

Chapter 17

Carbon storage and DNA adsorption in allophanic soils and paleosols*

Yu-Tuan Huang¹, David J. Lowe¹, G. Jock Churchman², Louis A. Schipper¹, Nicolas J. Rawlence³,
Alan Cooper⁴

¹Department of Earth and Ocean Sciences, University of Waikato, Private Bag 3105, Hamilton 3240, New Zealand yh224@students.waikato.ac.nz

²School of Agriculture, Food and Wine, University of Adelaide, Adelaide, SA 5005, Australia

³Allan Wilson Centre for Molecular Ecology and Evolution, Department of Zoology, University of Otago, PO Box 56, Dunedin 9054, New Zealand

⁴Australian Centre for Ancient DNA, School of Earth and Environmental Science, University of Adelaide, Adelaide, SA 5005, Australia

Abstract

Andisols and andic paleosols dominated by the nanocrystalline mineral allophane sequester large amounts of carbon (C), attributable mainly to its chemical bonding with charged hydroxyl groups on the surface of allophane together with its physical protection in nanopores within and between allophane nanoaggregates. C near-edge X-ray absorption fine structure (NEXAFS) spectra for a New Zealand Andisol (Tirau series) showed that the organic matter (OM) mainly comprises quinonic, aromatic, aliphatic, and carboxylic C. In different buried horizons from several other Andisols, C contents varied but the C species were similar, attributable to pedogenic processes operating during developmental upbuilding, downward leaching, or both. The presence of OM in natural allophanic soils weakened the adsorption of DNA on clay; an adsorption isotherm experiment involving humic acid (HA) showed that HA-free synthetic allophane adsorbed seven times more DNA than HA-rich synthetic allophane. Phosphorus X-ray absorption near-edge structure (XANES) spectra for salmon-sperm DNA and DNA-clay complexes indicated that DNA was bound to the allophane clay through the phosphate group, but it is not clear if DNA was chemically bound to the surface of the allophane or to OM, or both. We plan more experiments to investigate interactions among DNA, allophane (natural and synthetic), and OM. Because DNA shows a high affinity to allophane, we are studying the potential to reconstruct late Quaternary palaeoenvironments by attempting to extract and characterise ancient DNA from allophanic paleosols.

Keywords: Andisols, allophane, carbon sequestration, C NEXAFS, P XANES, ancient DNA

*Citation:

Huang, Y-T.; Lowe, D.J.; Churchman, G.J.; Schipper, L.; Rawlence, N.J.; Cooper, A. 2014. Carbon storage and DNA adsorption in allophanic soils and paleosols. *In:* Hartemink, A.E.; McSweeney, K. (eds). "Soil Carbon". Progress in Soil Science Series, Springer, New York, pp. 163-172. DOI : 10.1007/978-3-319-04084-4_17.

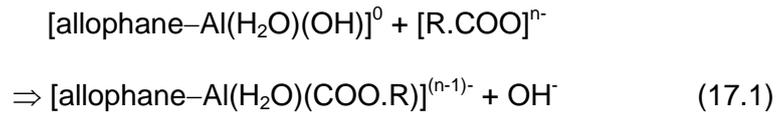
Introduction

Carbon storage in soils

Soils are the largest reservoir of terrestrial organic carbon (Batjes, 1996; Blanco-Canqui and Lal 2004), and soil organic matter (SOM) has become subjected to many diverse investigations because C is an important contributor to soil quality and because of its role as a potential store of CO₂. Nanocrystalline clay minerals including allophane and ferrihydrite have a greater affinity than crystalline clay minerals for organic functional groups, hence soil organic carbon (SOC), due to ligand exchange with their charged hydroxyl groups (Basile-Doelsch et al., 2005; Kleber et al., 2005; Churchman and Lowe, 2012). Soils developed mainly on free-draining, explosively erupted, fragmental volcanic material (collectively, 'tephra'), typically classed as Andisols, 'hold' more C than other mineral soils (Batjes, 1996; McDaniel et al., 2012) because of their constituent allophane (Chevallier et al., 2010; Calabi-Floody et al., 2011). Andisols occupy ~1 % of the world's land area, but contain 5% of global C (Dahlgren et al., 2004; McDaniel et al., 2012). Sequestration of organic materials in soils is an important research area, and we are specifically interested in DNA adsorption and storage by allophanic soils because preserved DNA or ancient DNA (aDNA) from buried paleosols could in turn provide a way of reconstructing past environments.

