

1 **Mineral physical protection and carbon stabilization in-situ evidence**
2 **revealed by nano scale 3-D tomography**

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14
15 **Abstract**

16 An approach for nano scale 3-D tomography of organic carbon (OC) and
17 associated mineral nano particles was developed to illustrate their spatial distribution and
18 boundary interplay, using synchrotron-based transmission X-ray microscopy (TXM). The
19 proposed 3-D tomography technique was first applied to in-situ observation of a lab-made
20 consortium of black carbon (BC) and nano mineral (TiO₂, 15 nm), and its performance
21 was evaluated under dual-scan absorption contrast and phase contrast modes. Then this
22 novel tool was successfully applied to a natural OC-mineral consortium from high
23 mountain soil at a spatial resolution down to 60 nm, showing the fine structure and
24 boundary of OC, distribution of abundant minerals at nano size, and in-situ 3-D organo-
25 mineral association. The stabilization of aged natural OC was found mainly attributed to

1 the physical protection of nano size Fe-containing minerals (Fe oxyhydroxides including
2 ferrihydrite, goethite, and lepidocrocite), and the strong organo-mineral complexation. In-
3 situ evidence reveals abundant mineral nano particles, in dense thin layers or nano-
4 aggregates/clusters, instead of crystalline clay-size minerals on or near OC surfaces. The
5 key working minerals for C stabilization are essentially reactive short-range-order (SRO)
6 mineral nano particles and/or poorly crystalline submicron size clay minerals. The
7 sorption of OC to SRO mineral (or vice versa) through organo-mineral multiple complex
8 bonds could occupy and consume their respective reactive surface sites, leading to
9 enhanced stabilization. The ubiquity and abundance of mineral nano particles on OC
10 surface, and their heterogeneity in natural environment could have been seriously
11 underestimated by traditional study approach. Our in-situ description of organo-mineral
12 interplay at nano scale provides direct evidence to substantiate the importance of mineral
13 physical protection for OC long term stabilization. This high resolution 3-D tomography
14 tool is promising for new insight on the interior 3-D structure of micro-aggregates, in-situ
15 organo-mineral interplay, and the fate of mineral nano particles including heavy metals in
16 natural environment.

17

18 **Introduction**

19 Mineral association with organic carbon (OC) may be an important stabilization
20 mechanism for carbon long-term sequestration, yet little is known about their in-situ
21 interplay and extent of association on aggregation level either chemically or physically
22 (Baldock and Skjemstad, 2000; Cusack et al., 2012; Mikutta et al., 2006; Torn et al., 1997;

1 Vogel et al., 2014). Traditional fractionation methods based on size and external force for
2 dissecting the association strength between OC and minerals in soils are limited to bulk
3 sample. High resolution information and in-situ knowledge is required for interpretation of
4 fractionation results and modeling (Kaiser et al., 2002; Kleber et al., 2007; Sollins et al.,
5 2009). Nano scale two-dimensional isotopic mapping discovered that only a limited
6 amount of the clay-sized surfaces contributed to OC sequestration (Vogel et al., 2014).
7 Understanding OC interplay with minerals in the fine fraction warrants study in a three
8 dimensional way (Kinyangi et al., 2006; Lehmann et al., 2007; Lehmann et al., 2008;
9 Solomon et al., 2012). Detailed in-situ association information between OC and minerals
10 may lead to breakthrough on mineral physical protection mechanism for OC long term
11 stabilization. To overcome the limitations of commonly used electron microscopic
12 methods (such as only on the surface layer, and undesirable artifacts due to
13 pretreatments), non-destructive high-resolution X-ray 3-D tomographic technique will be
14 used for exploring the fine structure of OC and boundary interplay with mineral nano
15 particles.

16 High resolution Synchrotron-based TXM has been demonstrated as a powerful
17 tool for understanding the internal 3-D structure of particles down to nano meter scale,
18 due to its large penetration depth and superior spatial resolution (Kuo et al., 2011; Wang
19 et al., 2015). This technique was successfully applied to reveal the discrete three
20 dimensional micro-aggregation structure of clay (kaolinite) in natural aqueous
21 environment, and generated remarkable tomography that revealed precise inter-particle
22 structure (Zbik et al., 2008). Clay particles with diameter below 500 nm were clearly visible

1 and their pseudo-hexagonal symmetry was recognized in details in a three dimensional
2 way.

3 The synchrotron-based TXM at the beamline BL01B1 of Taiwan Light Source
4 (TLS), which has been used in this study, provides two-dimensional imaging and three-
5 dimensional tomography at a spatial resolution of 30/60-nm with tunable energy (8-
6 11keV). It provides unprecedented opportunity for studying OC boundary interplays with
7 mineral particles at nano meter scale. Two image acquisition modes, absorption contrast
8 and phase contrast, can be used alternatively for recognizing OC and nano minerals.
9 Conventionally, X-ray images are taken in the absorption contrast mode, and the resulting
10 image contrast only depends on the difference of X-ray attenuation coefficient between
11 materials. This mode is especially useful for materials consisted of high atomic number
12 compositions. However, in organic materials, the difference of X-ray attenuation
13 coefficients between specimen and air is too small to distinguish each other. For this
14 reason, the structure of organic materials is often hard to be recognized due to low
15 contrast in absorption contrast images. Alternatively, phase contrast technique transfers
16 optical path length differences (optical phase) inside specimens into intensity contrast,
17 can be used for imaging low atomic number materials, which are poor to absorb X-rays.
18 It provides a unique opportunity to observe fine structures of organic specimens such as
19 OC. Little study has been done on OC and mineral nano particles using high-resolution
20 3-D X-ray tomography, though non-synchrotron-based 3-D X-ray microscopy was used
21 to observe occluded carbon in phytolith structure and kerogen in micrometer scale
22 (Alexandre et al., 2015; Bousige et al., 2016). We aim to develop a new dual-scan method

1 using phase contrast and absorption contrast modes of the TXM alternatively for the
2 observation of OC and mineral consortiums inside lab-made and natural samples in nano
3 meter scale. Lab-made OC in forms of black carbon (BC) will be examined in the artificial
4 consortium with added nano mineral (TiO₂) particles using synchrotron-based TXM for
5 the first time.

