A Simple Method to Measure Key Parameters of Soil-root Structure Using Medical X-ray Tomography Scanning Technique

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ROOTS have an extreme impact on soil properties and parameters are essential toward successful crop management. The study aims at modifying simple and quick method to identify key parameters of root structure by using medical X-ray CT (X-ray computed tomography) scanning technique and assesses its potentials.The results reveal that the modified simple procedures made it possible to isolate the root from the soil and partially overcome the problem of partial volume effect, and the main taproot was shown located at 0 to 30 mm depth. This procedure presented appropriate technique for segmenting large amounts of data. The modified method reduced the duration of the experiment and the risk of sample disturbance compared with the conventional methods to derive the architecture and the functional imaging. Results of plant growth parameters were closely related to the morphological parameters measured using the medical X-ray CT techniques.This approach enabledobservingroot growth manner and how individual roots overcome obstacles on their way. The technique modification shows great potential to provide new fundamental insights into soil-plant interactions and increase understanding of soil plant relationship.

Keywords: Medical X-ray computed tomography; Soil-plant interactions; Root morphology; Root architecture.

Soil-root interactions in relation to environmental effects and crop productivity are important in agricultural science (Anon, 2010). Roots embody nonvisible part of plant biology in which is involved (Waisel *et al*., 2002). They are represented in the activity of microorganisms and hence soil organic matter decomposition (Gregory, 2006). Several methodologies have been used to investigate root developments. Conventional techniques include semi-transparent nutrient agar growth media (Clark *et al*., 1999 and French *et al*., 2009) and/or gellan (Clark *et al*., 2011) as artificial growth media. Hence the root washing method is the most common technique to study plant root systems in soil (Smit *et al*., 2000 and Gregory, 2006). Spatial distribution of roots is lost using thismethod which involves breakage of fine roots. Rhizotron and minirhizotron techniques have extensively been used (Vamerali *et al*., 1999 and Johnson *et al*., 2001) in roots grown in soil, to identify the direction of root growth. However, observations indicate only small fractions of the entire root system, and are limited to the boundary surface. An alternative technique is X-ray computed tomography (CT), a non-destructivequantitative technique that can visualize and image opaque objects. It provides new parameter insights of soil-root-water interactions (Pierret *et al*., 2005 and Jahnke *et al*., 2009). This technique has been intensively used to show the soil morphology in three-dimensional set-up without soil destruction (Anderson *et al*., 1990 and Naveed *et al*., 2013). For identifying soil architecture and functions, Naveed *et al*. (2013) investigated linking X-ray CT measurements on soil pore parameters with the conventional soil physical measurements. Image analysis using X-ray CT scanners involve the use of medical CT from 500 mm to 200 nm along with nano-CT scanners to quantify three dimensional structures of soil pores and their connectivity. Cnudde *et al*. (2006) showed that images with medical CT facilitate scanning of larger (10–100 cm) samples, andits higher spatial resolution is limited to smaller (0.1–1 cm) samples.Sander *et al*. (2008) assessed paddy soil structure in soil columns

using medical CT with spatial resolution of 0.25 mm horizontally and 1 mm vertically. For the three dimensional visualization and quantification in soil science, Taina *et al*. (2008) consider the X-ray CTscanning technique is a unique tool, where its spatial resolution ranges from 10–500 µm depending on the scanner type and sample size.A CT scanner requires several projections for the different angles used for reconstructionof the CT data in three-dimensional systems. For visualization purposes, Mooney (2002) reported that the data are mapped to grayscale intensity values and are expressed in Hounsfield units. Gregory *et al*. (2006), Jenneson *et al*. (2003) and Tracy *et al*. (2010) showed that CT is an efficient tool to visualize plant root system. The overlap in x-ray attenuation values of organic matter and plant roots in soil was the limiting obstacle, where their variationsare caused by water stored in soil pores and/or retained in roots. Heeraman *et al*. (1997) reported that root extractions were difficult using current based image analysis thresholding approaches for quantification the root material using a per-voxel basis. These voxels were defined to different classes reflecting the scanned specimen components, and building a single model expected from materials of X-ray attenuation data. This approach was conscious to noise and required clear distinction between the grayscale values of the object and its background values (Pierret *et al*., 1999; Lontoc-Roy *et al*., 2006; Kaestner *et al*., 2006 and Perret *et al*., 2007). Although the thresholding techniques alone scarcely supplies sufficient accurate results, it is often combined with additional operations. Pierret *et al*. (1999) and Kaestner *et al*. (2006) preformed several morphological operations of post-processing for determining the remaining voxels that could belong to the final outcome or not. Lontoc-Roy *et al*. (2006) and Perret *et al*. (2007) provide an explicit connectivity check methods to coincide voxels in the threshold limits and not part of the roots. The objective of the present work was to investigate and modify an easy method to identify and describe the main root morphology growing using amedical CT scanning technique and assess the potentials and limitations of this technique.

