STXM-XANES analyses of carbonaceous matter in seafloor hydrothermal deposits from the ~3.5 Ga Dresser Formation in the North Pole area, Western Australia

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Supplementary Material

Samples

We analyzed the samples collected from a silica vein (96NP452) that intrudes basaltic greenstones of the ~3.5 Ga Dresser Formation in the North Pole area of Western Australia. The sampling locality of the silica vein was shown in previous study (Igisu *et al.*, 2018). Detailed descriptions of the localities and samples can be found in the works of Ueno *et al.* (2001, 2004, 2006). The ~3.5 Ga Dresser Formation is one of Earth's oldest records of seafloor hydrothermal deposits, where prominent silica-barite veins containing carbonaceous matter ubiquitously penetrate pillowed basaltic greenstones (Ueno *et al.,* 2004; Van Kranendonk, 2006; Van Kranendonk *et al.,* 2008). Carbonaceous matter present in the silica veins provides an important means to understanding how organic matter was synthesized in Earth's early hydrothermal systems. The occurrence of biological activity in the Dresser Formation has been inferred from observations of putative stromatolites (Walter, 1980; Van Kranendonk, 2006; Van Kranendonk *et al.,* 2008), microbially induced sedimentary structures (Noffke *et al.,* 2013), and putative microfossils (Ueno *et al.,* 2001), as well as geochemical signatures in silica-barite veins, including ^{13}C-depleted carbonaceous matter and methane (Ueno *et al.,* 2001, 2004, 2006; Glikson *et al.,* 2008; Morag *et al.,* 2016), ^{34}S-depleted pyrite with characteristic $\Delta^{33}S$ values (Ueno *et al.,* 2008; Shen *et al.,* 2009), and the molecular characteristics of carbonaceous matter (Derenne *et al.,* 2008; Duda *et al.,* 2018; Igisu *et al.,* 2018). The results of previous geochemical analyses suggest the possible presence of prokaryotic communities in ancient hydrothermal

ecosystems; however, abiotic origins have also been proposed for the putative morphological fossils and organic matter (Lowe, 1994; McCollom *et al.,* 1999; McCollom and Seewald, 2006).

Petrographic investigation of the thin section indicated that the silica vein is mainly composed of microcrystalline quartz (< 10 μm), carbonaceous matter, sulfides, and carbonates. We selected five carbonaceous clots in petrographic thin section of the silica vein that have been previously analyzed by Raman and Fourier transform infrared (FTIR) microspectroscopies (Igisu *et al.,* 2018; #01, #02, #13, #15, and #31 in Figure 1). Native Fe-Ni minerals and other preferred catalysts for Fischer–Tropsch-type (FTT) reactions were not observed in the studied thin section (Igisu *et al.,* 2018) or in previously reported samples (Ueno et al., 2006). The five carbonaceous clots show different infrared (IR) features of aliphatic CH_3/CH_2 groups (absorbance ratios of aliphatic CH_3/CH_2 groups = 0.22–0.51 for #01, #02, #13 and #15: Igisu *et al.,* 2018; and no signals aliphatic CH_3/CH_2 groups for #31).

Raman microspectroscopy

A laser Raman microspectrometer (RAMANtouch, nanophoton) was used to identify the presence of carbonaceous matter and examine its graphitization degree. The excitation laser was a green laser with a wavelength of 532 nm. The carbonaceous clots were analyzed using the X–Z mapping mode with an exposure time of 5 s and one accumulation. Raman spectra for wavenumbers in the range of ~2670–110 cm^{-1} were acquired with a 600 grooves/mm grating. The spot size was < 1 μm using a 100 × objective lens with numerical aperture of 0.9. The incident laser power density was ~1 × 10^5 W/cm^2, and the final power on the sample surface was estimated to be ~0.4 mW. Prior to sample analysis, wavenumber calibration was performed by comparing the Raman spectra with a standard silicon spectrum (520 cm^{-1}).