Allophane and carbon sequestration

Allophane comprises tiny (3.5–5 nm in diameter), Al-rich nanocrystalline hollow spherules ((1-2)SiO₂·Al₂O₃·(2-3)H₂O) (Fig. 17.1a) with extremely large surface areas (up to 1500 m² g⁻¹) (Parfitt, 2009; Calabi-Floody et al., 2011). Broken-bond defects in the spherules, also called perforations, are ~0.3 nm in diameter (Fig. 17.1a) and it is the protonation and deprotonation of (OH)Al(H₂O) groups exposed at these sites that give rise to allophane's variable (pH-dependent) charge (Parfitt et al., 1977; Yuan and Theng, 2012), and thus its anion exchange capacity. The adsorption of negatively charged humic substances on allophane through ligand exchange is represented by equation (17.1) (from Yuan and Theng, 2012). [R.COO]ⁿ⁻ denotes a humic molecule containing *n* carboxylate functional groups.



These high specific surface areas and surface properties help govern the strong association between allophane and SOM (Figs. 17.1b, c) (Buurman et al., 2007; Calabi-Floody et al., 2011; Yuan and Theng, 2012). In addition, allophane spherules tend to form nanoaggregates up to about 100 nm in diameter (Fig. 17.1d) (Chevallier et al., 2010; Calabi-Floody et al., 2011). We suggest that the interstices (nanopores) both within and between such nanoaggregates (Fig. 17.1d) provide a haven for SOM so that a portion of SOC is poorly accessible to microbes or enzymes and thus protected. Calabi-Floody et al. (2011) showed that significant SOC, which resisted the treatment of hydrogen peroxide, was strongly held by allophane and imogolite in Andisols, a finding consistent with studies of the physical protection and stabilisation of SOM within larger micro- and macro-aggregates (Elliott, 1986; Strong et al., 2004). It is possible that SOC is protected against the attack of enzymes in the narrowest interstices, such as in nanoaggregates, because the diffusion pathway for the passage of enzymes is constrained (McCarthy et al., 2008; Chevallier et al., 2010).

DNA adsorption on allophane

The adsorption of DNA on soil particles plays a major role in protecting extracellular DNA from degradation in soils (Paget and Simonet, 1994), and the interactions between DNA and common clay minerals have been investigated using FTIR spectroscopy (Cai et al., 2006a). Allophane adsorbs more DNA than other minerals in soils (Saeki et al., 2010; Harsh, 2012), and may be capable of preserving DNA as well. Greaves and Wilson (1970) suggested that nucleic acids in the centre of expandable montmorillonite crystal structures may be protected from attack by microbial enzymes, and hence we hypothesise that aDNA may also be physically protected and preserved within intra- and inter-nanoaggregate interstices of allophane in Andisols and andic paleosols. However, Cai et al. (2006b) revealed that DNA is more tightly adsorbed by organic matter-free clay than by clay containing organic matter, indicating that the abundant SOM in allophanic soils might hamper DNA adsorption on allophane. SOM is expected to increase the negative charge on

minerals (Equation 1), leading to a mutual exclusion between SOM-rich allophane and negatively-charged phosphate groups of DNA. In contrast to Cai et al. (2006b), Saeki et al. (2011) showed that humic acid had a high affinity for DNA, hence the influence of SOM on DNA (and potentially aDNA) adsorption in Andisol is still debatable.

A DNA strand comprises heterocyclic bases and a pentose-phosphodiester backbone, and it has been suggested that DNA binds to clays through the phosphate group (Cai et al., 2006a) but there was no direct evidence. We propose three main mechanisms for DNA adsorption on allophane: (1) direct chemical adsorption on the organic matter-free and protonated surface of allophane, (2) indirect chemical adsorption/dissolution on/into the SOM-rich allophane, and (3) physical adsorption in interstices/nanopores of allophane nanoaggregates (Figs. 1b-d). A clearer understanding of DNA adsorption on allophanic materials would help inform ways to extract preserved DNA or aDNA from buried paleosols.