Materials and Methods

Quantification of root architecture using medical CT

Imaging with a CT scanner is based on measurements of the different attenuation of X-rays passing through various kinds of matter of the sample. Image acquisition was done at El-Nour Radiology Centre (Egypt) with an X-ray CT of Siemens ECLOS (Medical Corporation). Soil pots with their plants were placed on the bench and scanned by the X-ray medical scanner (Fig. 1).
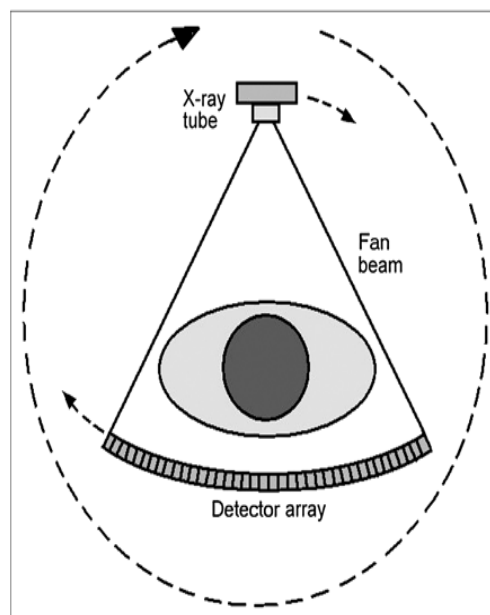

Fig.1. Detector+ X- ray source in rotation around the sample, it generates a3 D image from a series of 2Dimages taken around asingle axis of rotation as anon- destructive method

The scanner consisted of a gantry with a patient couch for positioning and advancing of the patient. The gantry opening and transaxial field of view (about 70 cm) allows for investigating large objects. The CT acquisition was performed as a helical scan using a voltage of 120 kV at 350mAs. The CT images were reconstructed on a 512 × 512 matrix using filtered back projection and an edge preserving filter with a convolution kernel (b30). The reconstruction kernelisis defined as the image processing filter applied to the raw data to yield a final scan image. The data were reconstructed on a three-dimensional matrix with the voxel values directly reflecting the linear attenuation coefficient in the field of view. The values are reported in Hounsfield units (HU) (Hounsfield, 1973), where attenuation of air and water are set to −1,000 and 0 respectively. After scanning and reconstruction, 256 slices were produced, in coronal view. The CT images (16 bit) had a voxel dimension of 0.39 × 0.39 × 0.6 mm for all the 96 image stacks (corresponding to the 32 samples scanned three times).

Image processing and analysis

Different steps were required for the root structure analysis. The analyses were made using the software ImageJ (Rasband, 2009). The images were selected and segmented using the automatic Otsu thresholding algorithm provided in the ImageJ software (version 1.45K) to isolate the root from the soil. This resulted in binary images from the original grey-level images. Subsequent, image analysis was carried out using the binary images.

The segmentation method used to isolate the root system was based on a thresholding method in similar to the method described by Tracy *et al*. (2010). Our method has been adapted as we were using ImageJ instead of VGS. The wand tool of ImageJ, permitted to select a certain grey-value range from a starting voxel, which is initially chosen on the probability of being most likely to be plant material. In this instance this can be done most easily by highlighting the plant stem material at the top of the column. Tolerance values were adjusted to ensure that only root material was included in the root region of interest (ROI) from the original seed points due to variations in grey-value resulting from slight variation in the density of the root material. As want tool works in 2D, this has been repeated over each slice. With the Particle analyzer plugin available in ImageJ,

only the connected voxels, which had the pixels in white, were selected. This procedure made it possible to isolate the root (main root) from the soil and partially overcome the problem of partial volume effect. We obtained binary images stack with the white corresponding to the main root.

Main root morphological parameters

Marching cubes algorithm, implemented in an ImageJ plugin, BoneJ, was used to render iso-surface (in the form of a triangular mesh) on 3D volumetric data to calculate surface area and volume of roots (Lorensen&Cline, 1987). An iso-surface represented the volume space, where each point of that space had the same constant value or iso-value. The iso-value was settled at 254. Voxels with values higher than 254 would considered inside the iso-surface. On binary images, only two values were represented: 0 (white) for roots and 255 (black) for the soil matrix, therefore voxels representing pore space would be outside the iso-surface. Surface area of the roots (AS) and its volume (AV) were calculated using this algorithm. With images of the binaries, the main root thickness could be defined as the diameter of the most fitting sphere with centre P (x, y, z) onto the root structure. The plug in local thickness (Dougherty & Kunzelmann, 2007) gave the mean thickness of the main root and the thickness map is displayed at each point P of the root.The 3D coordinates X, Y, and Z of the geometrical centre for each root were identified. A thinning algorithm was applied to reduce iteratively the diameter of root until only a skeleton remained and available as a plug-in, 3D skeletonize, in ImageJ. This process was performed symmetrically to keep the skeleton lines in a medial position and preserve the connectedness of the root volume. Besides, the ImageJ plugin was used and skeletonanalysis was done to analyse these networks. In this way, characterization of the largest shortest path for each network was attained. The largest shortest path is an estimate of the Euclidian distance obtained with the Floyd-Warshall Algorithm.

Plant material and growth conditions

A pot experiment was carried out to examine the effect of using compost and compost tea combined with chemical nitrogen fertilizers on growth of radish (*Raphanussativus*). Medical X-ray CT technique was used to study the interaction of soil with roots of the plant. Soil garbage compost "C" having organic carbon content of 30%, total nitrogen 1.0%, moisture 35%, and pH 7.5 was

