Coupling X-ray CT and diffraction measurements to monitor local mechanics associated with plant root soil bioturbation – a proof of concept using micro cone penetrometers

James Le Houx[1,2,*,†], Daniel McKay Fletcher[3,4,†], Alberto Leonardi[5,†], Katherine A. Williams[3,6,†], Nancy Walker[3], Fernando Albarez Borges[3], Ebrahim Afsar Dizaj[3], Madhu Murthy[3], Ronan Smith[3], Liam Perera[5], Navid Aslani[5], Andrew James[5], Sharif Ahmed[5], Tiina Roose[3], and Siul Ruiz[3,†]

[1]ISIS Neutron & Muon Source, Rutherford Appleton Laboratory, Didcot, OX11 0QX, United Kingdom
[2]The Faraday Institution, Harwell Science and Innovation Campus, Didcot, OX11 0RA, United Kingdom
[3]University of Southampton, Faculty of Engineering and Physical Sciences, University Road, Southampton, SO17 1BJ, United Kingdom
[4]Scottish Rural University College, Rural Economy, Environment and Society, West Mains Road, Edinburgh, EH9 3JG, United Kingdom
[5]Diamond Light Source, Rutherford Appleton Laboratory, Didcot, OX11 0QX, United Kingdom
[6]University of Portsmouth, Faculty of Science and Health, King Henry I Street, Portsmouth PO1 2DY, United Kingdom
[†]these authors contributed equally to this work

ABSTRACT

Soil compaction and increased global drought enhance soil mechanical strength and stiffness, which poses threat to soil health and ultimately adversely impacts society. Despite these restrictive conditions, some plant roots are able to grow under these harsh environments. This study was formed to develop a proof of concept measurement system that could be used to quaantify local plant biomechanical pressures during growth in in-situ assays. Using micro cone penetrometer root analogues, we tested the viability of combining X-ray computed tomography (XCT) with X-ray diffraction (XRD) measurements in order to understand whether we can characterize the mechanics during penetration associated with root growth. Cone penetrometers were used to penetrate into surrogate soil materials (e.g. gypsum), as these materials have porous and granular behaviour similar to natural soils but facilitate diffraction measurements due to their well defined atomic structures. Diffraction maps were generated across a sub region of the imaged space in order to quantify changes in the diffraction patterns. Diffraction maps are simplified representations by spectra peaks resulting from integrated Debeye-Scherrerr rings. Shifts in the spectra peaks are associated with deformations of the lattice crystals that comprise the surrogate soil material resulting from applied loads from the micro cone penetrometers. XCT images highlight compaction zones near the cone tip during penetration, with an increase in the compression zones radius as we push the cone deeper into the media. The XRD measurements peak shifts just outside of the local compacted zone of the cone. Qualitative finite element simulations illustrate that this is likely due to local inelastic deformations near the cone tip during penetration. However, the remote elastic deformations can be monitored as long as the inelastic zone remains local to the cone tip. While direct measurements near a root tip might be difficult due to the inelastic nature of the penetration process, the results show promise in terms of gaining insights into the locality of root growth biomechanical processes and the potential to elucidate root strategies under compacted soils in an in-situ test.

Keywords: X-ray Computed Tomography, Diffraction, Plant Roots, Root analogues, Soil bioturbation, Finite element modelling

Main

Rising frequency and severity of droughts threaten global food security and civil development in much of the world[1], and soil compaction resulting from intensified farming adversely impacts 25% of Europe's mechanized arable land area[2]. Drought and compaction both increase the stiffness of soils[3] creating environments that are challenging for plant roots and burrowing organisms to penetrate in order to forage for water and nutrients[4]. Plant roots, along with other tunnelling organisms, help sustain soil function through soil bioturbation[5]: forming biopore networks which enhance deep drainage of water that mitigate

flooding, enable diffusion of oxygen into deeper soil horizons, and prime soil microbial activity[5]. Together, these effects create ideal conditions for agricultural systems, and are being invoked to promote reduce tillage treatments in order to combat soil compaction.