6 Black C/biochar has received increasing research interest globally due to its
7 importance in global carbon cycling, soil fertility improvement and environmental pollutant
8 remediation (Bond et al., 2013; Jeffery et al., 2015; Kuhlbusch, 1998; Lehmann et al.,
9 2007; Liang et al., 2006; Liang et al., 2008; Schmidt, 2004). On top of method
10 development for 3-D tomography at nano meter scale, this study provides in-situ
11 evidences on the minerals physical protection on natural OC, and to explore the C
12 stabilization mechanism in natural soil.

13

14 **Methodology**

15 **Sample preparation and background**

16 Black C was made in lab using leguminous plant (*Sesbania roxburghii*) of 80 days'
17 harvest, which was first oven-dried (65 °C) and charred inside a muffle furnace at 300 °C
18 in loosely sealed stainless containers (Chen et al., 2014b). This consortium of low
19 temperature BC and mineral nano particles was constructed by dry deposition of
20 commercial TiO₂ (15 nm) on lab-made BC (3 mm chunk), and then embedded in Gatan
21 G-1 epoxy. The block were cross sectioned to a thickness of 100 to 200 µm using a

1 microtome (Leica Reichert Ultracut E ultra-microtome) and subsequently hand-polished
2 to a thickness of 30 to 50 μm . Each section was transferred onto Kapton tape and
3 mounted on a stainless steel sample holder for TXM observation. Before TXM analysis,
4 gold nano particles (50-150 nm or 400-500 nm in diameter) were deployed on the section
5 surface for image registration.

6 Thin section of natural OC and mineral consortium (NH) was prepared using
7 micron to millimeter size particulate sample from high mountain soil. Particulate organic
8 matter of mm-size with minerals embedded inside was taken from the lower dark layer at
9 depth of 72-93 cm in a Typic Humicryepts soil profile, located in Mt. Nanhua, Nantou
10 County, Taiwan (24°03'00", 121°17'02"). On top of this dark layer, iron stain was observed
11 within the depth of 63-72 cm in the profile. The soil has developed on top of sandstone
12 and slate, with some features of inceptisol and spodosol. The sampling elevation is 3092
13 m, the annual temperature is 7.57 °C, and the yearly rainfall is 2203.1 mm. The major
14 vegetation is arrow bamboo (*Yushania nittakayamensis*), with sporadic Hemlock (*Tsuga*
15 *chinensis*), fir (*Abies kawakamii*), and spruce (*Picea morrisonicola*).

16 The sequestration environment represents weak leaching and inactive chemical
17 weathering conditions. The age of soil organic C has been estimated to 3500 years B.P.

18

19 **Working conditions of TXM**

20 A superconducting wavelength shifter source provides a photon flux of 4×10^{11}
21 photons s^{-1} (0.1% bw) $^{-1}$ in the energy range of 5-20 KeV at the BL01B1 beamline. A

1 double crystal monochromator exploiting a pair of Ge (111) crystals selects X-rays within
2 the energy range of 8-11 KeV. The specimen is imaged using a Fresnel zone plate, which
3 is used as an objective lens for an image magnification of 44x by the first order diffraction
4 mode. Conjugated with a 20x downstream optical magnification, the TXM provides a total
5 magnification of 880x with a field of view of 15x15 μm^2 . By acquiring a series of 2D
6 images with the sample rotated 1° stepwise, 3-D tomography datasets is later
7 reconstructed based on 151 sequential image frames that are captured with azimuth
8 angle rotating from -75° to +75°.

9

10 **Image acquisition for 3-D tomography**

11 Under the most frequently used absorption contrast mode, 2-D images are
12 recorded based on the projection of the different X-ray absorption coefficient integration
13 along the optical pathway through samples on a detector. The absorption mode is useful
14 for materials of high absorption coefficient, such as minerals or high atomic number
15 materials, but it is poor for the observation of low atomic number materials, such as
16 organic or polymer materials. In order to recognize the OC structure more accurately, 2-
17 D/3-D images for the same sample region are recorded using absorption contrast and
18 phase contrast modes, respectively.

19 In the phase contrast mode, the gold-made phase ring positioned at the back-focal
20 plane of the zone plate is used to retard or advance the phase of the non-diffractive light

1 by $\pi/2$, generating (Zernike's) phase contrast images recording at the detector. The light
2 diffracted by specimen is interfered with the retarded non-diffractive light, generating
3 phase contrast image. The intensity difference in a phase contrast image shows the
4 combination of optical phase difference and absorption difference through specimens.
5 This ability is especially important for the observation of OC which has a low X-ray
6 absorption coefficient.