Raman spectroscopy has been used to obtain a metamorphic grade indicator of geological samples containing carbonaceous matter (Yui *et al.,* 1996; Beyssac *et al.,* 2002; Kouketsu *et al.,* 2014). Several indicators have been used to estimate peak metamorphic

temperatures using Raman spectral parameters: intensity ratio, area ratio, and full width at half maximum (FWHM) of the ~1580 cm^{-1} (graphite, G) and ~1350 cm^{-1} (disordered, D) bands. Following Kouketsu *et al.* (2014), we selected a five-band model: D1 band (~1350 cm^{-1}), D2 band (~1620 cm^{-1}), D3 band (~1510 cm^{-1}), D4 band (~1245 cm^{-1}), and G band (~1580 cm^{-1}). Using the FWHM of the D1 and D2 bands (FWHM-D1 and FWHM-D2, respectively), the maturation temperature of carbonaceous matter was estimated (Kouketsu *et al.,* 2014):

$$T\ (°C) = -2.15\ (\text{FWHM-D1}) + 478 \quad (1)$$

$$T\ (°C) = -6.78\ (\text{FWHM-D2}) + 535 \quad (2)$$

The error in equation (1) is approximately ± 30 °C (Kouketsu *et al.,* 2014). The FWHM-D1 and FWHM-D2 values are 65 ± 3 and 33 ± 5, respectively. The Raman spectra of carbonaceous clots are equivalent to those of CM from lower greenschist facies metasediments (Yui *et al.,* 1996; Kouketsu *et al.,* 2014), consistent with the metamorphic grade of basaltic greenstones surrounding the silica veins (below greenschist facies, < 350 °C; Kitajima *et al.,* 2001; Ueno *et al.,* 2001, 2004). We used equation (1) to calculate the maturation temperature of carbonaceous clots in order to compare the data with the previous spectral data of Archean samples (Alleon *et al.,* 2018, 2019, 2021). To estimate the maturation temperature of carbonaceous clots, we selected the spectral data of only the carbonaceous clots embedded within rocks below the surface of the thin section to avoid the effect of polishing, which can induce deformation of carbonaceous matter during sample preparation and possibly cause artificial modification of Raman spectroscopic feature (Pasteris, 1989). The Raman spectral data were processed using the PeakFit v.4.12 software (SeaSolve Software Inc.).

Focused ion beam scanning electron microscopy

STXM analysis requires ~100-nm-thick sections to transmit soft X-rays for chemical analysis. Therefore, approximately 100-nm-thick sections were extracted from five carbonaceous clots in the silica vein using a focused ion beam scanning electron microscope

(FIB-SEM, Helios G4 UX FEI).

Scanning transmission X-ray microscopy and X-ray absorption near-edge structure spectroscopy

Carbon and nitrogen X-ray absorption near-edge structure spectroscopy (C- and N-XANES) were performed using STXM at BL-19A of the Photon Factory at High Energy Accelerator Research Organization (KEK) (Takeichi *et al.,* 2014, 2016). The carbon map was obtained by acquiring the pairs of images below and on the carbon *K*-edge, at 280 and 290 eV, respectively, for each pixel. The energy steps (ΔE) for C-XANES spectra differed among FIB foils (Table S1). For N-XANES, ΔE was 0.2 eV in the 398–406 eV region, 0.3 eV in the 406–409.9 eV, and 0.5 eV in the 390–398 eV region. Stack measurements were performed with a dwell time of 2 or 5 ms per pixel for C-XANES analyses and 2 or 10 ms per pixel for N-XANES analyses. STXM-XANES data analysis was performed using the aXis2000 software (http://unicorn.mcmaster.ca/aXis2000.html).

The FIB foil extracted from carbonaceous clot #13 did not contain any black aggregates in its optical microscopic images, nor show any absorption peaks in its C-XANES spectra. This may be because a CM-free part of carbonaceous clot #13 was coincidentally extracted, and thus the results of carbonaceous clot #13 were excluded from the discussion.

