

Automated Characterization of the Mature Root System Form by a Double-Quadrangle-Shaped Polygon

Philippe Borianne

Amap - CIRAD
F-34398 Montpellier, France
philippe.borianne@cirad.fr

G rard Subsol

Research-Team ICAR, LIRMM
CNRS / University of Montpellier
France

Alain Audebert

Agap - CIRAD
F-34398 Montpellier, France

Abstract— We describe in this paper a new global approach to characterize the geometry of the root system area from black-and-white silhouettes. We introduce a covering polygon composed of two superimposed quadrangles to evaluate geometry features or to classify the growth strategy of root systems: the convex hull of the root system is progressively reduced to a double-quadrangle-shaped polygon by iterative suppression of least significant vertices to only keep the major geometry features. The root system density is addressed by the analysis of the hole distribution. The root system porosity is estimated from the decomposition of background regions in circular elements. An automated repartition of circular elements by 2-means clustering is introduced for a best appreciation of macro- and micro-holes, respectively associated to the anchorage and nutrient recovery potentials. The global parameters of the root system are evaluated from the geometry and density properties of the double-quadrangle-shaped polygon.

The experimental studies address the sensitivity of the double-quadrangle-shaped polygon, especially the evaluation of the geometrical stability of the polygon according to slight variations of the root system segmentation. Several experiments, performed using a representative set composed of 100 root system silhouettes of several species – *arabidopsis*, *chick pea*, *palm tree*, *rice*, *sorghum*, and *wheat* – at different stages of growth, underline the robustness of the shape parameters, especially the upper and lower penetration angles.

Keywords — *root system, root phenotyping, image processing, geometry and density characterization*

I. INTRODUCTION

Plant root system emphasizes several essential functions for whole plant development and growth, notably water and nutrient uptake and anchoring. Size, shape and spatial organization of the root system determine the explored soil volume accessible by the plant and *de facto* the access to water and nutrient resources which are major factors defining the adaptability of the plant, especially under stress conditions. Deciphering the genetic and molecular mechanisms which control the development of the root system and its adaptive plasticity to the availability of water and nutrient is of primary

importance for the sustainable establishment of a crop under adverse environments.

Understanding and measuring adaptation of cereal plants to their environment are now a key issue especially in the context of climate change. Cereal crop production is likely to decline with global warming [1][2]. This is especially true for cereals with shallow rooting. Roots allow nutrient and water uptake and are thus critical components of the overall plant productivity [3][4].

These challenges are based on high-throughput plant phenotyping approaches [5][6] developed for studying the relationships between plant traits and the genome. These approaches lie behind the development of many phenotyping platforms which more often use various non-invasive sensors [7][8] such as visible red-green-blue (RGB) imaging, chlorophyll fluorescence imaging (CFIM), thermo-imaging or hyper-spectral imaging. Moreover, it becomes now to perform “in vivo” high throughput root phenotyping (i.e. without extracting the root) by using transparent container connected with an imaging system. These phenotyping platforms are usually focused on shoot growth [9][10][11] and/or root growth [12][13][14].

A great diversity of overall architecture among root systems is fashioned as much by soil conditions as by genotype. Root System Architecture (RSA) is defined as a collection of root traits dedicated to the significant morphological features of the axes of the root system [15][16] which is largely used for characterizing the spatial configuration of young root systems. Its importance in plant productivity lies in the fact that major soil resources are heterogeneously distributed in the soil, so that the spatial deployment of roots will determine the ability of a plant to survive and growth [17]. This capability of plants is most often based on measuring the significant morphological features of the root system (and of its components). Beyond these measures, a classification scheme is essential to compare the root system behaviour. Different methods have been proposed based on phylogeny, morphology, topology or statistical traits [18][19][20][21].

This work is supported by Agropolis Fondation under the reference ID 1202-073 through the “Investissements d’avenir” program (Labex Agro: ANR-10-LABX-001-01).

For example, [19] defines the root system architectural taxonomy framework from four classes of root: the tap root which emerges from the seed, the lateral roots which are branches of other roots, the shoot-borne roots which arise from shoot tissues and the basal roots which develop from the stem basis. By contrast, the root system architectural topology framework identifies the morphological types of roots by their development pattern, their state of differentiation and their relationships [20]. A classification of root systems via their geometrical shape was introduced [22] and is boosted today by the increasing use of numerical images.

The geometry of the root system is more and more addressed through the measurement of several parameters from images acquired in phenotyping platforms or through observation glasses of pits dug in the soil.

In RSA approaches, the measures concern often the distribution of the root length and width, the cumulated lengths by diameter, the number of tips, curvature, etc. as done in RootTrace [23], EZ-Rhizo [24], Gia-Roots [25] or RootReader2D [13]. But these parameters are insufficient to globally characterize the form of the root system. Despite being a major indicator of the root growing strategy [28], the root cone angle remains difficult to assess: recent tool as DIRT [27] proposes to estimate it with varying success from RANSAC - *random sample consensus* - approach, an iterative method which identifies and excludes the outliers of the root system points.