used. Compost tea "CT" (water extract of compost) was prepared according to the method of Ingham (2005). Its properties were as follows: EC0.63 dSm^{-1},pH 7.2, total N, P, total K being 3.5, 23.9 and 450.0 mgL^{-1} respectively.The soil properties wereas follows: EC (past extract) 0.51 dSm^{-1}, pH 8.02, field capacity 9.14%, organic matter 4.8 gkg^{-1}, andCaCO$_3$5.0 gkg^{-1} (analysis according to methods cited by AOAC, 2002 and Baruah & Barthakur (1997).The soil was prepared by packing 6 kg pot^{-1} (Ø=20 cm, height=25 cm) and set in a vertical position. Plant seeds were sown on the 5th of November 2013. The experimental design was a randomized complete block with three replicates. Plants were thinned to one plant per pot after 15 days of sowing.There were ten treatments as follows: control, recommended dose of N–fertilizers "RDN" (240 kgNha^{-1}) as ammonium sulphate, recommended dose of compost "RDC" (24 ton. ha^{-1}), RDN + RDC, compost tea "CT" + RDC, CT "1:25"(*i.e.* diluted by water v/v) + RDN, CT "1:50" + RDN,CT "1:75" + RDN, CT "1:25" + 12/ RDN,CT"1:50" + 12/ RDN, and CT "1:75" + 12/ RDN. Micro-organism populations in compost and compost tea, (*i.e.* bacteria, aerobic N$_2$-fixing bacteria, actinomycetes and fungi) were determined using plate count and/or most probable number (MPN) technique (Table 1).

Results

Root morphological characteristics

Root morphological characteristics varied among treatments and have major effects on total root parameters and their 3-D architecture (Table 2). Main roots were traced and measured by following the shape of the root path, thereby providing accurate measurements of the root length as it extends down the soil pots (Table 2). Results of X-ray computed tomography reveal that soil amended with RDC+RDN gave the longest root depth of 149.3 mm (Table 2). The same treatment showed highest morphological root parameters of volume, surface, thickness, and tortuosity.

With the binary images, the main root thickness distribution versus the root depth expressedas the diameter of the best fitted sphere with centre P (x, y, z) onto the root structure is shown in Fig. 1. Using this processthe skeleton lines in a medial position was performed symmetrically and kept the connectedness of the root volume (Fig. 2). Results of Fig. 2 show that for all curves root thickness increase within the first mm of the root. It is followed then by decrease of thickness. RDC shows a maximum root thickness at 10 voxels depth (*i.e.* 3.9mm). In addition, using this technique, the root thickness was measured up to 250 voxels or 9.7cm for the RDC+ RDN treatment.

TABLE1 .Microbial populations in the compost and compost tea under investigation.

Treatment	Microbial populations			
	Bacteria	**Aerobic N$_2$fixing bacteria**	**Actinomycetes**	**Fungi**
Compost (log10 CFU/g)	8.32	1.23	6.54	4.41
Compost tea (log10 CFU/ml)	7.86	3.34	5.72	2.50

TABLE 2. Morphological parameters measured by medical X-ray CT of different treatments

Root morphometric parameters									
Sample ID	Volume (mm^3)	Mean thickness (mm)	Max thickness (mm)	Surface (mm^2)	Longest Shortest Path (mm)	tortuosity	Coordinate of centre of mass (mm)			Depth (mm)
							X	Y	Z	
C	1377.8	3.0	4.5	1919.4	71.3	0.8	14.3	30.1	6.4	56.9
RDN	12288.0	7.4	11.1	7646.4	169.6	0.8	16.6	52.1	10.6	139.7
RDC	7108.9	6.2	10.5	6432.2	169.8	0.5	30.0	25.6	20.5	93.2
RDC + RDN	13072.0	7.0	11.7	10155.1	188.2	0.8	22.6	82.6	9.4	149.3
CT2+RDN	5756.0	3.8	7.9	7352.1	190.6	0.6	23.8	34.3	31.3	117.1
CT3+RDN	4386.0	3.8	8.1	6190.9	214.0	0.6	22.3	39.1	25.8	121.2
CT1+1/2RDN	6958.0	5.8	9.1	5969.7	193.6	0.6	34.3	45.6	110.4	109.6

