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Abstract

The origin of organic microstructures in the ~ 3 Ga Farrel Quartzite is controversial due to their relatively poor state of preservation, the Archean age of the cherts in which they occur, and the unusual spindle-like morphology of some of the forms. To provide more insight into the significance of these microstructures, nano-scale secondary ion mass spectrometry (NanoSIMS) maps of carbon, nitrogen, sulfur, silicon, and oxygen were obtained for spheroidal and spindle-shaped constituents of the Farrel Quartzite assemblage. Results suggest that the structures are all bona fide ~ 3 Ga microfossils.

The spindles demonstrate an architecture that is remarkable for 3 Ga organisms. They are relatively large, robust, and morphologically complex. The NanoSIMS element maps corroborate their complexity by demonstrating an intricate, internal network of organic material that fills many of the spindles and extends continuously from the body of these structures into their spearlike appendages.

presputtering, spot size, e-gun) were used for analyzing the standard and the Precambrian structures. Elemental results from the Precambrian samples measured by NanoSIMS were normalized to results from the standard.

The NanoSIMS results were processed with L'Image software developed by L. Nittler, Carnegie Institution of Washington, Washington DC.

3. Results

NanoSIMS element maps were obtained for three categories of microstructures in cherts of the Farrel Quartzite: small spheroids, large spheroids, and spindle-shaped structures (Figs. 1–5). Comparisons were made with NanoSIMS element distributions in both undisputed microfossils from the ~0.83 Ga Bitter Springs Formation and secondary carbonaceous material in a hydrothermal vein in an Archean sample from the ~3.4 Ga Strelley Pool Chert (Oehler *et al.*, 2006, 2008a, 2008b, 2008c, 2009). For biogenicity assessment, characteristics utilized were the C^- and CN^- responses (size, shape, alignment and continuity), the relationship between intensities of C^- and CN^-, and the spatial relationships of C^- and CN^- to microstructures imaged in optical photomicrographs. For syngeneity assessment, Si^- and O^- concentrations

FIG. 1. NanoSIMS element maps of small spheroids in chert from the Farrel Quartzite of Western Australia. (**A**–**E**) NanoSIMS maps for carbon ($^{12}C^-$), nitrogen ($^{12}C^{14}N^-$), sulfur ($^{32}S^-$), oxygen ($^{16}O^-$), and silicon (

FIG. 2. NanoSIMS element maps of a large spheroid in chert from the Farrel Quartzite. (**A**–**E**) NanoSIMS maps for carbon ($^{12}C^-$), nitrogen ($^{12}C^{14}N^-$), sulfur ($^{32}S^-$), oxygen ($^{16}O^-$), and silicon ($^{28}Si^-$). Color bars and scales indicate yield of ions (intensity of response). The scales are set by the image processing software. (**F**) Optical photomicrograph in transmitted light

were compared to C^- and CN^- concentrations as well as to indications of Si^- and O^- in the chert matrix.

Results imply that the microstructures analyzed from the Farrel Quartzite are both biogenetic in derivation and syngenetic to the chert in which they are preserved.

3.1. [illegible]

Small spheroids have been described from this assemblage based largely on optical microscopy (Sugitani et al., 2007, 2009b). These spheroids (Fig. 1F) range in diameter from less than 5 mm to about 15 mm and occur in irregular clusters of a few to a few tens of individuals. The spheroids commonly appear to be loosely joined by strandlike connections or diffuse mucilaginous-like material. Within a cluster, individual cell-like bodies are irregularly and rather poorly preserved. No obvious internal contents have been noted. The walls of these spheroids are irregularly granular. More than 300 specimens in this category were documented by Sugitani et al. (2007).