An idea is to use a binary image where the root system was beforehand segmented (i.e. its contours were delineated either by an automatic image processing method or manually by an expert). This silhouette can lead to a global overview of the root system whose the spatial occupation can be evaluated with a covering form. The most commonly adopted approach, introduced in RootNav [28] or ARIA [29], is based on the convex hull, i.e. the smallest convex polygon including all the roots of the system. This polygon allows estimating the covering surface roots and some form factors as circularity, compactness or axial symmetry. One major problem is the sensitivity of the convex hull to small variations: a slightly longer root can modify considerably the hull. An alternative is proposed in GLO-Roots [30]: Elliptic Fourier Descriptors are used for providing a more precise boundary of the root system with parameters which are invariant to rotation, but this method is difficult to use. The landmark-based allometric method was introduced in RootScape [31] to overcome this difficulty: it is based on the measure of individual root traits for quantifying the variations in root architecture. For example, OpenAlea [32], Root System Analyser [33] or RootGraph [34] propose approaches for the detailed characterization of root traits based on a graph optimization process: primary roots are distinguished from lateral roots and a broad spectrum of root traits for each identified root axes is quantified. But these methods are rather adapted for studying sparse systems where the root axes cannot be individualized. In [35], a field-imaging protocol and algorithmic approach are combined to analyze mature root systems grown in the field: this work shows how image analysis can be utilized to estimate localized root traits that reliably capture heritable architectural diversity as well as

environmentally induced architectural variation of both monocot and dicot plants.

In order to classify root growing templates, a house-shaped polygon definition was proposed in GT-RootS [36] to estimate the general shape and spatial density distribution of mature rice root systems, which are too dense to allow the axis individualization. It is a polygon which is composed of two parts -a trapezoid for covering the upper part of the root system and a rectangle for the lower part - defined from the positions of the four most-external points and some significant vertical weighted-density profiles of the root system. It has been shown that such a polygon facilitates the evaluation of the root cone angle or the classification of the global root system shape.

But this house-shaped polygon appears particularly limited in its capability to precisely describe the global geometry of the deep roots. In this paper, we extend the house-shaped polygon concept to best characterize the shape and the density of the different root system levels. We propose an innovative method to define a covering polygon from an iterative simplification of the convex hull based on a local density descriptor. We propose also an alternative to the RSA density based on the identification of each root axis by introducing the notion of root hole granularity in order to refine precisely the estimation of root system density distribution.

II. METHOD

The method presented in this paper aims to obtain a global characterization of a mature root system silhouette. We consider that the root system is vertically presented in the binary image – *where the black points are roots and the white ones represent the background* – and oriented from top to bottom: the root neck is in the upper part of the image, the root meristems in the lower part.

A. The double-quadrangle-shaped polygon

The major challenge is to define a simple form for evaluating the penetration angle and estimating the soil occupation of the root system.

We propose to use a “house-shaped” polygon composed of two superposed quadrangles (cf. fig. 2). The upper quadrangle will allow the evaluation of the root cone angulation, the bottom one will give indications about the deep penetration angle. The geometrical relations, in particular the relative sizes, between the upper and the bottom quadrangles could lead to the classification of the root system shape. Contrary to some previous work based on the vertical profiles of root densities [36], the definition of the root system form is here only defined by its geometry. The idea is to progressively reduce the convex hull to a double-quadrangle-shaped polygon by suppressing the vertices which correspond to the less local dense parts of the root system. The resulting form is a closed polygon which best covers (but not necessarily includes completely) the root system. The double-quadrangle-shaped polygon is the best compromise between the root recovery rate and the surface loss rate. The root recovery rate is the percentage of roots included into the polygon; the surface loss rate is the percentage of the polygon reduction.