Materials and methods

Application of synchrotron radiation

Traditionally SOM was extracted chemically from soils, leading to possible artifacts of the treatment (e.g. see Schmidt et al., 2011; Churchman and Lowe, 2012). Synchrotron radiation has been developed for several decades for studying materials (Sutton et al., 2006) including the chemical composition of soil components such as 'resistant' C (Wan et al., 2007; Lehmann and Solomon, 2010). It allows the analysis of C at low concentrations and within bulk soil samples. C near-edge X-ray absorption fine structure analysis (NEXAFS) enables examination of speciation and structures of SOM (Lehmann, 2005; Lehmann et al., 2007). Phosphorus X-ray absorption near-edge structure (XANES) reveals the configuration for phosphorus bonding to Fe(III)- or Al(III)-rich minerals (Kizewski et al., 2011).

Carbon NEXAFS

We used the Beamline 24A1 at the National Synchrotron Radiation Research Center (NSRRC), Hsinchu, Taiwan, for our C NEXAFS study of the C compositions of SOM associated with allophane through the electron yield mode. Contaminant C contributions from the beamline have been eliminated by a golden mesh anterior to the sample chamber.

Allophanic materials were collected in northern New Zealand from an Andisol of the Tirau series (a medial, mesic Typic Hapludand: Bakker et al., 1996) at Tapapa and from buried andic paleosols at two sites at Lake Rerewhakaaitu (near Mt Tarawera) and one near Lake Rotoaira (adjoining Tongariro National Park). Clay fractions (<2 μm) were pressed into indium foil to enable them to be conductive, and analysis was accomplished under 10^{-9} torr using a chemical pump.

DNA adsorption studies

Salmon-sperm DNA from Sigma-Aldrich Co. LLC that ranged from 0 to 300 μg was added to the natural clays from all sites and to a 1 mL suspension of pure synthetic allophane (synthesized following the method of Ohashi et al., 2002), and the mixture was gently rotated at room temperature for 3 h. DNA in the supernatant was analyzed at 260 and 280 nm spectrophotometrically.

Phosphorus XANES

P XANES spectra were obtained for pressed pellets of clays from Tirau soil, pure salmon-sperm DNA, and salmon-sperm DNA-clay complexes, at the Beamline 16A1 at NSRRC.

Results and discussion

Carbon NEXAFS

The clay fraction of the upper subsoil of the Tirau soil contains about 10% total C, and C NEXAFS spectra showed its SOM comprises quinonic, aromatic, aliphatic, and carboxylic C (Fig. 17.2a). Although C contents varied between horizons and sites, the compositions of SOM were similar in all horizons of the andic paleosols (data not shown). We attribute this compositional similarity to either

(1) the downward leaching of dissolved SOM through the profiles vertically, effectively homogenising the composition of organic matter in each horizon, or (2) integrative developmental upbuilding pedogenesis whereby the profiles have formed by incremental additions of thin distal tephra over the past ~25,000 years that have interacted with the soil organic cycle whilst temporarily at the land surface, the land/soil surface slowly rising at an average rate of ~5 mm per century (Lowe et al., 2012; McDaniel et al., 2012), or both (1) and (2).

DNA adsorption studies

There was a negative relationship between total C and DNA uptake by all soil samples. However, at pH 6.5, humic-acid-free synthetic allophane adsorbed salmon-sperm DNA at the rate of up to 26 µg/mg allophane, which is about seven times more than that for humic-acid-rich allophane (Fig. 17.3). Hence humic acid (or SOM) occupied the active sites on allophane spherules, limiting their capacity for DNA adsorption.

Phosphorus XANES

The spectra showed that the absorption edge (absorption energy, the sharply rising peak of the spectrum) of phosphorus for pure DNA was 2153 eV, and that binding of DNA and clay had induced a shift of the absorption edge to 2514 eV (Fig. 17.2b). This shift indicates that the phosphate group of DNA has changed chemically after binding to natural clay. However, it is unclear whether DNA was bound to clay or to SOM, which is apparently pervasive in clay fractions from andic materials, and further experiments are needed to evaluate how DNA reacts to SOM and to organic-matter-free allophane. An improved understanding of DNA adsorption in Andisols should indicate how we can best potentially extract aDNA from allophanic paleosols that date back through time.

