rRNA analyses can show the phylogenetic diversity in hot spring environments, all studies have been limited to unmineralized samples. To determine the predominant microbial taxa that form particular sinters, 16S rRNA-based techniques will need to be extended to profiling the bacterial and archeal communities occurring on the surface and within the matrix of various sinters. It is likely that as the geochemical and physical conditions change during sinter formation, and therefore in response to these changes, the microbial diversity will also change. These successional changes in microbial diversity have never been studied and are clearly important for understanding what biosignatures may remain in sinters. One could therefore make systematic identifications of changes in microbial population structure between different sinter fabrics and any relationship between the physicochemical and morphological properties of the sinter and the microbial communities they harbour. Comparison of taxa from the surface and subsurface of the same sinter could also indicate if the microorganisms at depth are simply those that have been entombed in silica, perhaps with the loss of taxa that cannot protect themselves against silicification (i.e., those that do not produce ensheathed cells), or if they represent an entirely different community indigenous to the sinter. Therefore,

Fig. 4. SEM micrographs of silicified microbes from the central part of the discharge apron below Waikite Geyser in the Whakarewarewa Geothermal area, Rotorua, North Island of New Zealand. (A) Interwoven filamentous microbes with thick encrusting layers of opal-A that commonly varies in thickness along the length of individual filaments. (B) Small-diameter lumen surrounded by a thick layer of opal-A. In both figures the mineral has completely disguised all original features of the microbe.

combining rRNA analyses with electron microscopy could prove to be an invaluable way of ascertaining how different natural microbial assemblages survive silicifying solutions and perhaps assess the preservational potential of those species that make up the microbial mat community.

Sinter formation

Individual siliceous sinter deposits are architecturally complex with the lateral and vertical intercalation of various lithofacies and biofacies (i.e., geyserite, spicules, columnar and stratiform microstromatolites, oncoids, and coccoid microbial mats) being common on all scales (e.g., Jones et al. 1998). Each biofacies may be characterized by a unique microbial assemblage that developed in response to the operative hydrodynamic, geochemical, and temperature regimes (e.g., Walter 1976a; Jones et al. 1998; Renaut et al. 1998). The composition of the microflora is important because, as discussed earlier in the text, the microbes commonly act as templates for opaline silica precipitation, and thereby they must impart some control on the fabrics that develop in the sinter (e.g., Jones et al. 1998, 2001).

Sinter formation has been attributed to both abiogenic and biogenic processes (e.g., Walter 1976b; Jones et al. 1997). Indeed, the term geyserite, a dense, frequently laminated variety of sinter, was originally defined as an abiogenic siliceous precipitate that formed around the vents of hot springs and geysers where the high temperature setting (> 73 °C) was deemed sterile, except for scattered thermophilic microbes (Walter 1976b). Geyserite has received considerable attention because its internal laminated structures are similar to those

found in some Precambrian siliceous stromatolites. Examination of geyserite from Yellowstone and New Zealand, however, has shown that geyserite surfaces are commonly covered with biofilms and that their laminae generally contain silicified microbes (e.g., Cady et al. 1995; Jones et al. 1997). Thus, not all geyserite can be regarded as being abiogenic, and it appears that most siliceous sinters have been constructed, to some degree, around microbes. Siliceous sinters that formed on the more distal parts of the discharge aprons, where temperatures are much lower, are commonly characterized by complex fabrics that are controlled by the attitude of the microbes that lived in those settings (e.g., Jones et al. 1998).

Based on various studies in Yellowstone, New Zealand, Kenya, and Iceland, we now know that some species respond to complete encrustation by being motile, moving (or growing) in the direction of accretion faster than the rate of sedimentation. If this progression proceeds at an even rate, then the resulting stromatolite will either appear uniform (with degradation removing the dead organic matter at the bottom) or a finely laminated structure results (Golubic 1976). In Yellowstone, sinter laminae tens of micrometres thick have been attributed to daily growth patterns among unicellular cyanobacteria, *Synechococcus* sp., and *Chloroflexus* sp. (Walter et al. 1972; Doemel and Brock 1974). The upward migration of *Chloroflexus* at night, in response to low light levels and positive aerotaxis, causes the bacteria to accumulate at the surface, while the following day, rapid growth by *Synechococcus* results in the re-population by the cyanobacteria at the mat surface. In siliceous microstromatolites from Dragon's Mouth Geyser and Ohaaki Pool, New Zealand, Jones et al. (1997, 1998) have described erect, large-diameter filaments, aligned

parallel with each other and sub-perpendicular to the growth surface, alternating with layers in which numerous small-diameter filaments lie parallel to the surface. The cause of the alternating erect and prostrate laminae has been tentatively associated with seasonal changes. Other sinters, such as those in Krisuvik, Iceland, display much coarser laminations (Konhauser et al. 1999, 2001). There, the siliceous microstromatolites are formed of alternating layers, each ~ 250 mm thick, of filamentous cyanobacteria (predominantly *Calothrix* sp.) and pure silica, devoid of any microbial component. The cyclical pattern arises from active cell growth during spring and summer when the microorganisms can keep pace with silicification, while during their natural slow growth phase in the dark autumn and winter months silicification exceeds the bacteria's ability to grow upwards. When conditions once again become favourable for growth, re-colonization of the solid silica surface by free-living cyanobacteria occurs.

Understanding modern sinter formation has important implications for the rock record. As the pioneering work by Walter et al. (1972) first suggested, it may be possible to interpret some distinctive ancient stromatolite morphologies from modern hot spring analogues. For this to be useful, however, a general framework illustrating the association between the principal types of siliceous sinter and the dominant mat-forming communities needs to be developed. Unfortunately, this is where gaps in our knowledge emerge. Numerous studies suggest that cyanobacteria exert dominant control on sinter fabrics at temperatures below 73 °C (e.g., Brock 1978; Cassie 1989), but what happens at higher water temperatures or where anoxic conditions prevail (i.e., habitats of thermophiles and *Chloroflexus*, respectively) is indeterminate. Silicified hyperthermophilic Archea and viruses are probably also present in some geyserite, but as yet have not been identified with confidence.

At present, we can only surmise how such microorganisms interact with their solute-rich environment, and whether or not species-specific patterns of sinter morphology and fabric development truly exist. Stated simply, if we assume microbial community composition controls the sinter fabric then we would expect that different sinters would have different microbial community structures. Conversely, sinters with similar fabric from different springs or different parts of the same spring system should have microbial communities that are more similar to each other than they are to sinters of different structure and composition.

Conclusions

Molecular microbiology and electron microscopy have long been used in the study of hot spring microbiota. However, these techniques have not been properly integrated with the view of developing a framework with which to assess the primary community structure of Archean microbial mats. Ribosomal rRNA analyses of surface and subsurface hot spring sinters provide a picture of the indigenous microbial communities in both microenvironments, while SEM and TEM highlight which microorganisms biomineralize, whether they retain intact and recognizable cell morphologies at depth, and hence age (i.e., as fossils), and whether preservational biases occur in modern sinter communities. Moreover, electron microscopy can also generate detailed observations of the fabrics and the patterns of laminae development in a litany of sinter types. This is extremely significant for understanding what composed the Archean microfossil assemblages because by comparing hot spring sinters (where we can relate the preservation of silicified microbes at depth versus the surface populations) with microfossil-bearing stromatolites containing similar biosedimentary features, we may finally be able to extrapolate the primary community structure of the ancient microbial mats.

Acknowledgments

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