NanoSIMS element maps of a cluster of small spheroids are shown in Fig. 1A–E. Figure 1F shows an optical photomicrograph of the cluster, and the red rectangle illustrates the portion of that cluster that was analyzed by NanoSIMS. Most of the spheroids in this grouping are between about 4 and 7 mm in diameter. Since NanoSIMS analysis investigates only the topmost several nanometers of thin sections, three-dimensional structures that can be observed in optical microscopy are frequently only partially imaged in NanoSIMS. Consequently, the NanoSIMS element maps of the cluster of small spheroids captured only fragments of five individual structures. The structure at the center of this grouping was captured almost in its entirety, yielding secondary ions from the nearly complete perimeter. In the other four structures, the NanoSIMS maps yielded secondary ions from more irregular portions of the spheroids. Nevertheless, all the analyzed fragments of these small spheroids have the following characteristics in common:

- The distribution maps of C^-, CN^-, and S^- have a one-to-one correspondence with the structures observed by optical microscopy (Fig. 1A–C, F);
- Variations in C^-, CN^-, and S^- concentrations parallel one another (Fig. 1A–C);
- The C^-, CN^-, and S^- responses correspond to aligned element concentrations that are partially contiguous and form enveloping, wall-like boundaries (Fig. 1A–C).
- The C^-, CN^-, and S^- concentrations are globular and $\sim$0.2 to 0.5 m

Si^- and O^- in association with the microstructures are material that comprises this spheroid and show relationships among C^-, CN^-, and S^- that are identical to those for the small spheroids and suggestive of biogenicity. In addition, the C^-, CN^-, and S^- element concentrations of the large spheroid show the globular, aligned, and partially contiguous character that is reminiscent of remnants of cell walls (Oehler *et al.*, 2006). A comparison of element maps for O^- and Si^- with those for C^- and CN^- (Fig. 2A–B, 2C–D) shows a close correspondence between the organic material and the O^- and Si^-.

One difference between the NanoSIMS results for the large and small spheroids is that the bounding material of the large spheroids is more uniform in width than that of the small spheroids (~0.4 to 0.6 mm vs. 0.2 to 0.5 mm for the small spheroids). Another difference is the occurrence of numerous, subcircular granules with unusually high C^-, CN^-, and S^- content in the large spheroid. These granules occur within images, the appendages and the body appear to be made of the same material. They both exhibit nearly identical element compositions and thicknesses, both consist of the carbonaceous reticulate network, and the connection between the appendage and the body is continuous with no evidence of any jointing or separation.

Like the large spheroids, the spindles contain numerous, subcircular granules with high C^-, CN^-, and S^- content (Figs. 3–5). The C^-, CN^-, and S^- responses of these granules are much higher than typical responses for the wall-like material (with intensities ranging from 2 to 10 times that of the majority of the wall-like material). Their size range is similar to that of the granules described from the large spheroids (less than 0.3 mm to nearly 1 mm in diameter), and they appear to be ringed by O^- and Si^- (especially well illustrated by arrows in Fig. 5).

The spindles in Fig. 3 show patches of material with atypically high CN^- response (arrow, Fig. 3B). The sulfur map shows similar regions of enrichment (arrow, Fig. 3C), but the carbon map appears to lack equivalent enrichment in those regions (Fig. 3A). The patches are somewhat angular in shape and adjacent to the cavities in the upper spindle of Fig. 3.

4. Discussion

4.1. B

The correspondence and parallel variations among C^-, CN^-, and S^- in the Farrel Quartzite forms, coupled with the aligned (wall-like) and globular character of the element concentrations, suggest that the material comprising each of the structures analyzed is biogenic. Similar relationships have been seen in microfossils from the Bitter Springs Formation (Oehler et al., 2006, 2008a, 2008b, 2008c, 2009) and in NanoSIMS analyses of modern cyanobacteria (Eybe et al., 2007). In the modern cyanobacteria, it has been established that the NanoSIMS C^- and CN^- responses reflect the general morphology of the cells, and the S^- response reflects mainly protein distributions. Biogenicity of the spindles is discussed further in Section 4.6.

4.2.

