**The Nonesuch Formation Lagerstätte: A rare window into freshwater life one billion years ago**

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**Materials and methods with a detailed specimen list**

Samples from core and outcrop were processed at the University of Sheffield palynological processing facility by David Bodman and Charles Wellman with, additional samples processed at Boston College by Paul Strother. Samples were treated with dilute HCl and concentrated (ca. 52%) HF, water rinsed followed by heavy mineral separation and filtration through a 20µm mesh screen. The resultant residues were mounted on microscope slides using Evacite or glycerine jelly. All images are from a Zeiss Universal microscope fitted with a Leitz Apo 40x and a Zeiss Plan Apo 63x objective paired to a 1.3 NA condenser and a Nikon D700 full frame camera back. Raw files were imported into Photoshop™ using the Camera Raw module, which was used to set white balance and correct for exposure, in addition to some spot removal.

**Sample details for illustrated specimens**

Figure 2. Biological diversity in the Nonesuch assemblage. Sphaeromorphs and related forms. Scale bar in all images is10 *µ*m unless otherwise noted. Code in parentheses refers to England Finder coordinates.

a, *Leiosphaeridia crassa*. Well PC-1, depth 368 ft, slide PC1-368 (G38).

b, *Leiosphaeridia jacuta* Well WC-9, depth 1425 ft, NON18-66 (M38).

c, *Leiosphaeridia tenuissima*. WellDO-8, depth 1409, slide NON18-1 (V43).

d, *Leiosphaeridia minutissima*. Well DO-8, depth 1184 ft., slide NON18-13 (U39).

e, *Leiosphaeridia ternata.* Well DO-8, depth 1164, slide NON18-14.

f, cf. *Lophosphaeridium*. Well DO-8, depth 1409 ft., slide NON18-1 (S48).

g, *Nucellosphaeridium* sp. with a small circular patch/ spot. Well DO8, depth 1132 ft., slide DO8\_1132 (M43).

h, *Nucellosphaeridium* sp. with a large circular interior patch/ spot. Well PI-2, depth 642.5 ft., slide NON12-11.

i, Flat acritarch with sub-angular outline. DIC x63 image showing microbial perforations. Well DO-8, depth 1339 ft., NON18-5.

j. unnamed acritarch with a circular median laesura. Well PC-1, depth 303 ft, slide NON09-3B (E44).

k *Schizofusa* sp. Well DO-8, depth 1164, slide NON18-14.

l, *Zonosphaeridium* sp., in this case a darker, thick walled cyst preserved within a thin, somewhat delicate envelope. Well PI-2, depth759 ft., slide NON12-15.

m, *Nucellosphaeridium* sp with a granular envelope enclosing a blockish inner body. Well PI-2, depth 388 ft., slide NON12-10.

n, *Nucellosphaeridium* sp with a granular envelope enclosing a small inner body/spot. Well PI-2, depth 388 ft., slide NON12-10.

o, *Zonosphaeridium* sp. with large diffuse inner body. Well DO-8, depth 1184, slide NON18-13 (B30).

p, Ellipsoidal cell, cf. *Archaeoellipsoides*. Well WC-9, depth 1724 ft., slide NON18-49 (E40).

q, unnamed, large diffuse ovoid cell. Well DO-8, depth 1164.5, Slide NON18-14 (D44).

 r, Cylindrical form somewhat akin to *Germinosphaera*. Well WC-9, depth 1688 ft., slide NON18-51 (S34).

s, linear filament of *Archaeoellipsoides*-like cells. Well PC-1, depth 614 ft, slide NON09-12B (O30).

Figure 3. Case Studies – Some interesting taxa and their palaeoecological significance. All scale bars 10 *µ*m, unless otherwise indicated. Code in parentheses refers to England Finder coordinates.

a, *Valeria lophostriata*. Well DO-8, depth 1339 ft., slide NON18-5 (R36).

b, *V. lophostriata*. Well DO-8, depth 1154 ft., slide NON18-15 (R34)

c, *V. lophostriata* enlargement. Well DO-8, depth 1154 ft., slide NON18-15 (R34)

d, *Germinosphaera*-like specimen Well PI-2, depth 388 ft., slide NON12-10.

e, *Germinosphaera*. Well PC-1, depth 529 ft., slide NON09-9 (P48).

f, *Germinosphaera*. Well WPB-4, depth 396, slide NON09-31 (F30).

g, *Germinosphaera*-like specimen. Well PC-1, depth305 ft., Slide NON09-3B.

h, *Germinosphaera*-like specimen. Well WPB-4, depth 396, slide NON09-31 (Q31).

i, *Germinosphaera*. Well WPB-4, depth 396, slide NON09-31 (G37).

j, *Spuromoyeria* – like form. Well PC-1, depth 614 ft., slide NON09-12B (T36).

k, *Simia*-like form. Well PC-1, depth 614 ft., slide NON09-12B (F50).

l, perforated sphaeromorph. Well PC-1, depth 614 ft., slide NON09-12B (S34).

m, perforated protist. Well WPB-4, depth 396 ft., slide NON09-31 (N27).

Figure 4. Biological diversity in the Nonesuch assemblage. Cell clusters and simple multicellularity. Scale bar in all images is10 *µ*m unless otherwise noted. Code in parentheses refers to England Finder coordinates.

a, *Archaeoellipsoides*-like. Scale bar is 100 µm long. Well PC-1, depth 303 ft, slide NON09-3B.

b, *Synsphaeridium*. WellWPB-2, depth1430 ft, slide NON12-2.

c, *Symplassosphaeridium* sp. Well WC-9, depth 1892 ft., sample NON18-79 (W44).

d, cf. *Synsphaeridium.* Well WC-9, depth 1376, slide NON18-70 (O43).

e, two clusters. Well PC-1, depth 368 ft., slide PC1-368 (W43).

f, planar sheet of roughly isodiametric cells. Well DO-8, depth 1409 ft., slide NON18-1 (S48).

g, fragment of a planar sheet of cells. Well DO-8, depth 1339, slide NON18-5 (S50).

h, 3-celled cluster. Well WC-9, depth 1432 Ft., sample NON18-65 (H39).

i, linear array of large rounded cells. Well PC-1, depth 614 ft., slide NON9-12B (C34).

j, cf. *Gloeodiniopsis*. Well WC-9, depth 1688 ft., slide NON18-51 (D47).

Figure 5. Eukaryotic filaments and probable prokaryotes (cyanobacteria) in the Nonesuch assemblage. All scale bars 10 *µ*m, unless otherwise indicated. Code in parentheses refers to England Finder coordinates.

a, septate filament. Well PC-1, depth 285 ft., slide PC-1-285 (K31).

b, *Siphonophycus kestron*. Well DO-8, depth 1132 ft., slide NON18-16.

c, *Proterocladus* sp., Well WPB-4, depth 396 ft., slide NON09-31 (D37).

d, single branched axis. Well PC-1, depth 305 ft., slide NON09-3B (G35).

e, tapering axis. Well PC-1, depth 305 ft., slide NON09-3B.

f, pseudo branching. Well PC-1, depth 614 ft., slide NON9-12B.

g, *Siphonophycus* mat. Well WC-9, depth 1432 ft., slide NON18-65 (E3).

h, entangled sheaths. Well PC-1, depth 1132 ft., slide DO8\_1132 (Q41).

i, *Eohalothece lacustrinu*s. Well PC-1, depth 305 ft., slide NON09-3B.