7

8 **3-D reconstruction and analysis**

9 Three dimensional tomography reconstruction is performed using homemade
10 software, which is coded based on iterative image registration (Faproma) (Wang et al.
11 2017) and filtered back projection (FBP) reconstruction algorithms. Firstly, a serial of
12 single TXM image captured from -75° to $+75^\circ$ at rotational increments of 1° is loaded to
13 do image registration automatically using Faproma algorithm. Finally, the reconstruction
14 is processed using FBP algorithm. The reconstructed dataset is exported in cross-
15 sections, and later used for 3-D visualization using *Amira*. The intensity contrast of
16 reconstructed datasets is inversed for better visualization. Compositions with higher
17 absorption coefficients are shown in higher intensity and those with low absorption
18 coefficients are shown in lower intensity. The exported cross-section of 3-D tomography
19 (reconstructed datasets) shows the real distribution details and boundary interplay of OC
20 and mineral particles. The final 3-D tomographic structures for visualization and
21 illustration are generated using *Amira* 3-D software for image post-process and
22 computation (Fig. S1).

1

2 **Elemental mapping by SEM-EDS**

3 For correlated spatial distribution of selected elements (C, O, Fe, Al etc) in natural
4 OC particles from the high mountain soil, a low-vacuum scanning electron microscope
5 (JEOL W-LVSEM, JSM-6360LV) equipped with an energy dispersive X-ray spectrometer
6 (Oxford EDS) and a cathodoluminescence (CL) image detector (Gatan mini-CL) was used
7 for elemental mapping, at an accelerating voltage of 15 KeV.

8

9 **X-ray Diffraction for Mineralogy**

10 To analyze the forms of minerals associated with natural OC, particulate OC (with
11 minerals on the surface and embedded inside) was grounded and injected into capillary
12 tubes (Special Glass 10, Hampton Research, CA) for synchrotron high resolution X-ray
13 diffraction analysis at 09A beamline at Taiwan Photon Source (TPS), which is equipped
14 with a set of high-resolution monochromator (HRM). The wavelength is 0.8266 Å at the
15 energy of 15 KeV. The XRD spectra were recorded under room temperature for 240s
16 accumulation time and specific X-ray diffraction peaks and patterns were assigned ICDD
17 using PDF-2/4 program.

18

19 **Carbon functionality and interfacial mineral forms using SR-FTIR**

20 For FTIR analysis, mineral-bearing OC (NH) particles were grounded, dried (60 °C
21 overnight), and mixed with potassium bromide (KBr) at a ratio of 1:100, and molded into

1 disks using a hydraulic press. During the pressing process, a vacuum pump was used for
2 evacuating air and water. The samples were measured using Infrared
3 Microspectroscopy (IMS) at the BL14A1 beamline of the National Synchrotron Radiation
4 Research Center (NSRRC), Taiwan. The FTIR spectra were collected up to 1024 scans
5 in the mid-infrared range of 4000-400 cm^{-1} with a spectral resolution of 4 cm^{-1} , using a
6 FTIR spectrometer (Nicolet 6700, Thermo Fisher Scientific, Madison, WI, USA) with a
7 self-equipped light source. The automatic atmospheric suppression function in OMNIC
8 (OMNIC 9.2, 2012; Thermo Fisher Scientific Inc., Waltham, MA, USA) for bulk sample
9 analysis was activated for data analysis, to eliminate the rovibration absorptions of CO_2
10 and water vapor in ambient air.

11

12 **Results and Discussions**

13 **Distinguish the fine structure of BC and boundary interplay with mineral nano** 14 **particles**

15 High resolution 2-D X-ray photographs were captured for the identical regions in
16 lab-made BC and nano mineral consortium using dual-scan absorption contrast and
17 phase contrast modes (Fig. 1, a, e). The cross-section views exported from the
18 reconstructed 3-D datasets reveal subtle details of BC and mineral nano particles, and
19 clearly outline the fine boundary of BC and the distribution of TiO_2 nano particles (Fig. 1).
20 The shape, size, and distribution of mineral nano particles can be identified accurately
21 using absorption contrast mode due to their high X-ray absorptivity (Fig.1, b, c and d). In
22 comparison, the BC structure and contour of its boundary can be revealed much more

1 clearly using phase contrast mode (Fig.1, f, g and h). However, the bright halo artifacts in
2 phase-contrast image enhance the intensity of margin texture for nano minerals, and may
3 lead to overestimation of their volume (Fig. 1, e, f, g and h). Use of dual-scan mode allows
4 cross-checking of details and validation.

5 Cross-section views of the reconstructed 3-D tomography share consistent and
6 comparable features of BC and nano minerals in multi-angles (Fig. 2). According to the
7 display of different slicing planes (XY, XZ, YZ), it can be recognized that TiO₂ nano
8 particles deposit inside BC only sporadically contact with BC boundary (Fig. 2, b, e, c,
9 and f) due to the treatment of dry deposition. The nano scale gap between BC and nano
10 minerals has been clearly observed in absorption and phase-contrast images (Fig. 2, b,
11 e, c, and f). It is feasible to calculate the interplay surface and mineral volume
12 quantitatively by examining each cross-section views in a selected region. Our approach
13 is a success in thorough exploration of OC and minerals 3-D distribution, and verification
14 of their real in-situ spatial correlation under nano scale resolution.