Plant roots are able to grow through surprisingly rigid substrates, but this does have limits and the localised mechanisms enabling them to perform this feat are not well characterised. Hypothetically roots penetrate soil through a multi-scale processes where root turgor pressure allows the root to axially extend into the soil tip wise. Simultaneously, cells near the root tip multiply and, through an enzymatic process, reorient themselves acting to reduce local frictional effects and assemble past the root cap[6] (Figure 1 A). While together these processes could allow some roots to exert locally up to 1 MPa[7] of pressure, capable of fracturing solid chalk[8], there is no clear understanding as to how or where the loads are being applied. Root caps can swell under compacted soils[9], which may provide mechanical advantages for subsequent growth[10]. However, it is unclear whether this is loosening the forefront of soil or extending pre-existing faults in the media (e.g. cracks).

Recently, researchers have used X-ray computed tomography (XCT CT) to visualize soil displacements associated with growing plant roots on the order of 3-10 μ m resolution under loosely packed soil conditions[11]. However, soil deformation itself is more difficult to quantify under more compact soil conditions, which is expected to become more prominent in agricultural settings given land use intensification and increased machinery weight[12] . X-Ray diffraction (XRD) measurements have shown promise as a tool for inferring material's mechanical stresses state (i.e. internal forces acting within the material)[13]. This non-destructive technique infers residual stresses by using the materials crystallographic planes as a strain gauge. Under an applied load, changes in a materials lattice spacing can be determined based on Bragg's Law[14](Figure 1 C). Given a material with known mechanical properties, stresses can be inferred from local strains. The method has shown success in quantifying stress fields in granular media via individual particles under confined uniaxial loads[15][16], but has not to date been applied to understanding localised biological processes in soil.

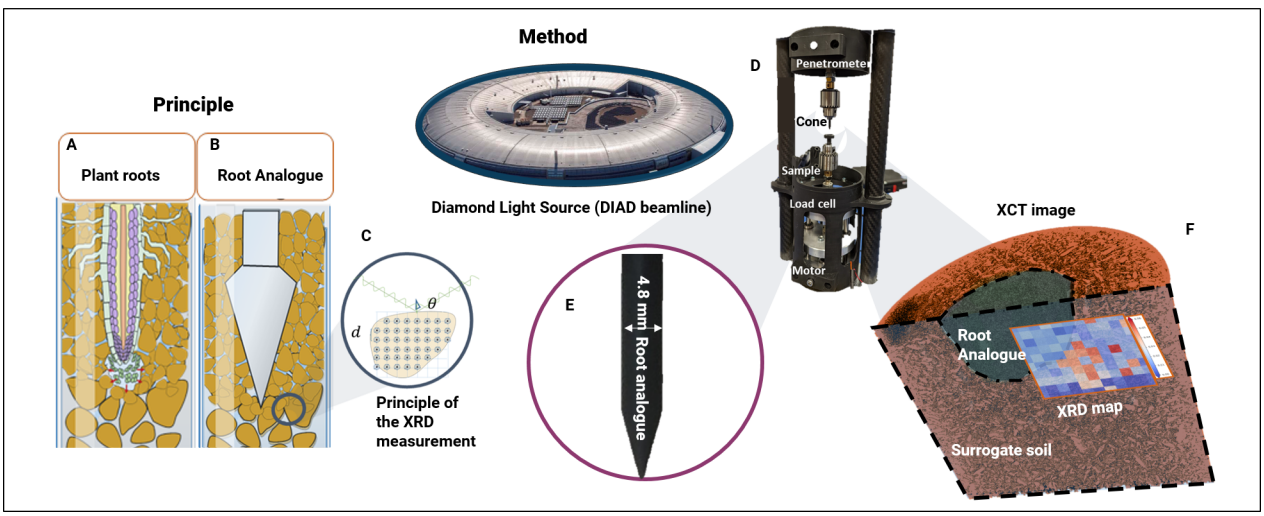

Figure 1. Methods and current experimental set up. The method is being developed for A – measuring biomechanics associated with root growth by using B – Cone penetrometer root analogues for C – XCT and XRD. D – developed in-situ rig with E - the cone penetrometer and sample near the detector using a root analogue cone. F - Overlaid XCT and XRD result from DIAD - XCT 3D image and 2D diffraction map representing the ensemble of stress values across the sample.