C : Control (without any additives); RDN: recommended dose of chemical N–fertilizers; RDC: recommended dose of compost; RDC +RDN; CT2 + RDN: Soil amended with CT (1:50 v/v water) + RDN; CT3 + RDN: Soil amended with CT (1:75 v/v water) + RDN; CT1 +1/2 RDN: Soil amended with CT (1:25 v/v water) + 1/2 RDN.
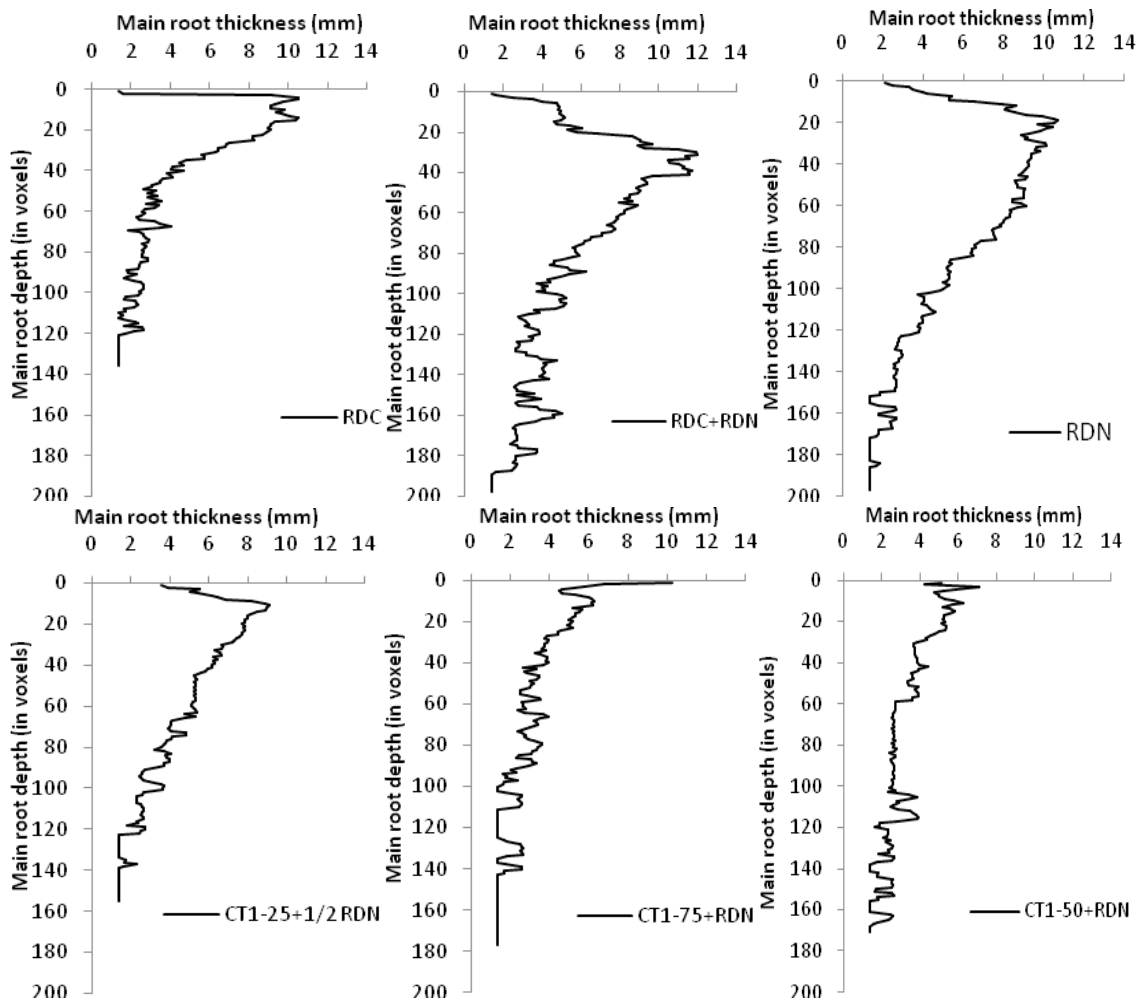

Fig.2. Characteristics main root thickness (mm) versus root depth (voxels) using X-ray CT data under different treatments; RDN: recommended dose of chemical N–fertilizers; RDC: recommended dose of compost; RDC +RDN; CT150- + RDN: Soil amended with CT (1:50 v/v water) + RDN; CT175- + RDN; Soil amended with CT (1:75 v/v water) + RDN; CT1 - 25 + 12/RDN: Soil amended with CT (1:25 v/v water) + 12/RDN

Root architecture using medical CT

Results of the isolated main root system are shown in Fig. 3 - 5. The obtained results were prepared whilst roots are still encased within the soil matrix. Figure 3 shows 3D visualization of the X-ray CT detected root of soil amended with RDN. The left image shows the pore thickness in 3D and 2D with the calibration thickness in mm. The right image is the main root network obtained after skeletonization. This level of details of growth medium structure could not be achieved using traditional methods for root studies, but using x-ray computed tomography enables following the root growth into the soil.

The centre of gravity for the plant root amended with CT175+-RDN was measured using X-ray CT binary images (Fig. 4A). Using this technique, the ability of the root to penetrate the soil matrix was established. Here the centre of gravity is located at 27mm from the soil surface. The images show the pore thickness in 3D with their respective calibration bar in mm for root of soil amended with CT175+-RDN (Fig. 4B), CT1-50+-RDN (Fig. 4C), and RDC+RDN (Fig. 4D).
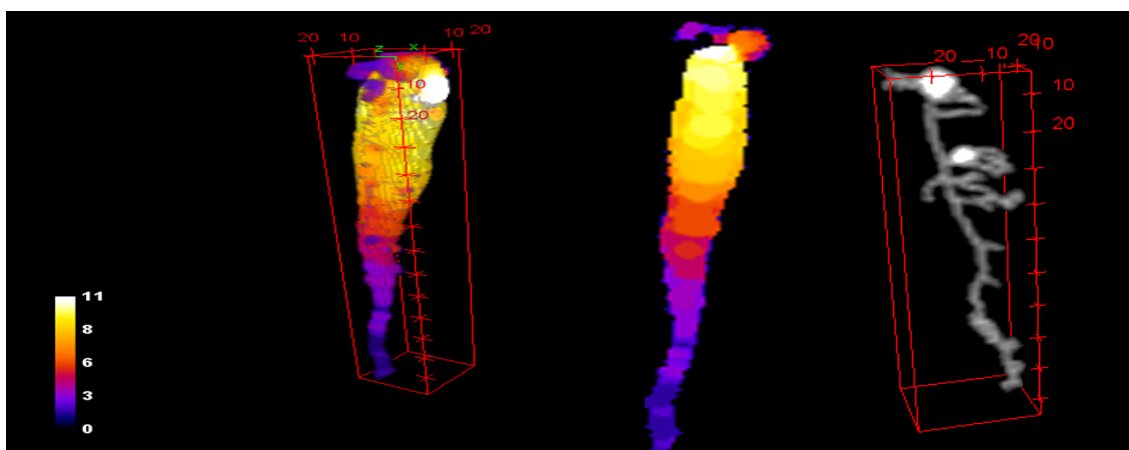