15

16 **3-D tomography for illustrating in-situ distribution of BC and mineral nano particles**

17 3-D tomography for visualization has been computed and generated to illustrate
18 the spatial correlation between BC and minerals based on post-process of reconstructed
19 3-D datasets. Unprecedented details of 3-D in-situ distribution of BC and mineral nano
20 particles are revealed in computed 3-D tomography (Fig. 3; Fig. SMOV1, 2). Results from
21 absorption mode and phase contrast mode are consistent and comparable. The fine

1 boundary feature of BC is contoured to more completeness in the phase contrast mode.
2 The OC was rendered by transparent mode and high absorptivity materials (such as
3 minerals and gold particles) were rendered by solid mode with various colors. All
4 renderings are combined to visualize their interaction. The illustration of 3-D computed
5 tomography allows randomly tilted and set angles for image and animated video exports,
6 thus any region of interest inside a specimen may be explored thoroughly.

7 The lab-made consortium was successfully tested by this dual-scan methodology
8 using both absorption contrast and phase-contrast acquisition modes (Figs. 1, 2, 3). Low
9 temperature BC, which is more similar to natural OC (especially recalcitrant OC) than that
10 made at high temperature, was especially made to test its applicability under absorption
11 contrast mode. Results show that the fine structure and boundary of low temperature BC
12 can be clearly observed under absorption contrast mode. Thus for environmental OC
13 samples, the use of absorption contrast mode is probably sufficient for capturing organo-
14 mineral features.

15 Different from field samples, the minerals observed within the lab-made consortium
16 often distribute in clusters and are only sparsely in association with BC surface. The
17 preservation of plant-like structures in BC could play a role for carbon stabilization in
18 natural environment, as their porosity and reactive surface provide large areas and sites
19 for mineral coating, which may contribute to their long residence and physical endurance
20 (Eusterhues et al., 2008; Rasmussen et al., 2005; Rawal et al., 2016).

21

1 **Interplay of OC and minerals and C stabilization in high mountain soil**

2 Nano scale 3-D tomography revealed a high heterogeneity within natural OC-
3 mineral consortium, and most of the particulate OC surface is coated by minerals. Natural
4 OC exhibited strong organo-mineral association on its surface at nano scale (Fig. 4; Fig.
5 SMOV3). Abundant SRO minerals in forms of subhedral particle or anhedral nano-
6 aggregates/clusters have direct association with the boundary of OC, and develop coating
7 on the tracheid surface (Fig. 4 b and c) (Mikutta et al., 2006). Sheet-like mineral coating
8 is observed on OC surface, which are dense and thin layers, likely originated from
9 absorption (Fig. 4 b, Fig. S2). The densely-packed mineral texture suggested significant
10 physical protection on OC surface (Kaiser and Guggenberger, 2007). The sorbed
11 minerals not only form physical protection, but could also shield OC from chemical
12 weathering (Mikutta et al., 2006). The key working minerals for OC-mineral interplay are
13 essentially SRO mineral nano particles, and/or poorly crystalline submicron size clay
14 minerals, instead of crystalline, micron/clay-size minerals. Another distinct texture is
15 recognized as nano-aggregates/clusters of various shapes, indicating possible OC-
16 mineral co-precipitation (Fig. 4 b, Fig. S3). Mineral aggregation by poorly crystalline nano
17 particles renders natural sub-micron porosity, which may contribute to elevated sorption
18 capacity in soil (Rawal et al., 2016).

19 The nature of associated minerals was confirmed to be mainly SRO Fe
20 oxyhydroxides, specifically ferrihydrite (ICDD 01-073-8408), goethite (ICDD 01-073-
21 6522), lepidocrocite (ICDD 00-044-1415), and quartz (ICDD 00-033-1161) (Fig. 5; Table
22 S1), analyzed using high resolution synchrotron-based X-ray diffraction. Quartz may be

1 at most a minor component on OC surface, considering their chemistry and particle size.
2 Yet siliceous mineral surfaces may be coated with a veneer of hydrous Al- and Fe- oxides,
3 which could confer net positive charge and promote their reactivity in tropical
4 environments (Chen et al., 2014a; Sposito, 1989).

5 Considering their high surface area and reactivity, the abundant nano size Fe
6 oxyhydroxides could play a significant role for OC long-term stabilization through
7 chemical bonding and physical shielding (Eusterhues et al., 2005; Kaiser et al., 2002;
8 Kiem and Kogel-Knabner, 2002; Mikutta et al., 2006), and contribute to the longevity of
9 OC in high mountain soil. According to elemental mapping, aluminosilicates may also be
10 present, however, their portion and crystallization level should be low due to their minimal
11 signal in the XRD spectra (Figs. 5, 6). The FTIR analyses reveal the chemistry of organo-
12 mineral association (Fig. 7; Table S2). The aged OC is highly aromatic when at the same
13 time highly reactive, likely originated from pyrogenic C, as broad bands are observed
14 centered at 1596 cm^{-1} and 1706 cm^{-1} for aromatic C=C stretching and carboxylic $\nu\text{ C=O}$,
15 (Özçimen and Ersoy-Meriçboyu, 2010; Sharma et al., 2004). Both aromatic and carboxyl
16 C functional groups normally have high affinity with Fe (III) (Mikutta et al., 2007; Zhao et
17 al., 2016). Broad bands indicate a significant degree of association between OC and
18 minerals (Chen et al., 2016; Gu et al., 1994; Kaiser and Guggenberger, 2007). The
19 sorption of OC to Fe oxyhydroxides through organo-mineral multiple complex bonds could
20 occupy their reactive surface sites, and enhance their respective stabilization (Chorover
21 and Amistadi, 2001; Cornell and Schwertmann, 2006; Hall et al., 2016; Kaiser and
22 Guggenberger, 2007; Mikutta et al., 2007). The discovery of SRO mineral ferrihydrite in

1 air-dried OC particles and later ground-samples indirectly validates its stabilization due to
2 organo-mineral interplay. As a SRO/metastable mineral, ferrihydrite is hard to estimate
3 accurately in dry soil samples due to its transient nature and the limitation of traditional
4 extraction and spectroscopic methods (Cornell and Schwertmann, 2006). The specific
5 mineral phase in direct contact with OC on surface at nano scale warrants future study
6 (Fig. SMOV3). In-situ mineral mapping of different SRO minerals/Fe oxyhydroxide on OC
7 surface will provide mechanistic understanding on OC stabilization. Mineral physical
8 protection on OC may represent the end stage of carbon stabilization, especially in a
9 weak leaching and weathering environment.