Table S1. Energy steps for carbon X-ray absorption near-edge structure spectroscopy (C-XANES)

Foil#	Range [eV]	Energy step [eV]
#01, #02	280–284.5	0.5
	284.5–292	0.1
	292–300	0.5
	300–320	1
#09, #31	280–284	0.5

	284–291.5	0.15
	291.5–299	0.3
	299–320	0.5
#15	280–284.5	0.5
	284.5–292	0.1
	292–295	0.2
	295–310	1

References

Alleon, J., Bernard, S., Le Guillou, C., Beyssac, O., Sugitani, K. and Robert, F. (2018) Chemical nature of the 3.4 Ga Strelley Pool microfossils. *Geochem. Perspect. Lett.* 7, 37–42. DOI: 10.7185/geochemlet.1817

Alleon, J., Bernard, S., Olivier, N., Thomazo, C. and Marin-Carbonne, J. (2021) Inherited geochemical diversity of 3.4 Ga organic films from the Buck Reef Chert, South Africa. *Commun. Earth Environ.* 2, 6. DOI: 10.1038/s43247-020-00066-7

Alleon, J., Flannery, D. T., Ferralis, N., Williford, K. H., Zhang, Y., Schuessler, J. A. and Summons, R. E. (2019) Organo-mineral associations in chert of the 3.5 Ga Mount Ada Basalt raise questions about the origin of organic matter in Paleoarchean hydrothermally influenced sediments. *Sci. Rep*. 9, 16712. DOI: 10.1038/s41598-019-53272-5

Beyssac, O., Goffé, B., Chopin, C. and Rouzaud, J. N. (2002) Raman spectra of carbonaceous material in metasediments: a new geothermometer. *J. Metamorph. Geol.* 20, 859–871. DOI: 10.1046/j.1525-1314.2002.00408.x

Derenne, S., Robert, F. and Skrzypczak-Bonduelle, A. (2008) Molecular evidence for life in the 3.5-billion-year-old Warrawoona chert. *Earth Planet. Sci. Lett.* 272, 476–480. DOI: 10.1016/j.epsl.2008.05.014

Duda, J. P., Thiel, V., Bauersachs, T., Mißbach, H., Reinhardt, M., Schäfer, N., Van Kranendonk, M. J. and Reitner, J. (2018) Ideas and perspectives: hydrothermally driven redistribution

and sequestration of early Archaean biomass – the “hydrothermal pump hypothesis.” *Biogeosciences* 15, 1535–1548. DOI: 10.5194/bg-15-1535-2018

Glikson, M., Duck, L. J., Golding, S. D., Hofmann, A., Bolhar, R., Webb, R., Baiano, C. F. and Sly, L. I. (2008) Microbial remains in some earliest Earth rocks: Comparison with a potential modern analogue. *Precambrian Res.* 164, 187–200. DOI: 10.1016/j.precamres.2008.05.002

Igisu, M., Ueno, Y. and Takai, K. (2018) FTIR microspectroscopy of carbonaceous matter in ~3.5-Ga seafloor hydrothermal deposits in the North Pole area, Western Australia. *Prog. Earth Planet. Sci.* 5, 85. DOI: 10.1186/s40645-018-0242-1

Kitajima, K., Maruyama, S., Utsunomiya, S. and Liou J. G. (2001) Seafloor hydrothermal alteration at an Archaean mid-ocean ridge. *J. Metamorph. Geol.* 19, 583–599. DOI: 10.1046/j.0263-4929.2001.00330.x

Kouketsu, Y., Mizukami, T., Mori, H., Endo, S., Aoya, M., Hara, H., Nakamura, D. and Wallis, S. (2014) A new approach to develop the Raman carbonaceous material geothermometer for low-grade metamorphism using peak width. *Isl. Arc* 23, 33–50. DOI: 10.1111/iar.12057

Lowe, D. R. (1994) Abiological origin of described stromatolites older than 3.2 Ga. *Geology* 22, 387–390. DOI: 10.1130/0091-7613(1994)022<0387:AOODSO>2.3.CO;2

M

McCollom, T. M, Ritter, G. and Simoneit, B. R. T. (1999) Lipid synthesis under hydrothermal conditions by Fischer-Tropsch-type reactions. *Orig. Life Evol. Biosph.* 29, 153–166. DOI: 10.1023/A:1006592502746

McCollom, T. M. and Seewald, J. S. (2006) Carbon isotope composition of organic compounds produced by abiotic synthesis under hydrothermal conditions. *Earth Planet. Sci. Lett.* 243, 74–84. DOI: 10.1016/j.epsl.2006.01.027