Fig. 3.3D visualization of the X-ray CT detected root of soil amended with RDN. The left image shows the pore thickness in 3D and 2D with the calibration thickness in mm. The right image is the main root network obtained after skeletonization
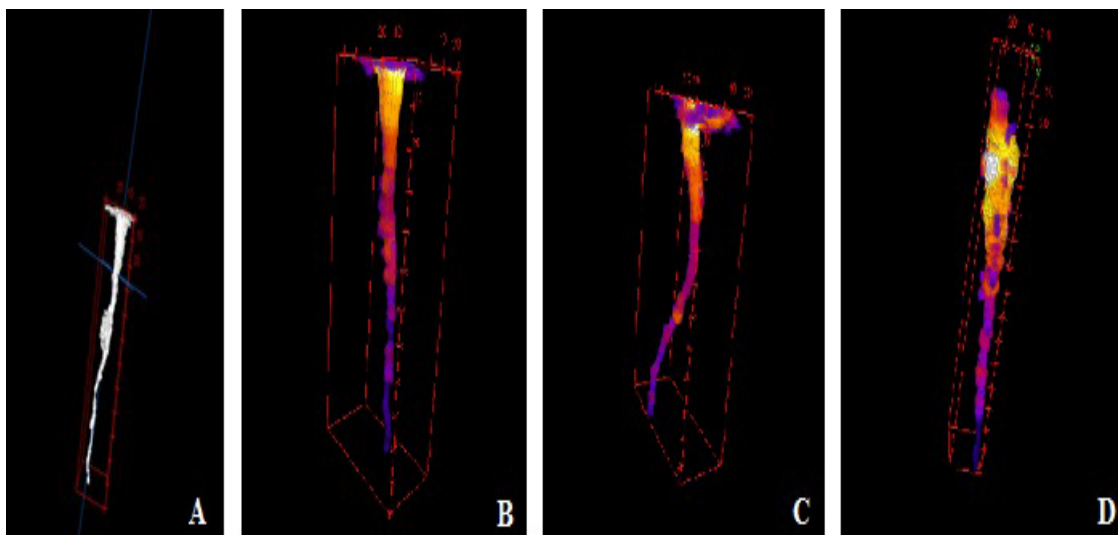

Fig. 4.3D view of the main root of soil amended with CT175+-RDN (A). In blue is indicated the axis of the moment of inertia. The centre of gravity is located at 27mm from the soil surface. The images show the pore thickness in 3D with their respective calibration bar in mm for root of soil amended with CT175+-RDN (B), soil amended with CT150+-RDN (C) and soil amended with RDC+RDN (D)

The treatment effect was distinct using X-ray CT binary images (Fig. 5). The images show the pore thickness in 3D with their respective calibration bar in mm for root of soil amended with RDC (Fig.5A), soil without any additives (Fig. 5B), and soil amended with CT12/1+25-RDN (Fig. 5C)

Plant parameters

Table 3 shows the growth parameters of number of leaves per plant, root fresh and dry weight, shoot fresh and dry weight. Soil amended with RDC + RDN gave the highest fresh and dry weight of roots (31.80 g plant^{-1} and 3.47 g plant^{-1}, respectively) as well as the highest fresh and dry weight of shoots (54.10 g plant^{-1} and 6.58 g plant^{-1}, respectively). Growth parameters were closely related to the morphological parameters measured by X-ray CT techniques (Table 2). That the treatment (RDC + RDN) gave the highest values of longest root depth, root volume, surface, thickness, and tortuosity.
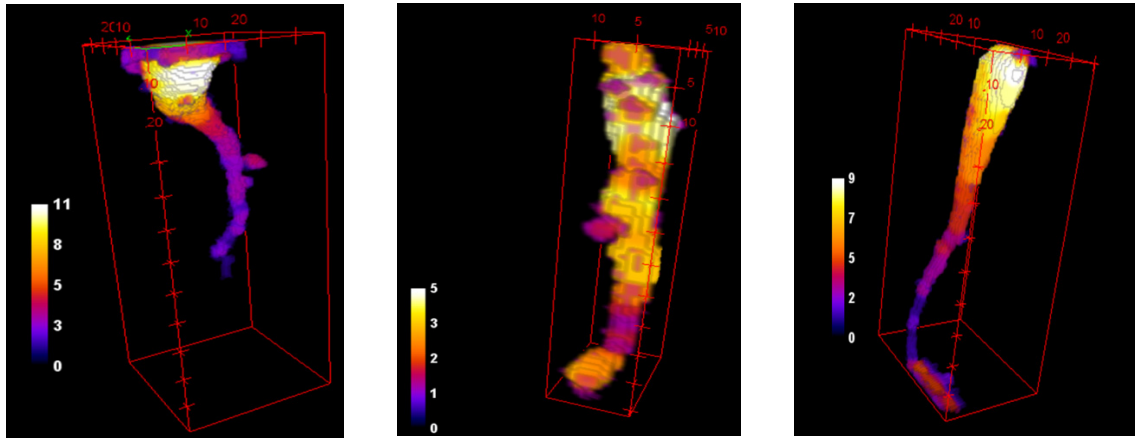

Fig. 5. Pore thickness in 3D with their respective calibration bar in mm for root of soil amended with RDC (A), soil without any additives (B) and soil amended with CT12/1+25- RDN (C)

TABLE 3. Effect of different treatments on root and shoot parameters of Radish plants

