Proceedings of the 7th International Conference on Functional-Structural Plant Models, Saariselkä, Finland, 9 - 14 June 2013. Eds. Risto Sievänen, Eero Nikinmaa, Christophe Godin, Anna Lintunen & Pekka Nygren. http://www.metla.fi/fspm2013/proceedings. ISBN 978-951-651-408-9.

A combined method for quantifying 3D root architecture of field-grown maize

Jie Wu, Bo Yang and **Yan Guo***
Key Laboratory of Arable Land Conservation (North China), Ministry of Agriculture, College of Resources and Environment, China Agricultural University, Beijing 100193, China
**correspondence:* yan.guo@cau.edu.cn

Highlights: A new method for reconstructing 3D root architecture of field-grown maize was developed through integrating the spatial deployment of maize axile roots determined by digitizing *in situ* in the field with geometrical and topological information of scanned lateral roots. Spatial root length density and other root traits of maize can be estimated basing on this integrated method.

Keywords: root system architecture, topological structure, maize, root traits, model

INTRODUCTION

Root system architecture (RSA), refers to the topological arrangement of root segments and their geometrical characteristics (Fitter 1987; Danjon and Reubens 2008), has high capacity of plasticity responding to its rooting environment (Hodge 2004; Pagès et al. 2004). Camera and 3D laser scanner have been applied to non-invasively measure 3D dynamic RSA traits of young crop plants grown in transparent gel media (Fang et al. 2009; Clark et al. 2011). However, the root traits screens from non-soil media could be significantly different from or even reversed to those obtained from soil media (Gregory et al. 2009; Clark et al. 2011). To image and quantify 3D RSA in soil media, X-ray micro-computer tomography (CT) and neutron tomography have been utilized (Moradi et al. 2011; Mairhofer et al. 2012). However, high cost, restricted container size and limit of environmental factors restricts their applications in the field.

The objective of this study is to develop a combined method for quantifying 3D RSA of field-grown maize. We utilize digitizer to measure axile roots of individual plants *in situ* in the field, and develop a method to sample and collect topological and geometrical information of lateral roots using a software for root image analysis. We match the lateral root information with the axile roots to reconstruct and visualize the entire root system of individual plants. Different types of 2D and 3D root traits can be estimated based on the developed root model.

MATERIALS AND METHODS

Maize hybrids ZD958 (*Zea mays* L.) was seeded with row and plant spacing of 0.6 m and 0.3 m respectively on May 6, 2011 at the Shangzhuang experimental farm (40°08’ N, 116°10’ E) of the China Agricultural University.

For lateral roots measurement, three plants were selected at 75 days after sowing (10 days after silking). A self-made root auger (55 cm high, 50 cm diameter), composed by two half hollow cylinders, was put on soil surface with one sample plant at the center of the root auger. The root auger was hammered into the soil 50 cm in depth. A self-manufactured lift system was used to lift the root auger with sampled root system. Adjustable hydraulic nozzles were used to wash away soil with a disc kept at the base of the sampled root system to avoid root broken during washing. Then the root system was stored in a freezer with temperature kept 3 ℃. Two roots from the 4th to 8th nodal root whorls were selected for scanning. Branched zone of axile roots was cut into 5 cm segments from the base and first-order lateral roots along each axile root segment were cut. Axile root segment and its cut lateral roots were placed in a rectangular glass dish (25 cm × 20 cm) with water. Lateral roots were untangled carefully to minimize root overlap and then scanned using a scanner (ScanMaker i800 plus, Microtek, China). The eraser tool of WinRHIZO Pro 2009 software (Régent Instruments, Canada) was used to separate the overlapping of root segments in the images and remove noise. The developmental analysis function and Largard algorithm of the software were adopted to analyze the root developmental order, length and diameter of lateral roots while the former was also used to estimate the length and diameter of axile roots. Visual Basic Application embedded in Microsoft Excel 2007 was programmed to extract topological and geometrical information of lateral roots, length and diameter distribution of non-branched axile roots, and diameter of each 5 cm axile root segment.