The silicon and oxygen maps for each of the microstructures studied from the Farrel Quartzite show enhancements in these elements that are intimately associated with (and almost mimic) the carbon, nitrogen, and sulfur distributions.

A similar enhancement of silicon and oxygen was observed in the Bitter Springs microfossils (Oehler et al., 2006), and it has been suggested that such enhancement of Si^- and O^- reflects the intimate association between silica and organic matter that results from silica permineralization (Oehler et al., 2009). Silica permineralization involves nucleation of silica on organic surfaces, with initial weak bonding between the silica and functional groups in biological materials. Silica permineralization of biological structures is well known from (1) laboratory experiments (Oehler and Schopf, 1971; Oehler, 1976; Toporski et al., 2002), (2) observations of natural silica nucleation on modern microbes (Phoenix et al., 2000; Benning et al., 2002; Renaut et al., 2002), and (3) electron microscopic analyses of ancient microfossils (Moreau and Sharp, 2004). The latter study showed this intimate relationship in spheroidal microfossils from the ∼2 Ga Gunflint Formation, where the wall was comprised of kerogen intimately intermixed with silica. The silica in the walls is very fine grained (100 by 300 nm) compared to the larger crystals (750 by 1000 nm) in the chert matrix, and similar size relationships have been observed in other studies of organic matter and silica (Oehler and Logan, 1977; Altermann and Schopf, 1995; Kempe et al., 2005).

Such intimate intermixing of fine-grained silica and kerogen seems likely to account for the enhanced Si^- and O^- response in the walls of the Bitter Springs microfossils. The mechanism by which this occurs could be due to a "matrix effect" in NanoSIMS where Si^- and O^- from the silica associated with kerogen ionize more readily than Si^- and O^- from the matrix chert. Such preferential ionization could result from the comparatively small size of silica grains associated with the kerogen or the weak bonding at the kerogen-silica interface, or both. Alternatively, as carbonaceous material is known to sputter faster than chert (Dr. C. House, personal communication, 2008), the enhancement may result from the more rapid sputtering of the organic material during analysis, which would allow comparatively more silica mixed with the kerogen of the cell wall to be exposed to the primary ion beam. In either case, enhanced responses of Si^- and O^- associated with C^- and CN^- would be an indicator of an intimate association between silica and kerogen, and that is suggestive of the process of silica permineralization of biological materials. Since permineralization involves functional groups, it must occur prior to their loss through diagenesis, in the earliest stages of preservation. The fact that primary cherts preserve organic microfossils three-dimensionally, prior to their having been significantly compressed by burial, similarly attests to very early diagenetic occurrence of this process.

In contrast, carbonaceous matter in a secondary vein of the Eo-Archean Strelley Pool Chert shows no enhancement of Si^- associated with C^- or CN^- (Oehler et al., 2009), which perhaps reflects a lack of intermixed, fine-grained silica such as has been imaged in the Gunflint microfossils (Moreau and Sharp, 2004). Petrographic relationships demonstrate that the vein formed after the chert matrix (Oehler et al., 2009), and it and the carbonaceous material within are clearly epigenetic. The lack of intimately intermixed silica and carbon might reflect the fact that silica crystallization in the vein occurred later in diagenesis, when few functional groups remained in the carbonaceous material and the potential for bonding between silica and organic materials, therefore, was eliminated.

Thus, the spatial relationships among Si^-, O^-, C^-, and CN^- in the Farrel Quartzite microstructures provide support for their syngeneity with the chert matrix in which they occur. This conclusion is in line with (1) petrographic observations that the Farrel Quartzite microstructures are an integral part of the primary sedimentary fabric of the matrix chert (Sugitani et al., 2007), and (2) results of laser-raman spectroscopy (Dr. J.W. Schopf, personal communication, 2009).