Treatment	leaves/ plant	Root F.W (g/ plant)	Root D.W (g/ plant)	Shoot F.W (g/ plant)	Shoot D.W (g/ plant)
Control	9.00	8.67 c	0.49 c	9.17 e	0.86 e
RDN	14.00	31.77 a	2.47 ab	46.17 a	3.57 b
Compost	11.33	24.67 ab	1.63 b	27.73 c	2.40 cd
RDC + RDN	12.67	31.80 a	3.47 a	54.10 a	6.58 a
CT (1:25) + RDN	10.67	17.77 bc	2.64 ab	32.60 bc	3.65 b
CT (1:50) + RDN	13.00	19.33 bc	2.39 ab	37.32 b	3.19 bc
CT (1:75) + RDN	11.33	18.03 bc	2.80 ab	29.35 bc	2.49 cd
CT (1:25) + 1/2 RDN	10.67	22.70 ab	1.99 b	17.27 d	1.96 d
CT (1:50) +1/2 RDN	13.00	26.47 ab	2.12 b	32.27 bc	1.72 d
CT (1:75) + 1/2 RDN	12.00	18.17 bc	2.07 b	30.03 bc	1.97 d

Numbers in the same column with different letters are significantly different ($P \leq 0.05$).
CT: Compost tea; RDN: Recommended dose of nitrogen; F.W.: Fresh weight; D.W.: Dry weight

Discussion

With the binary images, it is possible to define the main root thickness as the diameter of the best fitted sphere with centre P (x, y, z) onto the root structure. Even the plug in Local thickness (Dougherty & Kunzelmann, 2007) gave the mean thickness of the main root. The 3D coordinates X, Y, and Z of the geometrical centre for each root were identified. A thinning algorithm was applied to reduce iteratively the diameter of root until only a skeleton remained (Lee et al., 1994), and available as a plug-in3D skeletonize, in ImageJ. These processes were performed symmetrically to keep the skeleton lines in medial position and preserve the connectedness of the root volume. In the current study, medical CT scanning was used to provide structural and functional information about root and soil interaction. In fact, this technology gave visible images of the architecture and morphology of root in soil (Mairhofer et al. 2012). It was possible by this modification to reduce the duration of the experiment as well as the risk of sample disturbance compared with the conventional techniques (Jahnke et al., 2009; Dunbabin et al., 2013) to derive the architecture and functional imaging. Using medical CT scanner allowed placing the soil-plant system on the bench where it remained undisturbed during the whole experiment period (i.e. both initial labelling the subsequent scanning phase). The environment was also kept undisturbed during the experiment, with constant temperature and light intensity. The medical CT scanner enabled scanning relatively large samples, and keeping the experiment closer to the natural conditions. By instituting environmental control, it was possible to manipulate the environment and monitor the root-soil response. Witha resolution up to 0.6 mm, the CT images were rather similar to the resolution achieved by (Jahnke et al., 2009) using magnetic resonance imaging (MRI) (0.4 mm). A more effective resolution was achieved with the micro-CT scanner (90 μm). In this study, by using the medical CT scanner, it was possible to extract information on soil and root structure, that previous studies on the structure and functioning of roots were not able to integrate the component structure of the soil (Jahnke et al., 2009). The images of CT fluctuated with depth at higher values at the bottom of the VOI which could be attributed to the distribution of plant material and air surrounding the roots. This implies increased density with depth. The CT scanning technique also allows detailed visualization and quantification of structural properties in differently

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structured soils (Taina et al., 2008). In other studies, Amin et al. (1996) showed the positron emission tomography (PET) combined with MRI for structural and functional analysis of plant. The MRI is based on proton detection so that a sample contains a low amount of protons will not give a clear signal and the image resolution will be poor. With x-ray CT scan, it was not necessary to deal with such constraints (Garbout *et al.*, 2011). Using the medical CT scanner enables visualizing the main taproot and detecting it to 30 mm depth. Nevertheless, scanning with a medical CT scanner yielded no information on the fine roots and the main taproot was distinguished from fine or lateral roots. The micro-CT scanner showed the presence of fine roots down to 100 mm depth, and the PET showed signal until a depth of 80 mm (Garbout et al., 2011). The present study demonstrates the capability of medical CT scanning to supply datasets for non-destructive and non-invasive spatio-temporal studies on soil-plant interactions at high resolution. With this, it was possible to envisage a study taking place on plant growth and development subjected to stress, and treatment effects on plants with special focus on the root system. Further understanding and experiments on root growth, resource acquisition, and root-shoot communication under different effects of multiple and combined abiotic stresses (e.g. drought, waterlogging, salinity, nutrient status, soilphysical status) are required. These new technologies may help to elucidate how roots respond to below-ground abiotic stresses in terms of development, root-shoot communication and interactions with the biotic environment. Effects of above-ground stresses (e.g., light and temperature) can also be identified. Therefore hence the medical CT scanner can be used to provide information on root development. Rescanning with a micro-CT scanner may demand termination of plant growth, freezing and transport over long distances. In the present study, for simplicity, homogeneous sand was used as the test soil. This allowed a clear differentiation of roots from sand in CT scanning as emphasized by Lontoc-Roy et al. (2006). Natural soils, with a more complex structure, can be used in future studies using some modifications of the principles described in the current study and other previous studies (Taina et al., 2008; Choat et al., 2015).