Axile roots of two neighboring maize plants in a row were digitized *in situ* in the field using 3Space

Fastrak (Polhemus, USA) at 90 days after sowing (grain-filling stage). The axile roots were cut immediately after digitization and stored in a freezer for scanning to determine diameter and length of axile root segments.

3D RSA model of individual maize plant was built through matching measured lateral roots on the digitized axile roots using C++ language, boost library, SQLite and Visualization Toolkit. Model visualization was realized using ParaView software.

RESULTS AND DISCUSSION

The spatial deployment of axile roots of two neighboring maize plants at grain-filling stage was illustrated based on the dataset digitized *in situ* in the field (Fig.1A). Through matching measured lateral roots (Fig.1B) with digitized axile roots, the complete 3D RSA of an individual maize plant was reconstructed (Fig. 1C).

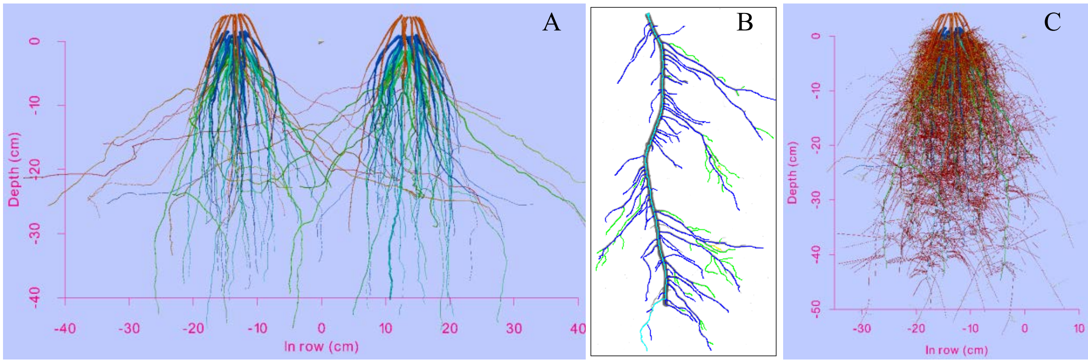

Fig. 1. A. 3D visualization of axile roots of two neighboring maize plants in a row, different colors indicate distinct root types. B. A sample of lateral root system output from WinRHIZO software, light blue, dark blue and green indicate first, secondary and tertiary order lateral roots. C. Visualization of the complete root architecture of an individual maize plant, red, yellow and green segments were first, secondary and tertiary order lateral roots, respectively.

Different root traits can be extracted based on the constructed RSA model. 2D root traits include the branching number and length density, the highest order of lateral roots, and the total length of each order lateral root on each 5 cm axile root segment of different root types. 3D RSA traits include angle and horizontal trajectory of different types of axile roots, and root length distribution of different orders of lateral roots and types of axile roots in each soil cell.

The distribution of lateral root length density of two neighboring maize plants at different soil depth was presented (Fig. 2). More than 70% of the lateral root length of the 0-40 cm soil profile was distributed in the 0-20 cm soil horizon. Significant variation of lateral root length density occurred in distinct soil horizons. For the top 10 cm soil horizon, most of root length concentrated around the plant base. In contrast, the distribution of root length density is much lower and more homogeneous at 20-40 cm soil horizon.