This work develops first steps for applying coupled XCT and XRD measurements for quantifying the mechanical stresses associated with root growth in soil. This benchmark used an artificial root (i.e. cone penetrometer Figure 1 B) moving into a surrogate soil material (Gypsum). All measurements were taken at the Diamond Light source at the K11 Dual Imaging and Diffraction (DIAD) beamline. At this beamline, a custom designed load frame was developed to emulate root growth (Figure 1 D) with a conical tip (Figure 1 E) loading tool to emulate the root growth into the soil sample. The load frame used a stepper motor to drive the sample upward into the conical tip, penetrated into the sample. A load cell measured the forces during penetration. Surrogate soil (Gypsum) was selected for its relatively soft crystalline structure and due to its use as an analogue for Martian soil[17]. We expected this would facilitate the diffraction measurements. We loaded the sample holders with wet paste of soil and let the samples dry to emulate strong capillary potentials in the domain. Once the sample was loaded, simultaneous XCT and XRD measurements were taken (Figure 1 F), where XRD provides a 2D map of the averaged diffraction peak shifts through the sample.

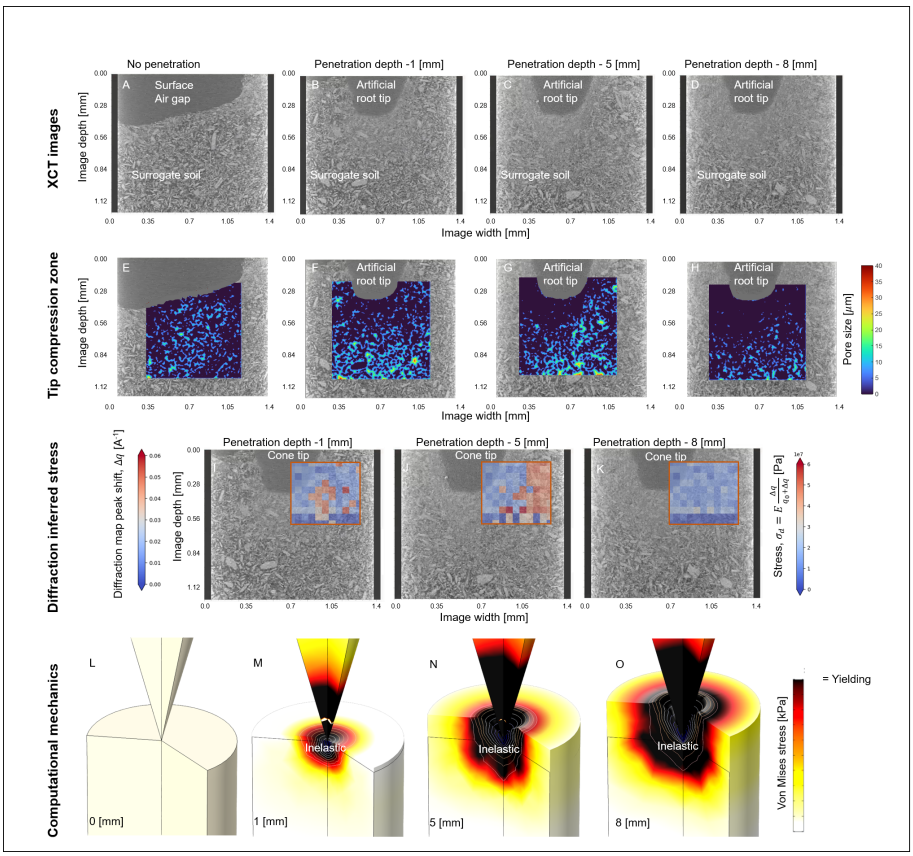

Figure 2. Coupled XCT imaging and XRD stress maps during root analogue penetration through surrogate soil (dried gypsum). A - the surface of the sample without the cone. B- The root analogue conical tip penetrating into the surface 1 mm. C - penetrating 5 mm depth. D - penetrating 8 mm deep. Compression zone can be quantified considering the changes in the pore sizes around the root tip for E - surface, F 1 mm penetration depth C - 5 mm penetration depth, and D - 8 mm penetration depth. XRD inferred stresses overlay the CT slices for I-1 mm, J- 5mm, and K - 8 mm depths, where the stresses ae inferred from the diffraction peak shifts and the elastic modulus of gypsum. Computational models are used to assess the yielding behavior of the system, where the artificial root penetrates into the surrogate soil continuum from the surface (L), 1 mm (M), 5 mm (N), and 8 mm depth (O). The model highlights the mechanical yield zone expanding, which creates a zone that cannot be readily measured via XRD.