10 Our in-situ description of organo-mineral interplay at nano scale provides direct
11 evidence on the importance of mineral physical protection for OC long term stabilization.
12 High amounts of ferrihydrite and other Fe oxyhydroxides were also found associated with
13 lignin-like OC in soil under an aquic moisture regime (Eusterhues et al., 2011). The
14 abundance of mineral nano particles, and their high heterogeneity and short-range-order
15 nature could be common in humid environment, however, they could have been seriously
16 underestimated by traditional analysis methods, such as electron microscopy, X-ray
17 diffraction and fractionation approaches, which focus on clay-size minerals (Mikutta et al.,
18 2005). Mineral physical protection for OC stabilization may be more important than
19 previous understanding. More research is proposed to explore: i) whether it is a general
20 phenomenon that the minerals interact with OC are essentially SRO mineral nano particle
21 and or poorly crystalline submicron size clay minerals, ii) whether the major mineral for C
22 stabilization is nano size SRO minerals instead of clay-size minerals in soils. Perspective

1 on C stabilization and saturation should be revolutionized to take the role of SRO minerals
2 into account in modelling soil C dynamics, in addition to parameters such as clay type
3 and content.

4 In summary, a high resolution 3-D tomography tool is required for exploring the in-
5 situ interplay of OC and mineral nano particles in natural environment. Nano scale 3-D
6 tomography provides direct evidence and new insight for the mineral physical protection
7 mechanism of OC in soil. This high resolution 3-D tomography approach is a promising
8 technique for probing the multi interfacial features between OC and minerals in lab and
9 field samples, and may provide new perspective on the fate of nano particles including
10 heavy metals in natural environments.

11

12

13 **Figure Captions**

14 **Figure 1.** The 2-D X-ray images for BC and mineral nano particle consortium. Two images
15 are taken for the same region using absorption contrast mode **(a)** and phase contrast
16 mode **(e)**, respectively. Cross-section views of the reconstructed 3-D tomography under
17 each mode at different depths relative to the position of gold nano particle along Z-axis
18 as a reference. **(b)** and **(f)** are sections extracted at the position of the gold particle. **(c)**
19 and **(g)** are sections extracted at 800 nm above the gold particle. **(d)** and **(h)** are sections
20 extracted at 800 nm below the gold particle. The scale bar is 5 μm .

1 **Figure 2.** Three-directional orthogonal sections of lab-made BC and mineral nano particle
2 consortium. The upper row sections are extracted from absorption contrast tomography
3 **(a, b, c)**, and the lower row sections are extracted from phase contrast tomography **(d, e,**
4 **f)**, specifically **(a)** and **(d)** are for XY plane, **(b)** and **(e)** are for YZ plane, and **(c)** and **(f)**
5 are for XZ plane. The scale bar is 5 μm .

6 **Figure 3.** 3-D tomography illustration of lab-made BC and mineral nano particle
7 consortium observed at -45° **(a, d)**, 0° **(b, e)**, and $+45^\circ$ **(c, f)** azimuthal viewing angles
8 under absorption contrast **(a, b, c)** and phase contrast mode **(d, e, f)**. The scale bar is 5
9 μm .

10 **Figure 4.** Three-directional orthogonal sections of high mountain mineral-bearing OC
11 from absorption contrast tomography **(a)** for XY plane, **b** for XZ plane, and **c** for YZ plane). .
12 The scale bar is 5 μm . Minerals mainly present two types of textures, subhedral particles
13 and anhedral nano-aggregates. The lower row images highlight the free surface of
14 specimen (red line in **d**), the boundary of OC (green dotted-line in **e**), and the subhedral
15 mineral particles (pink arrow in **e** and **f**)

16 **Figure 5.** The X-ray diffraction pattern of minerals within OC particles from high mountain
17 soil. Highly reactive Fe oxyhydroxides are identified and denoted with lines of different
18 colors: ferrihydrite (ICDD 01-073-8408, orange), goethite (ICDD 01-073-6522, blue), and
19 lepidocrocite (ICDD 00-044-1415, green). Q stands for Quartz (ICDD 00-033-1161).
20 Details are included in Table S1.

1 **Figure 6.** Elementary mapping by SEM-EDS for mineral-bearing OC from high mountain
2 soil. Left: SEM backscattering image (The bright spots inside are gold nano particles for
3 coating). Right: Elemental mapping of C, O, Fe and Al. Scale bars are 20 μm .

4 **Figure 7.** The FTIR spectra for the chemistry of organo-mineral association. The aged
5 OC is highly aromatic (1596 and 1386 cm^{-1}), and highly reactive with obvious carboxyl
6 functional group (1706 cm^{-1}). The broad bands point to possible significant degree of
7 association between OC and minerals. Some minor bands near 1274 , 1062 , 1024 , and
8 989 cm^{-1} indicate the lignin-derived nature of OC. Those bands near 476 , 534 , 798 , 910
9 and 1025 cm^{-1} have similar characteristics of soil inorganic/mineral matrix. More details
10 are included in Table S2.