Conclusions

(1) C NEXAFS spectra for an Andisol showed that OM comprises quinonic, aromatic, aliphatic, and carboxylic C. In other Andisols, C contents varied but C species were similar, attributable to pedogenic processes operating during developmental upbuilding, downward leaching, or both.

(2) The presence of OM in natural allophanic soils weakened adsorption of DNA on clay: HA-free synthetic allophane adsorbed seven times more DNA than HA-rich synthetic allophane.

(3) P XANES spectra for salmon-sperm DNA and DNA-clay complexes showed that DNA was bound to allophane clay through the phosphate group, either to the surface of allophane or to OM, or both.

Acknowledgements

This research was supported by the Marsden Fund (10-UOW-056) through the Royal Society of New Zealand. We thank NSRRC, Taiwan, and especially Dr Tsan-Yao Chen for technical instruction and support, Ling-Yun Jang for P XANES spectra for salmon-sperm DNA, Prof Shin-Ichiro Wada (Kyushu University) for advice on allophane synthesis, Dr Emma Summers, Janine Ryburn, and Lynne Parker (Waikato University) for help with experiments, and Prof Kevin McSweeney (University of Wisconsin–Madison) for reviewing the paper.

References

- Bakker L., Lowe D.J., and Jongmans A.G. (1996) A micromorphological study of pedogenic processes in an evolutionary soil sequence formed on Late Quaternary rhyolitic tephra deposits, North Island, New Zealand. *Quaternary International* 34-36: 249-261.
- Basile-Doelsch I., Amundson R., Stone W.E.E., Masiello C.A., Bottero J.Y., Colin F., Borschneck D., and Meunier J.D. (2005) Mineralogical control of organic carbon dynamics in a volcanic ash soil on La Réunion. *European Journal of Soil Science* 56: 689-703.
- Batjes N.H. (1996) Total carbon and nitrogen in the soils of the world. *European Journal of Soil Science* 47: 151-163.
- Blanco-Canqui H., and Lal R. (2004) Mechanisms of carbon sequestration in soil aggregates. *Critical Reviews in Plant Sciences* 23: 481-504.
- Buurman P., Peterse F., and Almendros Martin G. (2007) Soil organic matter chemistry in allophanic soils: a pyrolysis-GC/MS study of a Costa Rican Andosol catena. *European Journal of Soil Science* 58: 1330-1347.

- Cai P., Huang Q.-Y., and Zhang X.-W. (2006a) Interactions of DNA with clay minerals and soil colloidal particles and protection against degradation by DNase. *Environmental Science and Technology* 40: 2971-2976.
- Cai P., Huang Q., Jiang D., Rong X., and Liang W. (2006b) Microcalorimetric studies on the adsorption of DNA by soil colloidal particles. *Colloids and Surfaces B: Biointerfaces* 49: 49-54.
- Calabi-Floody M., Bendall J.S., Jara A.A., Welland M.E., Theng B.K.G., Rumpel C., and Mora M.L. (2011) Nanoclays from an Andisol: extraction, properties and carbon stabilization. *Geoderma* 161: 159-167.
- Chevallier T., Woignier T., Toucet J., and Blanchart E. (2010) Organic carbon stabilization in the fractal pore structure of Andosols. *Geoderma* 159: 182-188.
- Churchman G.J., and Lowe D.J. (2012) Alteration, formation, and occurrence of minerals in soils, in: P.M. Huang, Y. Li, M.E. Sumner (Eds.), *Handbook of Soil Sciences*. 2nd edition. Vol. 1: Properties and Processes. CRC Press, Boca Raton. pp. 20.1-20.72.
- Dahlgren R.A., Saigusa M., and Ugolini F.C. (2004) The nature, properties and management of volcanic soils. *Advances in Agronomy* 82: 113-182.
- Elliott E.T. (1986) Aggregate structure and carbon, nitrogen, and phosphorus in native and cultivated soils. *Soil Science Society of America Journal* 50: 627-633.
- Greaves M.P., and Wilson M.J. (1970) The degradation of nucleic acids and montmorillonite-nucleic-acid complexes by soil microorganisms. *Soil Biology and Biochemistry* 2: 257-268.
- Harsh J. (2012) Poorly crystalline aluminosilicate clay minerals, in: P.M. Huang, Y. Li, M.E. Sumner (Eds.), *Handbook of Soil Sciences*. 2nd edition. Vol. 1: Properties and Processes. CRC Press, Boca Raton. pp. 23.1-23.13.
- Kizewski F., Liu Y.-T., Morris A., and Hesterberg D. (2011) Spectroscopic approaches for phosphorus speciation in soils and other environmental systems. *Journal of Environment Quality* 40: 751-766.
- Kleber M., Mikutta R., Torn M.S., and Jahn R. (2005) Poorly crystalline mineral phases protect organic matter in acid subsoil horizons. *European Journal of Soil Science* 56: 717–725.