The cone penetrated progressively into the sample to 4 different depths – 0mm, 1mm, 5mm, and 8mm. At each depth, XCT measurements were taken (Figure 2 A-D) to visualise the pore space changing with cone penetration (Figure 2 E-H). These images highlight the "compression zone", a region with fewer and smaller pores around the cone tip as it pushes into the soil.

While the XCT results were as expected, the overlaid diffraction maps for the associated regions were less intuitive. (Figure 2 I-K) plot the diffraction peak shifts associated with each loading step. For the 1mm load step (Figure 2 I), we monitor a small region just away from the cone tip where we see peak shifts associated with the loading. In Figure 2J, we can see that there is a region just adjacent to the cone tip where no clear peak shifts are visible. However, just outside of this region ($\tilde{2}00$ micron), we can see peak shifts acting as a boundary to this un shifted region. When we moved the artificial root 8 mm in (Figure 2 K), we no longer resolve peak shifts within our diffraction window.

In order to gain physical insights into the measurements a conceptual finite element mechanics simulation was carried out emulating the experiment (Figure 2 L-O). The simulation considered a cylinder of gypsum treating it as an elasto-perfectly plastic body using a Von Mises yield criteria[18]. The simulation results highlighted the development of the inelastic region around the penetrating cone tip, where in 1mm (Figure 2M) the inelastic zone is very local to the cone tip, and that progresses to reaching the outer radius of the cylinder at 8mm (Figure 2 O). This result gives an explanation as to why we see an un-shifted zone just around the cone tip in the diffraction maps (Figure 2 I-K). The macroscopic manifestation of inelasticity is caused by the ensemble of microscale material failure, flow, and reorientation[19]. The diffraction measurement provides information pertaining to elastic strains of the materials crystalline structure. Therefore, when those crystals dislocate, reorient, and relax in

their new configuration, they are no-longer loaded elastically. It should be noted that the loading carried out in this work is geometrically more sophisticated than the confined uniaxial tests previously demonstrated. The material was subject to shear stresses that would likely induce immediate inelastic deformations in the material. This novel method offers great potential for understanding fundamental biomechanics associated with root growth, while considering the limitations highlighted in this study with regards to direct local measurements. Roots tip wise growth is unlikely to drag surface material to lower depths, thus the diffraction maps are more likely to follow Figure 2I-J rather than Figure 2 K.

Controlled tests with a natural plant root can be used to generate inferences regarding location of root loads (i.e. radial or axial) or whether plant roots are exploiting imperfections in their growth medium (i.e. crack propagation). Imaging data similar to what was taken in Figure 2 A-H can be used to infer morphological features of plant roots that can be beneficial for penetrating through mechanically harsh soils, and diffraction maps (Figure 2 L-O) can demonstrate how those growth patterns can be mechanically advantageous. Traits that are more beneficial for growing through harsh soils can be considered for cover crop breading programs for regenerative agricultural treatments. This potential in-vivo measurement method has implications for understanding root growth and adaptation in challenging soil conditions, which could ultimately be used to inform rewilding strategies for desertified areas.

While direct measurements cannot be made directly adjacent to the root tip, coupling the measurement with mathematical modelling can provide insights as to what the applied stresses are. For example, use of an analytic cavity-expansion model can be used to estimate cavity surface pressures based on the cavity size and the inelastic zone. Future studies will also make use of CT images to generate image based modelling domains for more accurate characterization of the soil heterogeneities.

While this work had been developed in the context of sustainable agriculture and land management, the results present broader applicability in terms of material science and even geotechnical engineering. Characterization of plasticity is often difficult in geotechnical engineering. However, we have shown that the diffraction measurements readily provide this information. This could aid with precision design of earth works.

Data Records

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Competing interests

(mandatory statement)

The corresponding author is responsible for providing a competing interests statement on behalf of all authors of the paper. This statement must be included in the submitted article file.