

■ Chemical nature of the 3.4 Ga Strelley Pool microfossils

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■ Supplementary Information

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Methods

Raman

Raman microspectroscopy measurements were performed using a Renishaw INVIA microspectrometer (IMPMC, Paris, France) on a freshly fractured sample at room temperature using a 514.5-nm wavelength 50-mW Modulaser Argon laser (green laser). Focusing was achieved using a Leica DMLM microscope with a long working distance × 100 objective (numerical aperture = 0.75). This configuration yields a horizontal resolution of $\approx 1 \mu\text{m}$ for a laser power below 1 mW, intended to prevent irreversible laser-induced thermal damage (Beyssac *et al.*, 2003; Bernard *et al.*, 2008; Beyssac and Lazzeri, 2012). Extraction of spectral parameters from peak fitting procedures and estimation of peak temperature experienced by organic carbon were done following the procedure described by Beyssac *et al.* (2002), Lahfid *et al.* (2010), and Beyssac and Lazzeri (2012).

SEM

SEM was used to locate the organic microfossils within the silica matrix of Strelley Pool chert investigated herein, as well as for their subsequent extraction using a FIB. To minimise contamination that may arise during sample preparation, freshly fractured fragments were directly observed after mounting and gold coating on aluminium stubs. These SEM observations were made using the SEM–field emission gun ultra 55 Zeiss at IMPMC (Paris, France), exploiting a 15-kV accelerating voltage and a working distance of 3 mm.

FIB

Focused ion beam (FIB) ultrathin sections were extracted from the organic microfossils using an FEI Strata DB 235 (IEMN, Lille, France). Milling at low Ga-ion currents minimises common artefacts including: local gallium implantation, mixing of components, creation of vacancies or interstitials, creation of amorphous layers, local compositional changes or redeposition of the sputtered material on the sample surface (Schiffbauer and Xiao, 2009).

TEM

Transmission electron microscope (TEM) analyses were performed on FIB sections to document the textural nature of the Strelley Pool microfossils and identify the mineral phases with which the organics are associated at the nanoscale. These TEM observations



were performed using a FEI TECNAI G2 20 microscope (UMET, Lille, France) operating at 200 kV with a LaB₆ filament. Scanning TEM Z-contrast imaging was performed using the high-angle annular dark field mode.

XANES

The XANES data were collected on the 10ID-1 STXM beamline at the Canadian Light Source (Kaznatcheev *et al.*, 2007) and on the HERMES STXM beamline at the synchrotron SOLEIL (Belkhou *et al.*, 2015; Swaraj *et al.*, 2017). At CLS, a 100 nm thick titanium filter is used to remove the contribution of second-order light. At SOLEIL, beamline optical elements are exposed to a continuous flow of pure O₂ to remove carbon contamination. Microscope chambers were evacuated to less than 100 mTorr after sample introduction. Energy calibration was achieved using the well-resolved 3p Rydberg peak of gaseous CO₂ at 294.96 eV for the C *K*-edge and using the 1 → π* photoabsorption resonance of gaseous N₂ at 400.8 eV for the N *K*-edge. X-ray absorption spectroscopy was performed by collecting image stacks with a spatial resolution of 25 nm, thereby rastering selected areas in the x–y domain at energy increments of 1 eV over the 270–450 eV energy range using the low-energy grating of the 10ID-1 SM beamline. Additional image stacks were collected at energy increments of 0.1 eV over the carbon (270–340 eV) and the nitrogen (390–450 eV) absorption ranges, to resolve the fine structures near the C and N *K*-edges (XANES spectroscopy). Stack measurements were performed with a dwell time of ≤1 ms per pixel to prevent irradiation damage. Alignment of images of stacks and extraction of XANES spectra were achieved using the aXis2000 software (ver2.1n). The C- and N- XANES spectra shown in the present contribution correspond to homogeneous organic-rich areas of several hundreds of square nanometres. N/C atomic ratio values were estimated with an uncertainty of ± 0.02 following the methodology and calibration outlined in Alleon *et al.* (2015). According to the Beer–Lambert law (Stöhr, 1992), the intensity of the radiation passing through the sample (*I*) is related to the intensity of the incident radiation (*I*₀) following the equation $I = I_0 e^{-\mu l}$; with μ the mass absorption coefficient, *l* the thickness of the sample and ρ its volumetric mass density. The absorption signal ($A = -\ln(I/I_0)$), *i.e.* the absorbance or optical density, is thus the product of the thickness times the volumetric mass density times the mass absorption coefficient ($A = \mu l \rho$). According to Henke *et al.* (1982; 1993), for a given element, the mass absorption coefficient (μ) is directly proportional to the atomic photoabsorption cross section (σ_a), which is itself directly proportional to the *f*₂ component (imaginary part) of the complex atomic scattering factor. Thus, for a given area of a sample (*i.e.* for a given thickness and a given volumetric mass density), an atomic ratio can be directly estimated by dividing the coefficients used to fit the sum of the *f*₂ components of carbon and nitrogen to the measured absorption signal below and above the edge regions. Here, we used the *f*₂ components of carbon and nitrogen reported by Henke *et al.* (1982; 1993) to estimate the N/C values of the Strelley Pool microfossils from their XANES spectra. Note that the *f*₂ components of most elements can be found online (http://henke.lbl.gov/optical_constants/).

In some cases, the presence of potassium and/or calcium nitrates (N-XANES absorption features at 401.7 and 405.4 eV) may slightly increase the estimated N/C values. Yet, the very weak absorption steps observed at the potassium (295–300 eV) and calcium (340–360 eV) *L*-edges on the Strelley Pool microfossils indicate that their calcium and potassium contents are very low and thus do not significantly impact the estimated N/C values.

Supplementary Information References

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