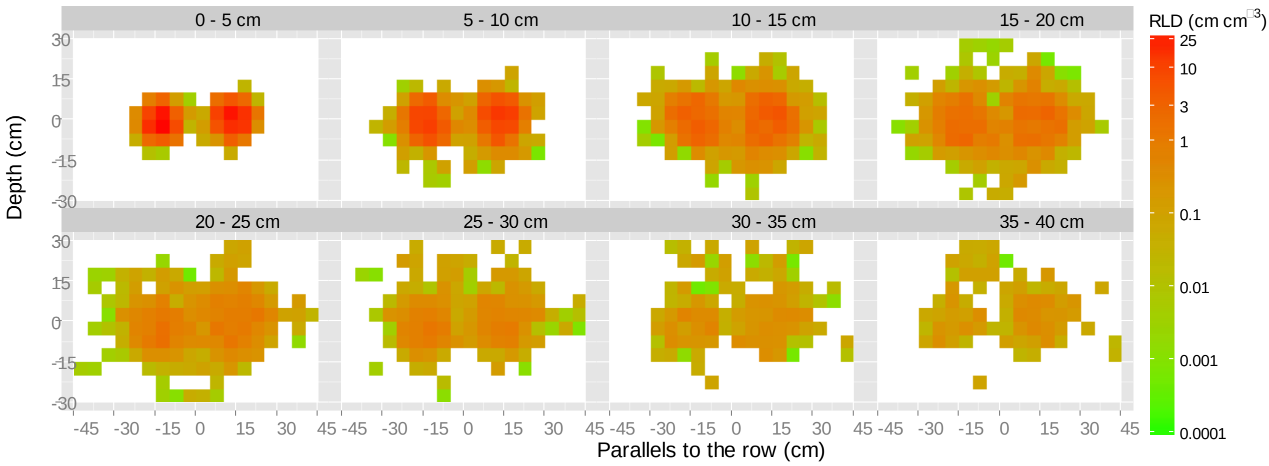

Fig. 2. The distribution of lateral root length density of maize at different soil horizons. The origin of the horizontal and vertical axis indicates the centre between two neighboring plants in a row and the vertical axis is perpendicular to

the row direction. The color mapping the values of root length density (RLD) in the legend is under logarithmic transformation.

ACKNOWLEDGEMENTS

This work was supported by the National Natural Science Foundation of China (41071205) and National Scientific and Technological Support Plan (2012BAD35B02).

LITERATURE CITED

Clark RT, MacCurdy RB, Jung JK, Shaff JE, McCouch SR, Aneshansley DJ, Kochian LV. **2011**. Three-dimensional root phenotyping with a novel Imaging and software platform. *Plant Physiology* **156**: 455 –465.

Danjon F, Reubens B. **2008**. Assessing and analyzing 3D architecture of woody root systems, a review of methods and applications in tree and soil stability, resource acquisition and allocation. *Plant and Soil* **303**: 1–34.

Fang S, Yan X, Liao H. **2009**. 3D reconstruction and dynamic modeling of root architecture in situ and its application to crop phosphorus research. *The Plant Journal* **60**: 1096–1108.

Fitter AH. **1987**. An architectural approach to the comparative ecology of plant root systems. *New Phytologist* **106**: 61–77.

Gregory PJ, Bengough AG, Grinev D, Schmidt S, Thomas WTB, Wojciechowski T, Young IM. **2009**. Root phenomics of crops: opportunities and challenges. *Functional Plant Biology* **36**: 922–929.

Hodge A. **2004**. The plastic plant: root responses to heterogeneous supplies of nutrients. *New Phytologist* **162**: 9–24.

Iyer-Pascuzzi AS, Symonova O, Mileyko Y, Hao Y, Belcher H, Harer J, Weitz JS, Benfey PN. **2010**. Imaging and Analysis Platform for Automatic Phenotyping and Trait Ranking of Plant Root Systems. *Plant Physiology* **152**: 1148 –1157.

Mairhofer S, Zappala S, Tracy SR, Sturrock C, Bennett M, Mooney SJ, Pridmore T. **2012**. RooTrak: Automated Recovery of Three-Dimensional Plant Root Architecture in Soil from X-Ray Microcomputed Tomography Images Using Visual Tracking. *Plant Physiology* **158**: 561 –569.

Moradi AB, Carminati A, Vetterlein D, Vontobel P, Lehmann E, Weller U, Hopmans JW, Vogel H-J, Oswald SE. **2011**. Three-dimensional visualization and quantification of water content in the rhizosphere. *New Phytologist* **192**: 653–663.

Pagès L, Vercambre G, Drouet J-L, Lecompte F, Collet C, Le Bot J. **2004**. Root Typ: a generic model to depict and analyse the root system architecture. *Plant and Soil* **258**: 103–119.