11

12

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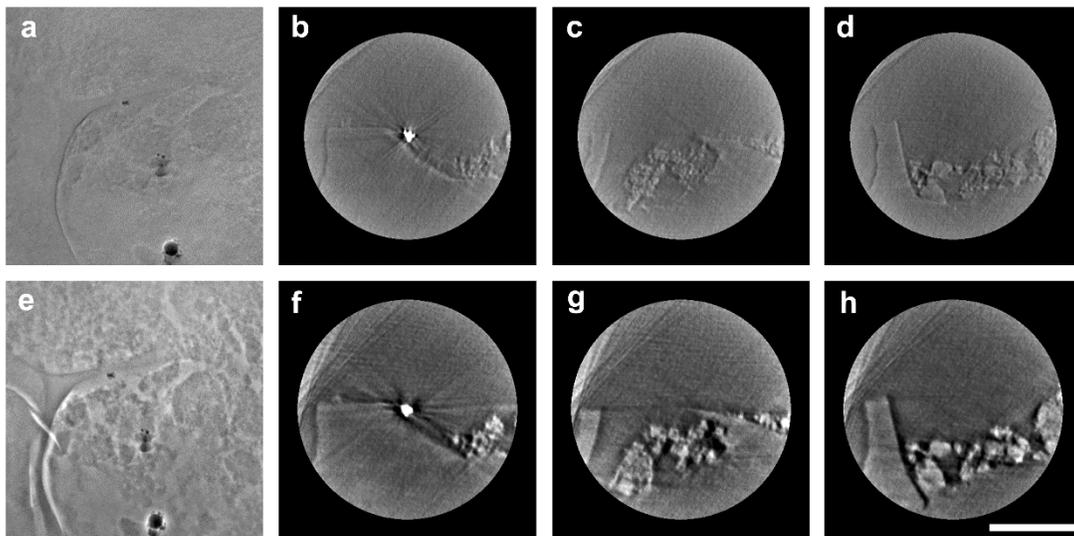
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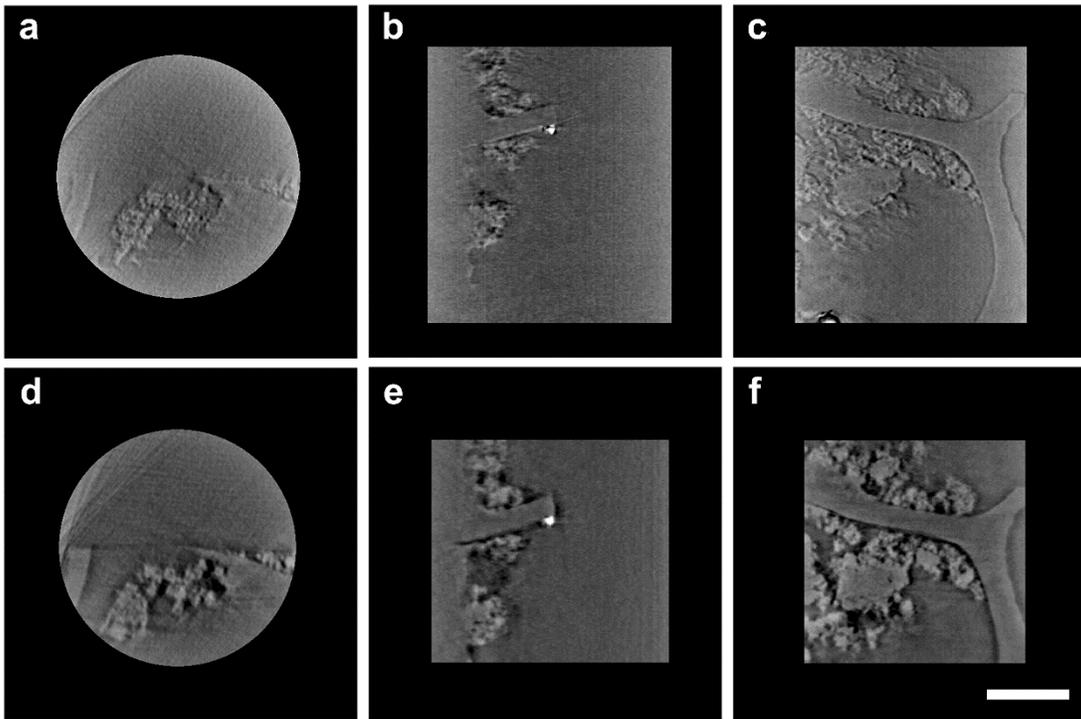
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Figure 1.

