Supplementary Information Captions

ESI 1: Transmission FTIR spectra from diatom mounted on polyimide loop (in air) and later mounted on CaF_2 window. Spectra from approximately same location within the chloroplast.

ESI 2: Visible and FTIR movies showing the rotation of two *Nitzschia* sp. cells mounted on a polyimide microloop recorded at the IRENI beamline. FTIR data were processed for and Amide I and SiO bands. Reconstructed intensities were displayed with a rainbow scale. Red and blue correspond to high and low integrated band areas, respectively.

ESI 3: Reconstructed 3D FTIR voxelated display of two *Nitzschia* sp. cells, processed for SiO, Amide I and CH. Movie shows full 3D viewing angles, recorded at the IRENI beamline.

ESI 4: Isosurface display of 3D FTIR image of two *Nitzschia sp.* cells processed for SiO and Amide I. Red shaded area indicates edge where remainder of cell was cut off from view. Isosurface offers smoother surface than voxelated version.

ESI 5: Tomography accessory installed on the stage of Agilent Cary 620 FTIR microscope. A micro-sample is held at the tip of the device and rotated below the microscope objective lens.

ESI 6: Isosurfaces and voxelated display of a 3D FTIR image of a portion of an *Entomoneis spp*. cell, processed for SiO (1080 cm^{-1}) and integrated area of CH bands. SiO semi-transparent surface shows outer volume of cell; chloroplast rich in lipid (CH) is visible within, and corresponds to chloroplast location in visible image, Figure 6 Panel A.

ESI 7: Undersampled proof-of-principle images. A) Visible image of individual cell mounted on polyimide microloop B) FTIR false-colour 2D mosaic images of integrated spectral bands: SiO (grey scale) overlaid with image processed for CH (rainbow scale). Red box denotes location captured in under-sampled tomographic data set (single tile, 6 views, 30° intervals). C) False-colour 3D FTIR images processed for CH, Amide I and SiO. Here, and in other tomographc images, pixels or voxels with very low band areas have been rendered as transparent for these rainbow scale images. D) Spectrum associated with a voxel in the chloroplast region. E) Axial cross-section processed for SiO (grayscale), overlaid with cross-section processed for CH (rainbow scale). Pink box in (C) CH cube indicates location of slice.

The ~0.7 mm metal shaft of the polyimide microloop with diatom was mounted into the lead chuck of a mechanical pencil with 12 faceted sides (Pentel Sharp™ P207 Mechanical Pencil). The pencil was placed flat on the microscope stage, permitting six non-redundant views at ~30° rotational angle intervals. A *Pleurosigma* sp. cell was selected as a test case for single tile tomographic imaging with the mechanical pencil sample holder. A visible image (ESI 7A) and a 2D FTIR mosaic image of the entire cell mounted on a microloop was acquired with the Agilent thermal source FTIR microscope. After each image acquisition and a manual rotation, the diatom was re-centred in the field of view of the microscope with the motorized stage. This particular cell was quite long (~375 μm) and a mosaic of 8×2 tiles was required to image its entirety, at 1.1 × 1.1 μm^2 pixels. A minimal tomographic data set was recorded on the central region containing a visibly pigmented chloroplast using the 6 non-redundant viewing angles available. Three views of the reconstructed 3D image, processed for SiO, Amide I and CH, are shown in ESI 7 C, D.