Morag, N., Williford, K. H., Kitajima, K., Philippot, P., Van Kranendonk, M. J., Lepot, K., Thomazo, C. and Valley, J. W. (2016) Microstructure-specific carbon isotopic signatures

of organic matter from ~3.5 Ga cherts of the Pilbara Craton support a biologic origin. *Precambrian Res.* 275, 429–449. DOI: 10.1016/j.precamres.2016.01.014

Noffke, N., Christian, D., Wacey, D. and Hazen, R. P. (2013) Microbially induced sedimentary structures recording an ancient ecosystem in the ca. 3.48 billion-year-old Dresser Formation, Pilbara, Western Australia. *Astrobiology* 13, 1103–1124. DOI: 10.1089/ast.2013.1030

Pasteris, J. D. (1989) In situ analysis in geological thin-sections by laser Raman microprobe spectroscopy: A cautionary note. *Appl. Spectrosc.* 43, 567–570. DOI: 10.1366/0003702894202878

Shen, Y., Farquhar, J., Masterson, A., Kaufman, A. J. and Buick, R. (2009) Evaluating the role of microbial sulfate reduction in the early Archean using quadruple isotope systematics. *Earth Planet. Sci. Lett.* 279, 383–391. DOI: 10.1016/j.epsl.2009.01.018

Takeichi, Y., Inami, N., Suga, H., Miyamoto, C., Ueno, T., Mase, K., Takahashi, Y. and Ono, K. (2016) Design and performance of a compact scanning transmission X-ray microscope at the Photon Factory. *Rev. Sci. Instrum.* 87, 013704. DOI: 10.1063/1.4940409

Takeichi, Y., Inami, N., Suga, H., Ono, K. and Takahashi, Y. (2014) Development of a compact scanning transmission X-ray microscope (STXM) at the photon factory. *Chem. Lett.* 43, 373–375. DOI: 10.1246/cl.130948

Ueno, Y., Isozaki, Y., Yurimoto, H. and Maruyama, S. (2001) Carbon isotopic signatures of individual Archean microfossils(?) from Western Australia. *Int. Geol. Rev.* 43, 196–212. DOI: 10.1080/00206810109465008

Ueno, Y., Ono, S., Rumble, D. and Maruyama, S. (2008) Quadruple sulfur isotope analysis of ca. 3.5 Ga Dresser Formation: New evidence for microbial sulphate reduction in the early Archean. *Geochim. Cosmochim. Acta* 72, 5675–5691. DOI: 10.1016/j.gca.2008.08.026

Ueno, Y., Yamada, K., Yoshida, N., Maruyama, S. and Isozaki, Y. (2006) Evidence from fluid inclusions for microbial methanogenesis in the early Archaean era. *Nature* 440, 516–519. DOI: 10.1038/nature04584

Ueno, Y., Yoshioka, H., Maruyama, S. and Isozaki, Y. (2004) Carbon isotopes and petrography of kerogens in ~3.5-Ga hydrothermal silica dikes in the North Pole area, Western Australia. *Geochim. Cosmochim. Acta* 68, 573–589. DOI: 10.1016/S0016-7037(03)00462-9

Van Kranendonk, M. J. (2006) Volcanic degassing, hydrothermal circulation and the flourishing of early life on Earth: A review of the evidence from c. 3490–3240 Ma rocks of the Pilbara Supergroup, Pilbara Craton, Western Australia. *Earth Sci. Rev.* 74, 197–240. DOI: 10.1016/j.earscirev.2005.09.005

Van Kranendonk, M. J., Philippot, P., Lepot, K., Bodorkos, S. and Pirajno, F. (2008) Geological setting of Earth's oldest fossils in the ca. 3.5 Ga Dresser Formation, Pilbara Craton, Western Australia. *Precambrian Res.* 167, 93–124. DOI: 10.1016/j.precamres.2008.07.003

Walter, M. R., Buick, R. and Dunlop, J. S. R. (1980) Stromatolites 3,400–3,500 Myr old from the North Pole area, Western Australia. *Nature* 284, 443–445. DOI: 10.1038/284443a0

Yui, T.-F., Huang, E. and Xu, J. (1996) Raman spectrum of carbonaceous material: A possible metamorphic grade indicator for low-grade metamorphic rocks. *J. Metamorph. Geol.* 14, 115–124. DOI: 10.1046/j.1525-1314.1996.05792.x