Conclusion

X-ray CT is an innovative technique for studying root-soil interaction offering a major

advancement in our understanding the dynamic nature of the soil-plant relationship. The technology was modifiedin the current study and proved of considerable potential to show the immediate soil environment affects root architecture. With the binary images, defining the distribution of main root thickness versus root depth, the root tortuosity could be done. The 3D visualization of the X-ray CT detected the root whilst still encased within the soil matrix. At such level of details, the growth medium structure cannot be achieved using traditional methods for root studies.Using medical X-ray computed tomography enables following the root growth. By exploiting this technique, the below-ground impact of plant genotype on its phenotype regarding its roots and their soil environment can be visualised. This may help in identifying roles of novel genes in optimising the acquisition of water and nutrients from soil.

References

AOAC. (2002) *Official Methods of Analysis.* Association of Analytical Chemists (AOAC). International 17 th ed. Washington, D.C., USA.

Amin, M.H.G., Richards, K.S., Chorley, R.J., Gibbs, S.J., Carpenter, T.A. and Hall, L.D. (1996) Studies of soil-water transport by MRI. *Magn. Reson.Imag.* **14**, 879–882.

Anderson, S.H., Peyton, R.L. and Gantzer, C.J. (1990)Evaluation of constructed and natural soil macropores using X-ray computed tomography. *Geoderma* **46**, 13–29. Anon. 2010. How to feed a hungry world. *Nature* **466**, 531–532.

Baruah, T.C. and Barthakur, H.P. (1997) *Soil Analysis.* Vikas House. New Delhi.

Brady, N.C. and Weil, R.R. (1999) *The nature and properties of soils*.13th ed., Printice Hall, Upper Saddle River, New Jersey. USA.

Choat, B., Brodersen, C.R. and McElrone, A.J. (2015) SynchrotronX-ray micro tomography of xylemembolism in Sequoiasem pervirens saplings during cycles of drought and recovery. *New Phytol.* 205, 1095–1105.doi: 10.1111/nph.13110

Clark, L., Whalley, W., Leigh, R., Dexter, A. and Barraclough, P. (1999) Evaluation ofagar and agarose gels for studying mechanical impedance in rice roots.*Plant Soil,* **207**, 37–43.

Clark, R.T., MacCurdy, R.B., Jung, J.K., Shaff, J.E., McCouch, S.R., Aneshansley, D.J. and Kochian, L.V. (2011) Three-dimensional root phenotyping

with anovel imaging and software platform.*Plant Physiol.* **156**, 455–465.

Cnudde, V., Masschaele, B., Dierick, M., Vlassenbroeck, J., Hoorebeke, L.V. and Jacobs, P. (2006) Recent progress in X-ray CT as a geosciences tool. *Appl. Geochem.***21**, 826–832.

Dougherty, R. and Kunzelmann K., (2007) Computing local thickness of 3D structures with ImageJ. *MicroscMicroanal.***13**, 1678–1679.

Dunbabin, V M, Postma, J A, Schnepf, A, Pagès, L, Javaux, M and Wu, L. (2013) Modelling root–soil interactions using three–dimensional models of root growth, architecture and function. *Plant and soil,* 372 (1124–93 :(2-.

French, A., Ubeda-Toma´s, S., Holman, T.J., Bennett, M.J. and Pridmore, T. (2009) High-throughput quantification of root growth using a novel image analysis tool. *Plant Physiol.* **150**, 1784–1795.

Garbout, A., Munkholm, L..J., Hansen, S.B., Petersen, B.M., Munk, O.L. and Pajor, R. (2011) The use of PET/CTscanning technique for 3D visualizationand quantification of real-time soil/plant interactions. *J. Plant Soil,* **352**, 113127-.

Gregory P.J. (2006) Roots, rhizosphere and soil: the route to a betterunderstanding of soil science? *Eur. J. Soil Sci.* **57**, 2–12.

Heather, M.D., Alexandra, G.S. and Richard, P.D. (2006) Compost and manure mediated impacts on soil borne pathogens and soil quality. *Soil Sci. Soci. Am. J.* **70**, 347–358.

Heeraman, D., Hopmans, J. and Clausnitzer, V. (1997) Three dimensional imagingof plant roots in situ with x-ray computed tomography. *Plant Soil,* **189**, 167–179.

Hounsfield, G.N. (1973) Computerized transverse axialscanning (tomography). Part 1: Description of system. *British J. Radiology* **,46**, 101622-.

Ingham, E.R. (2005) *The compost tea brewing manual*. 5th ed. Soil Food web Inc., Corvllis, Oregon, USA

Jahnke, S., Menzel, M.I., van Dusschoten, D., Roeb, G.W., Bühler, J., Minwuyelet, S., Blümler, P., Temperton, V.M., Hombach, T., Streun, M., Beer, S., Khodaverdi, M., Ziemons, K., Coenen, H.H. and Schurr, U. (2009) Combined MRI-PET dissects dynamic changes in plant structures and functions. *Plant J.***59**, 634–644.

Jenneson, P.M., Gilboy, W.B., Morton, E.J. and Gregory, P.J. (2003) An x-ray micro-tomographysystem optimised for the low-dose study of living organisms. *Appl. Radiat. Isot.* **58**, 177–181.

Johnson, M.G., Tingey, D.T., Phillips, D.L. and Storm, M.J. (2001) Advancing fine root research with mini rhizotrons. *Environ Exp. Bot.* **45**, 263–289.

Kaestner, A., Schneebeli, M. and Graf, F. (2006) Visualizing three-dimensionalroot networks using computed tomography. *Geoderma,* **136**, 459–469.

Lontoc-Roy, M., Dutilleul, P., Prasher, S.O., Liwen, H., Brouillet, T. and Smith, D.L. (2006) Advances in the acquisition and analysis of CT scan data to isolatea crop root system from the soil medium and quantify root systemcomplexity in 3-d space. *Geoderma,* **137**, 231–241.