4.3. I

In all specimens analyzed, the oxygen response is more intense than that of the silicon. This was true of the Bitter

Springs microfossils as well. The intensity of oxygen may result from a high ionization yield for oxygen in the cesium beam of the NanoSIMS, or it may, in part, reflect additional oxygen that was originally a constituent of sugars, proteins, or lipids, as has been shown by the NanoSIMS mapping of modern cyanobacteria (Eybe et al., 2007).

4.4. *a* [illegible] C^-, C [illegible] $^-$, *a* [illegible] $^-$

The granules have only been seen in the large spheroids and spindles; they do not appear in the small spheroids analyzed to date. In the granules, the C^-, CN^-, and S^- yields appear to vary in parallel. In the high-resolution C^- and CN^- maps (Fig. 5A–B), the correspondence of high C^- and CN^- is apparent. Figure 4A–C also illustrates the correspondence of high S^- with high C^- and high CN^-. This correspondence suggests that the granules may have been derived from organic constituents similar to those that make up the bulk of the large spheroids and spindles. The fact that the granules are associated with enhanced silicon and oxygen concentrations at their perimeters (Figs. 4D–E, 5C) suggests that the granules were present prior to silicification. In all characteristics mentioned above, the granules are different from the secondary carbonaceous material that was analyzed by NanoSIMS from the secondary vein in a

oxyge36.7(8r)5.59servn S ap7-OF131Tf6.7246005.9768270.1(C)61(h.549n)74()TjF41Tf8.9663008.9663154.584i8755f5a7he cesieatche3,6.7(C)c

experimentation that may have no analogy among modern microbial forms.

The Archean age of the spindles might suggest prokaryotic affinities. Although their large sizes are more typical of eukaryotic microbes (Schopf, 1992), some prokaryotic spores can reach large and even macroscopic sizes (Walsh, 1992). Certainly the organizational complexity of the spindles (with their flanges, spearlike appendages, and internal reticulate networks) attests to a relatively advanced morphology.

Recent biomarker studies by Waldbauer *et al.* (2009) concluded that biochemical innovation in the Archean laid the groundwork for all three domains of life (Bacteria, Archaea, and Eukaryota); accordingly, it is even conceivable that the spindles could be early representatives of the Domain Eukaryota. At a minimum, results from our work support the existence of a diverse microbiota by 3 Ga with individual constituents (the spindles) exhibiting an advanced level of structural organization. These results align with conclusions from Waldbauer *et al.* (2009). Both studies point to surprising evolutionary diversification in the Archean.

5. Conclusions

NanoSIMS element distributions for small spheroids, large spheroids, and spindles preserved in chert of the ~3 Ga Farrel Quartzite suggest that each of these is a bona fide microfossil of an Archean microorganism. This assemblage, then, joins the few handfuls of examples of organically preserved microbiotas of Archean age (summarized in Schopf, 2006). And these microbiota join a host of recent stromatolitic and geochemical results indicative of a multifaceted Archean biosphere (references listed in the Introduction). Together, these lines of evidence are building a picture of Archean life that was surprisingly diverse and probably metabolically and ecologically more advanced than might have been surmised just a few years ago.

The spindle-like forms are of particular interest because of their large size and morphological complexity—with flanges, spearlike appendages, and an intricate internal network that is integral to both the body and appendages. This level of organization is in contrast to that of the simpler and generally smaller spheroids and filaments common to many Precambrian microbiotas. The spindles are abundant in the Farrel Quartzite cherts, and they exhibit an apparent robustness that might suggest an origin from a resistant organism—possibly one that developed on an environmentally hostile young planet.

Thus, the microbiota of the Farrel Quartzite is notable, not only because it comprises a ~3 Ga assemblage of organically preserved microfossils but also because it is diverse and includes spindle-shaped forms with relatively advanced morphology. If the recent suggestion by Waldbauer *et al.* (2009) is correct—that biochemical innovation in the Archean laid the groundwork for development of the three domains of life—then our work, which supports the biogenicity and syngeneity of the Farrel Quartzite forms, complements and aligns with this view of early evolution on Earth.

Evolution of life on other planets, similarly, may involve

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