- Lehmann J. (2005) Near-edge X-ray absorption fine structure (NEXAFS) spectroscopy for mapping nano-scale distribution of organic carbon forms in soil: Application to black carbon particles. *Global Biogeochemical Cycles* 19: GB1013.
- Lehmann J., Kinyangi J., and Solomon D. (2007) Organic matter stabilization in soil microaggregates: implications from spatial heterogeneity of organic carbon contents and carbon forms. *Biogeochemistry* 85: 45-57.
- Lehmann J., and Solomon D. (2010) Organic carbon chemistry in soils observed by synchrotron-based spectroscopy, in: B. Singh and M. Gräfe (Eds.), *Synchrotron-based techniques in soils and sediments*. Elsevier, Amsterdam. pp. 289-312.
- Lowe D.J., Lanigan K.M., and Palmer D.J. (2012) Where geology meets pedology: Late Quaternary tephras, loess, and paleosols in the Mamaku Plateau and Lake Rerewhakaaitu areas. *Geoscience Society of New Zealand Miscellaneous Publication 134B*, pp. 2.1-2.45.
- McCarthy J., Ilavsky J., Jastrow J., Mayer L., Perfect E., and Zhuang J. (2008) Protection of organic carbon in soil microaggregates via restructuring of aggregate porosity and filling of pores with accumulating organic matter. *Geochimica et Cosmochimica Acta* 72: 4725-4744.
- McDaniel P.A., Lowe D.J., Arnalds O., and Ping C.-L. (2012) Andisols, in: P.M. Huang, Y. Li, M.E. Sumner (Eds.), *Handbook of Soil Sciences*. 2nd edition. Vol. 1: Properties and Processes. CRC Press, Boca Raton. pp. 33.29-33.48.
- Ohashi F., Wada S.-I., Suzuki M., Maeda M., and Tomura S. (2002) Synthetic allophane from high concentration solutions: nanoengineering of the porous solid. *Clay Minerals* 37: 451-456.
- Paget E., and Simonet P. (1994) On the track of natural transformation in soil. *FEMS Microbiology Ecology* 15: 109-118.
- Parfitt R.L. (2009) Allophane and imogolite: role in soil biogeochemical processes. *Clay Minerals* 44: 135-155.
- Parfitt R.L., Fraser A.R., and Farmer V.C. (1977) Adsorption on hydrous oxides. III. Fulvic acid and humic acid on goethite, gibbsite and imogolite. *Journal of Soil Science* 28: 289-296.
- Saeki K., Ihyo Y., Sakai M., and Kunito T. (2011) Strong adsorption of DNA molecules on humic acids. *Environmental Chemistry Letters* 9: 505-509.