Lorensen, W. and Cline, H. (1987) Marching cubes: a high resolution 3d surface construction algorithm. *Computer Graph.* **21**, 163–169.

Mairhofer, S., Zappala, S., Tracy, S. R., Sturrock , C., Bennett, M. and Mooney, S.J. (2012) RooTrak: automated recovery of three-dimensional plant root architecture in soil from X-ray microcomputed tomography images using visual tracking. *Plant Physiol.* 158, 561–569.

Mooney, S.J. (2002) Three-dimensional visualization and quantification of soil macroporosity and water flow patterns using computed tomography. *Soil Use Management,* **18**, 142–151.

Naveed, M., Moldrup, P., Arthur, E., Wildenschild, D., Eden, M., Lamandé, M., Vogel, H.J. and de Jonge, L.W. (2013) Revealing soil structure and functional macroporosity along a clay gradient using X-ray computed tomography. *Soil Sci. Soc. Am. J.* **77**, 403–411.

Page, A.L., Miller, R.H. and Keeney, D.R. (1982) *Methods of Chemical Analysis.* Part 2: *Chemical and microbiological properties* (Second edition). American Society of Agronomy, Inc. and Sci. Soc. of America, Inc. Publishers, Madison, Wisconsin U.S.A.

Perret, J., Al-Belushi, M. and Deadman, M. (2007) Non-destructive visualizationand quantification of roots using computed tomography. *Soil Biol. Biochem.***39**, 391–399.

Pierret, A., Moran, C.J. and Doussan, V. (2005) Conventionaldetection methodology is limiting our ability tounderstand the roles and functions of fine roots. *NewPhytol.***166**, 967–980.

Rasband, W. (2005) ImageJ, 1.44b.United States National Institutes ofHealth.Available at: http://rsb.info.nih.gov/ij/.

Sander, T. and Gerke, H.H., (2007) Preferential flow patterns in paddy fields using a dye tracer. *Vadose Zone J.***6**, 105–115.

Sander, T., Gerke, H.H. and Rogasik, H. (2008) Assessment of Chinese paddy-soil structure using X-ray computed tomography. *Geoderma,* **145**, 303–314.

Smit, A.L., Bengough, A.G., Engels, C., van Noordwijk M., Pellerin, S. and van deGeijn, S.C. (2000) *Root Methods: A Handbook.* Springer-Verlag, Berlin, Germany.

Taina, I.A., Heck, R.J. and Elliot, T.R. (2008) Application of X-raycomputed tomography to soil science: a literature review. *Can. J. Soil Sci.* **88**, 1–20.

Tracy, S.R., Roberts, J.A., Black, C.R., McNeill, A., Davidson, R. and Mooney, S.J. (2010) The x-factor: visualizing undistorted root architecture in soilsusing X-ray computed tomography. *J. Exp. Bot.* **61**, 311–313.

Vamerali, T., Ganis, A., Bona, S.&Mosca, G. (1999) An approach to minirhizotronroot image analysis. *Plant Soil,* **217**, 183–193.

Waisel, Y., Eshel, A., Beekman, T. and Kafkafi,U. (2002) *Plant roots: The hiddenhalf.* CRC Press, NY, USA.

طريقة بسيطة لقياس الخواص الرئيسية للجذورفي التربة باستخدام تقنيةالمسح المقطعي بالأشعة السينية الطبية

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أقيمت تجربة أصص على تربة رملية ماخوذة من مزرعة التجارب بالخطارة – محافظة الشرقية – مصر، لدراسة تأثير أستخدام تقنية المسح المقطعي بالأشعة السينية الطبية (X-Ray Computed Tomography) بهدف أيجاد طريقة بسيطة وسريعة لتحديد المعايير والخواص الرئيسية للجذور داخل التربة وتقييم إمكانياتها. تم دراسة تأثير استخدام السماد العضوي ومستخلص السماد العضوي جنبا إلى جنب مع الأسمدة النيتروجينية الكيميائية على نمو الفجل(Raphanussativus) لاظهار تداخلات التربة مع جذور النباتات تحت المعاملات المختلفة. أظهرت النتائج المتحصل عليها تطوير طريقة عزل الجذور من التربة والتغلب على مشكلة تأثير الحجم الجزئي للتربة، كما أظهرت الخواص المورفولوجية للجذور داخل التربة. باستخدام هذه التقنية أمكن تجزئة كميات كبيرة من البيانات وتقليل التشوهات التى تحدث للصور الرقمية للجذور مقارنة مع الطرقا لتقليدية مع تقليل مدة التجربة على جهاز ال CT. باستخدام تقنية التصوير الرقمى تم تحديد ودراسة وتوزيع سمك الجذور الرئيسية على الاعماق المختلفة للتربة. وقد أظهرت النتائج المتحصل عليها أرتباط خواص نمو النبات المختلفة أرتباطا وثيقا بالخواص المورفولوجية للجذور التى تم قياسها باستخدام تقنيات الأشعة السينية الطبية CT-X Ray.

إستخدام هذه الطريقة المعدلة بالأشعة السينية CT ساعدت على كشف الجذر الرئيسى ومراقبة نمو الجذور داخل التربة تحت الظروف البيئية المختلفة داخل مصفوفة التربة. ويبين التعديل تقنية كبيرة لتقديم رؤى جديدة في تفاعلات التربة والنبات وزيادة فهم علاقة النبات بالتربة.