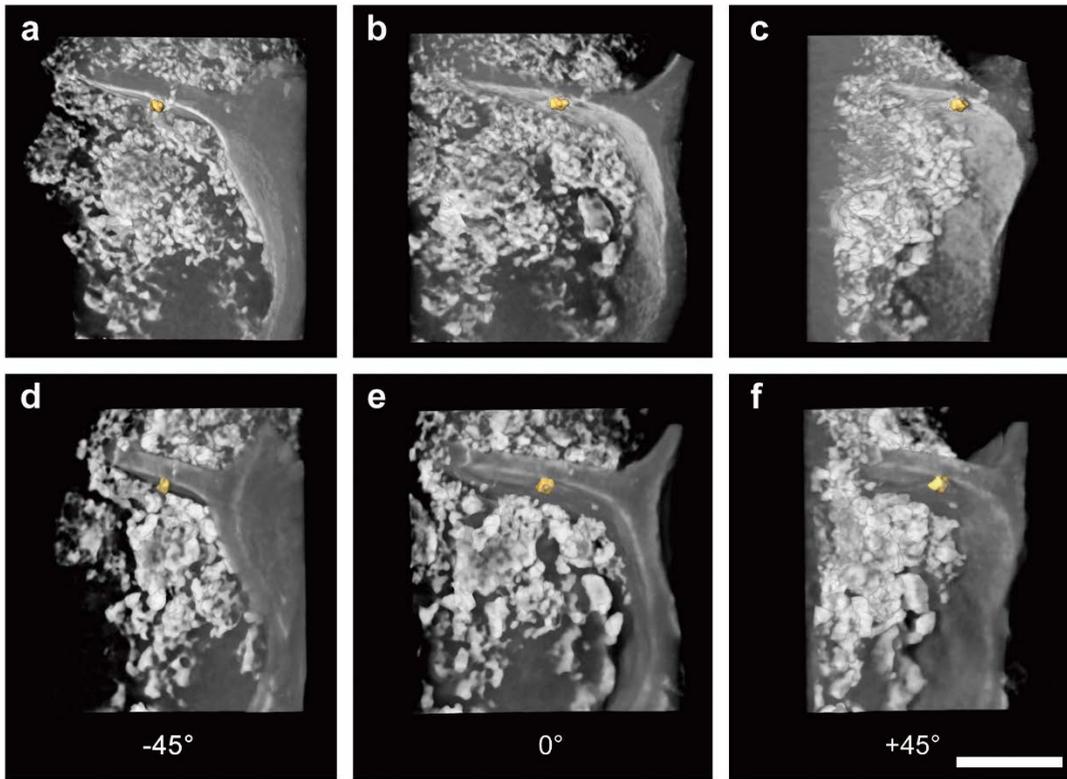


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2 **Figure 2.**

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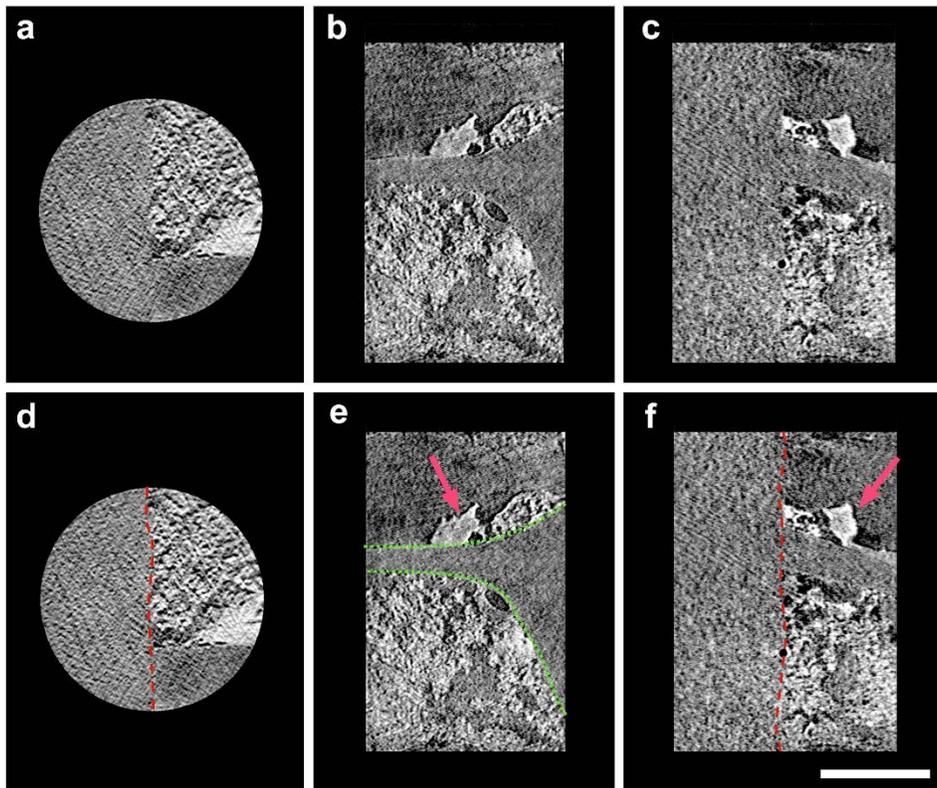
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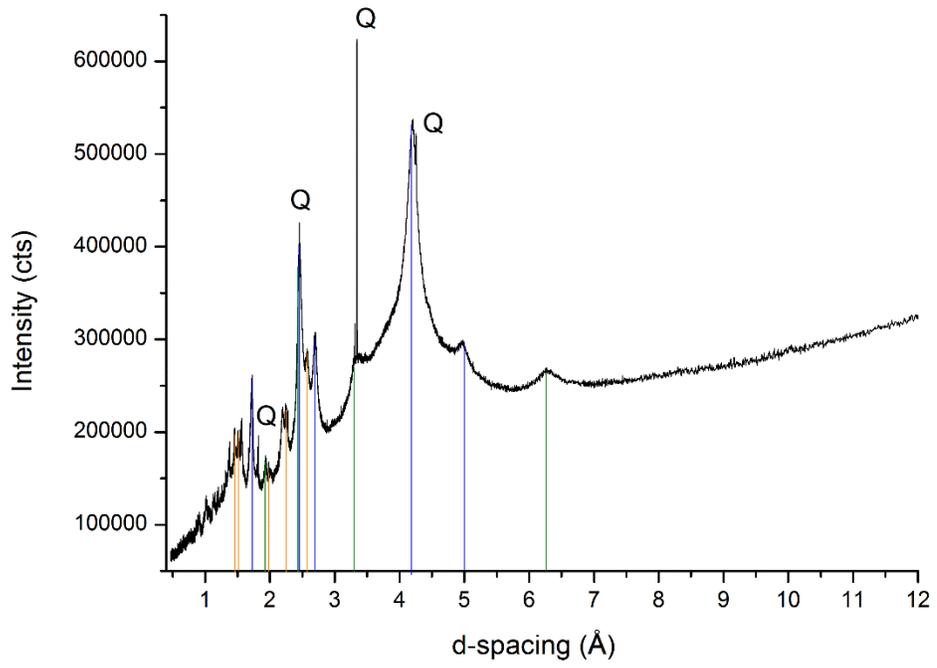
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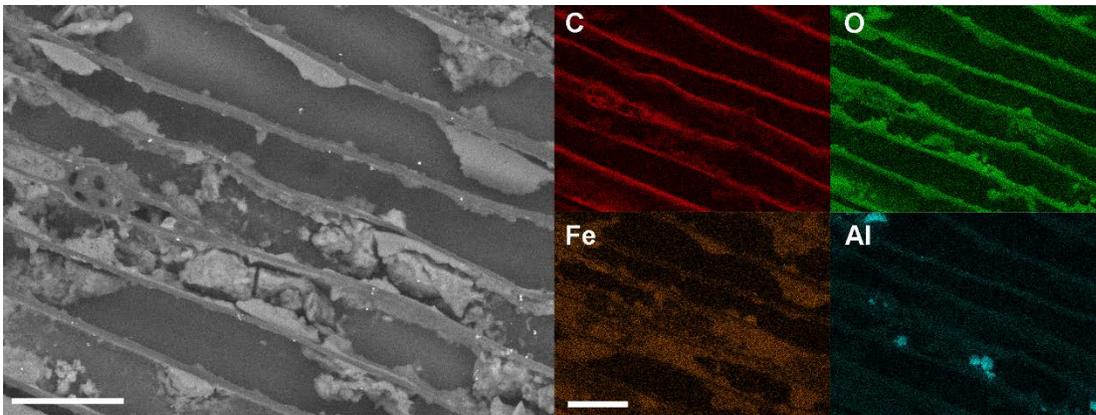