- Saeki K., Sakai M., and Wada S.-I. (2010) DNA adsorption on synthetic and natural allophanes. *Applied Clay Science* 50: 493-497.
- Schmidt M.W.I., Torn M.S., Abiven S., Dittmar T., Guggenberger G., Janssens I., Kleber M., Kögel-Knabner I., Lehmann J., Manning D.A.C., Nannipieri P., Rasse D.P., Weiner S., and Trumbore S. (2011) Persistence of soil organic matter as ecosystem property. *Nature* 478: 49-56.
- Strong D.T., Wever H.D., Merckx R., and Recous S. (2004) Spatial location of carbon decomposition in the soil pore system. *European Journal of Soil Science* 55: 739-750.
- Sutton S.R., Caffee M.W., and Dove M.T. (2006) Synchrotron radiation, neutron, and mass spectrometry techniques at user facilities. *Elements* 2: 15-21.
- Wan J., Tyliczszak T., and Tokunaga T. (2007) Organic carbon distribution, speciation, and elemental correlations within soil microaggregates: Applications of STXM and NEXAFS spectroscopy. *Geochimica et Cosmochimica Acta* 71: 5439-5449.
- Yuan G., and Theng B.K.G. (2012) Clay-organic interactions in soil environments, in: P.M. Huang, Y. Li, M.E. Sumner (Eds.), *Handbook of Soil Sciences*. 2nd edition. Vol. 2: Resource Management and Environmental Impacts. CRC Press, Boca Raton. pp. 2.1-2.20.