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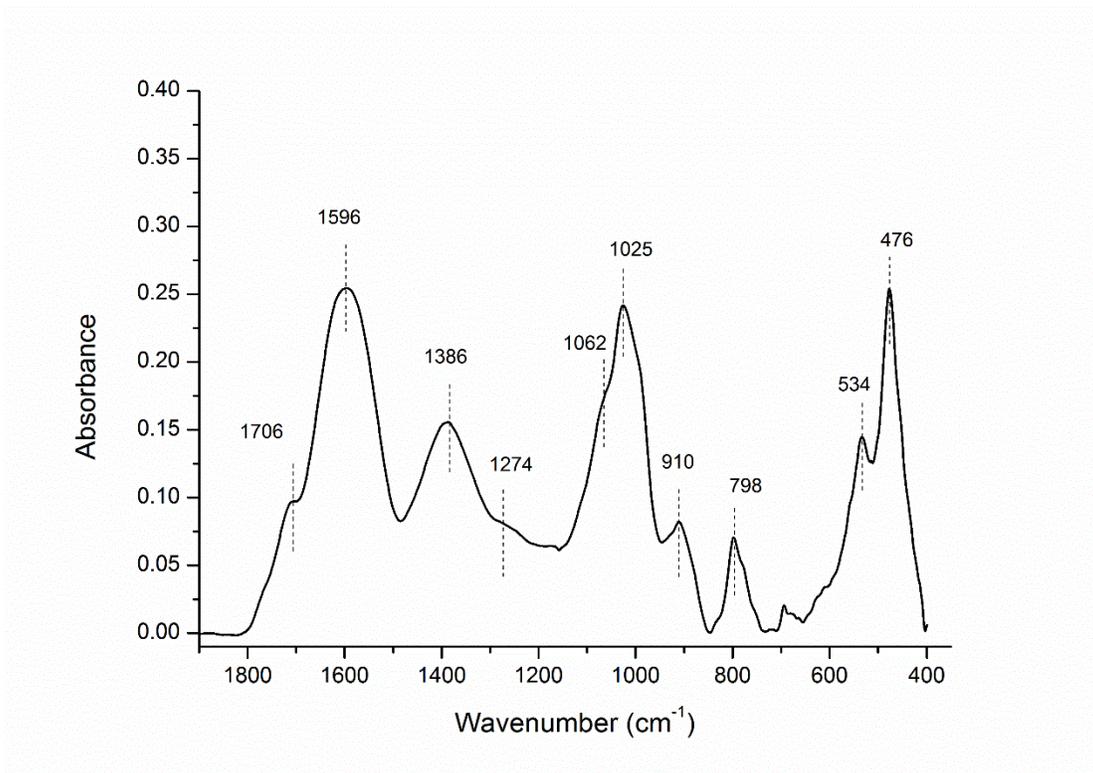
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6 **Figure 6.**



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2 **Figure 7.**

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