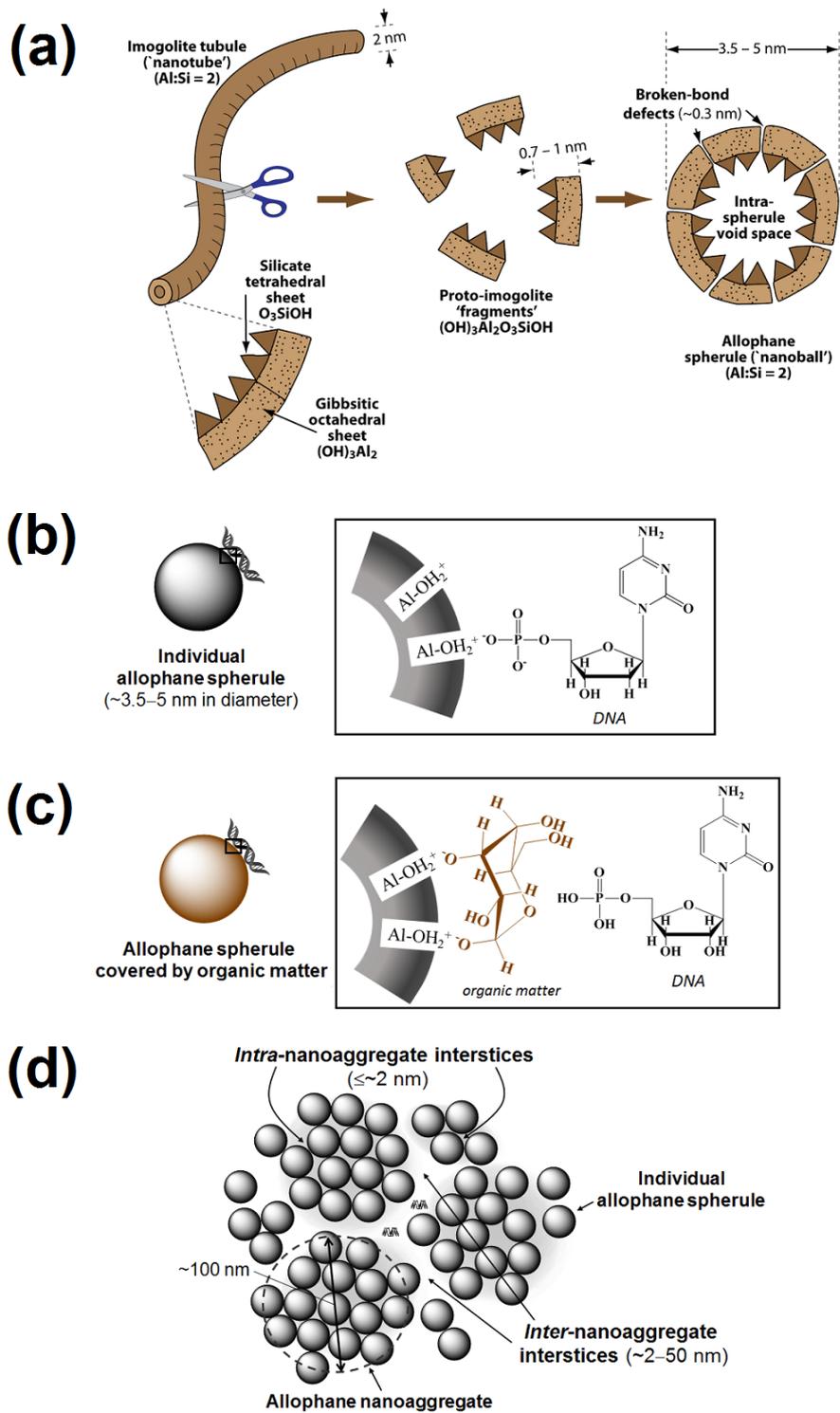


Fig. 17.1 Illustration of **(a)** nanoscale dimensions and composition of imogolite tubules and allophane spherules (from McDaniel et al., 2012), **(b)** direct chemical adsorption of DNA on allophane, **(c)** indirect chemical adsorption of DNA on organic matter-rich allophane, and **(d)** physical adsorption of DNA in the interstices of allophane nanoaggregates.

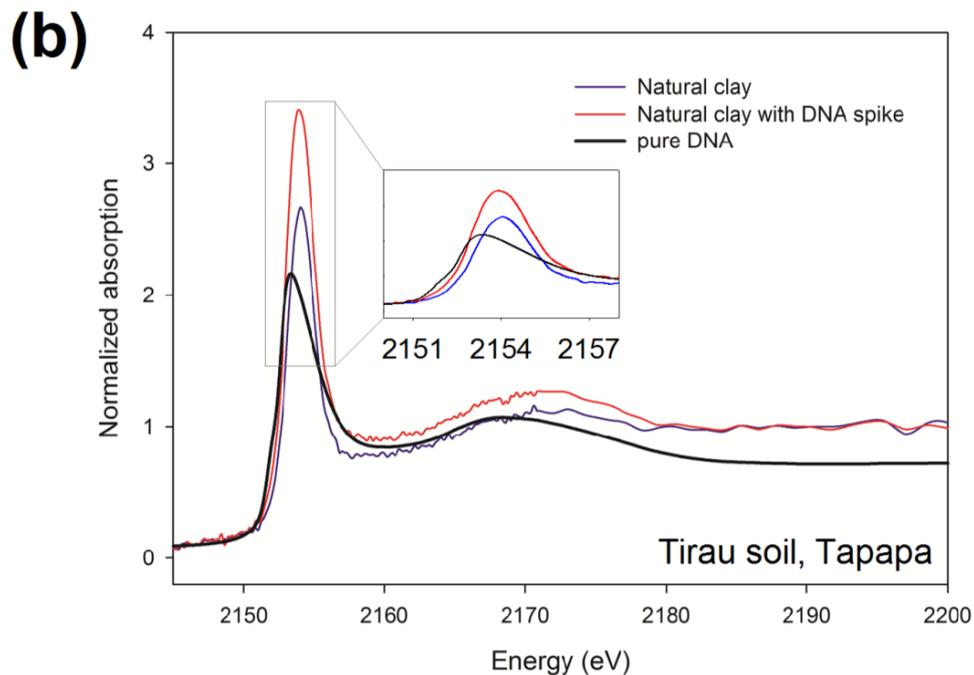
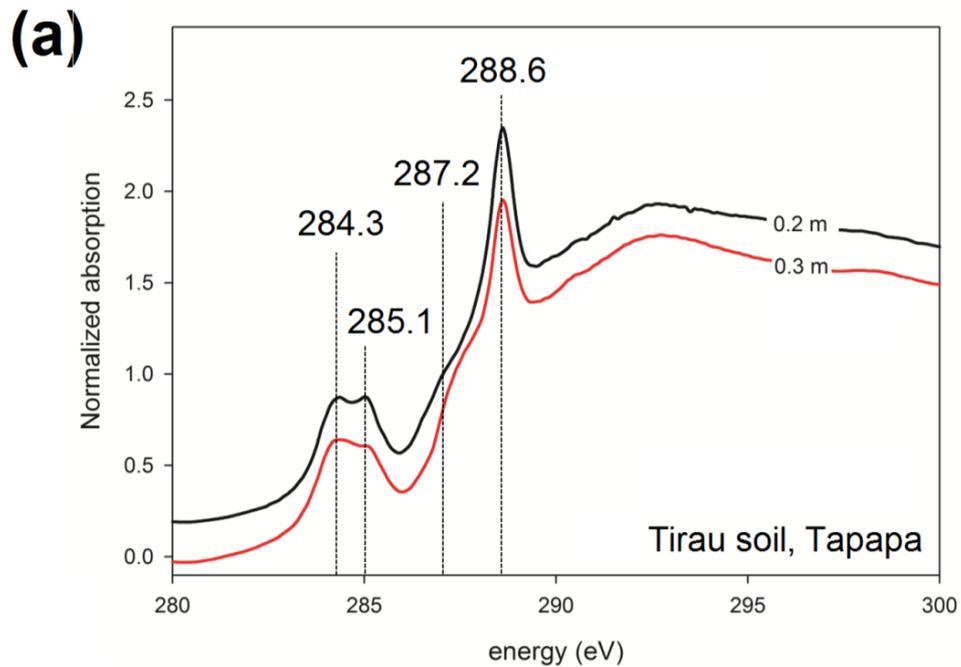


Fig. 17.2 Normalized **(a)** C NEXAFS spectra for organic matter in clays from the upper subsoil (~20–40 cm depth) of the Tirau soil, an Andisol at Tapapa in New Zealand (37° 59' 41" S, 175° 52' 59" E: Lowe et al., 2012), and **(b)** phosphorus XANES spectra for salmon-sperm DNA, for natural allophanic clay from the Tirau soil at Tapapa, and for allophanic clay with a salmon-sperm DNA spike. Peaks at 284.3, 285.1, 287.2, and 288.6 eV represent quinonic, aromatic, aliphatic, and carboxylic C, respectively.

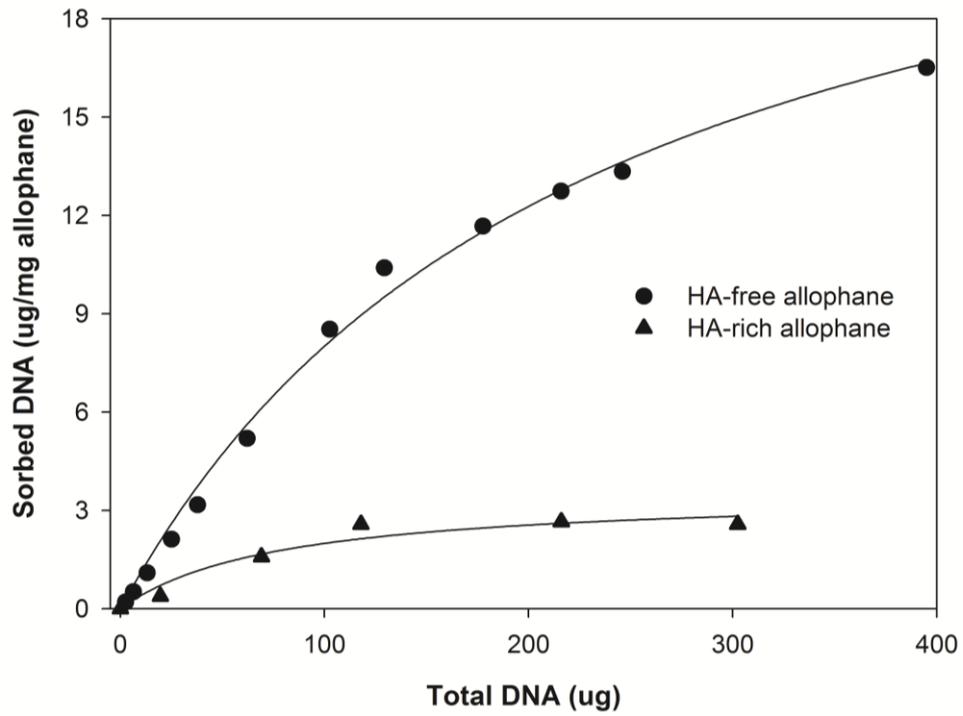


Fig. 17.3 Salmon-sperm DNA adsorption isotherm for humic-acid (HA)-free and for HA-rich synthetic allophane. This experiment was based on work by Saeki et al. (2010), and DNA in the supernatant was measured by UV spectrophotometry at 260 and 280 nm.