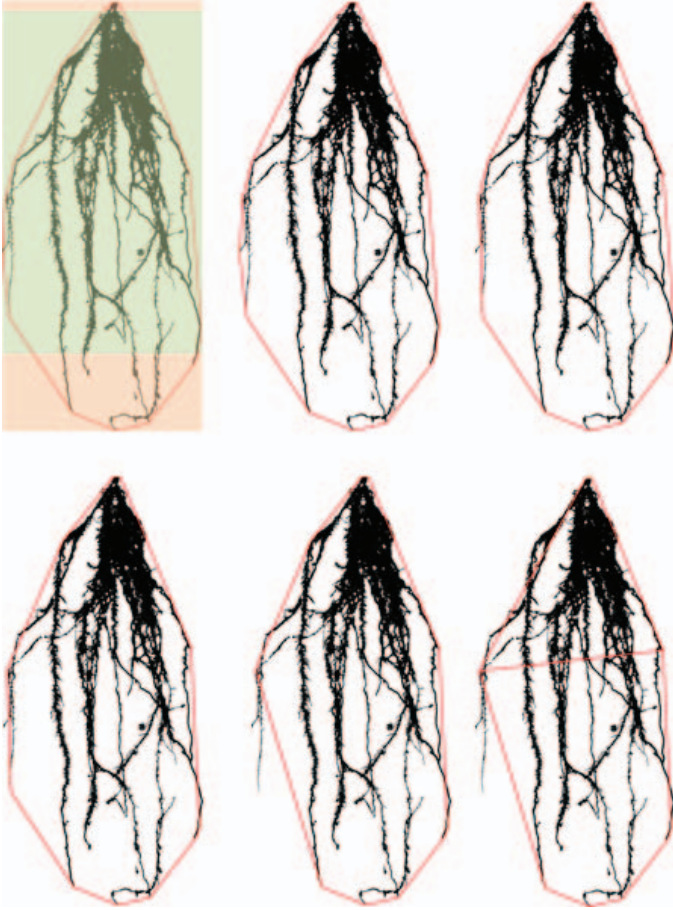


Fig. 1. The double-quadrangle-shaped polygon and its masks. Different steps of the convex hull regression are presented on a mature and dense rice root system (30 days). From left to right, and top to bottom: the convex hull and the 9th, 15th, 16th, 17th, 18th step with respectively 28, 19, 13, 12, 11 and 9 vertices. The covering border is represented in red. The 1%-upper and 20%-lower masks (in orange) cover the zones where the vertices of the convex hull could not be removed, and the complementary zone – in green – where the convex hull reduction will be applied.

The reduction of the convex hull is achieved by the following algorithm (cf. fig. 1):

1. Compute the convex hull of the binary image. The result is a polygon composed of n vertices v_i .
2. Evaluate the local density at each vertex of the polygon.

We call T_i the local neighbourhood at v_i defined by the triangle $[v_{i-1}, v_i, v_{i+1}]$. We define the local density d_i at v_i by the harmonic mean between two parameters R_i and S_i :

$$d_i = \frac{2 \times R_i \times S_i}{R_i + S_i} \quad \Leftrightarrow \quad \frac{1}{d_i} = \frac{1}{2} \times \left(\frac{1}{R_i} + \frac{1}{S_i} \right) \quad (1)$$

where

- R_i is the ratio between the number of black image points – also called *pixels* – which are included into T_i and the total number of black points (present in the binary image),
- S_i is the area neighbourhood contribution at v_i , i.e. the ratio between the area of T_i and the total area of the current polygon.

3. Remove the « less dense » vertex.
4. Go to step 2 until the stopping condition is verified

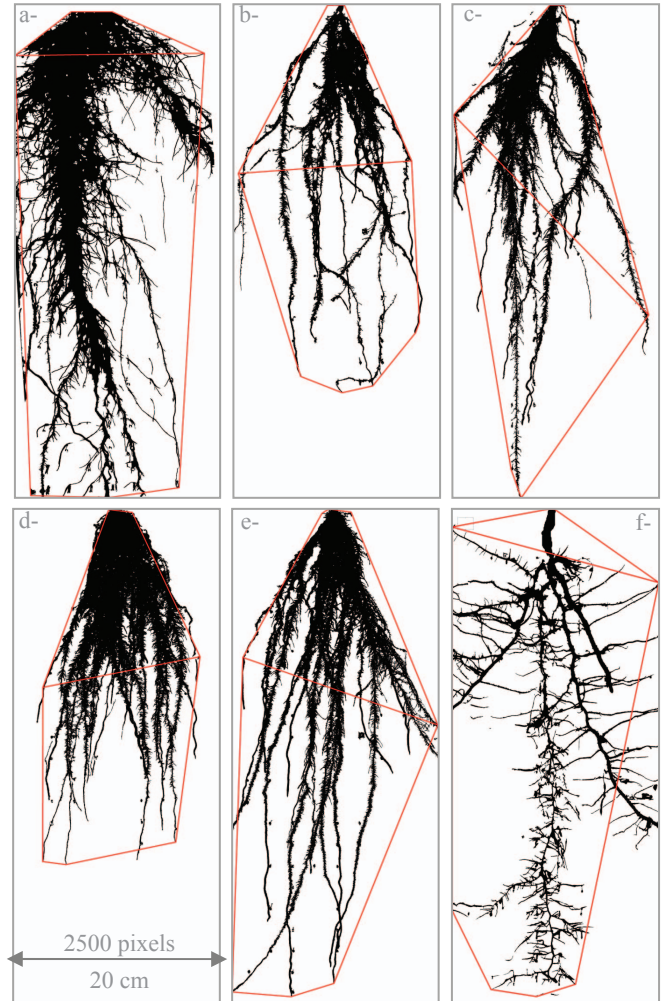


Fig. 2. The double-quadrangle-shaped polygon. Mature root systems in black and their covering polygon composed of two superposed quadrangles in red. a- 3-week-old sorghum, b- 4-week old wheat, c- 3-week-old Japonica rice (*Azucena*), c- 4-week old Indian rice, d- 4-week old wheat, e- week-old Japonica rice (*Cicli Beton*), f- 6-week-old palm tree.

In order to provide more significant form, it is important to conserve the main geometrical landmarks of the root system, especially around the principal root neck and meristems. The vertices of the convex hull corresponding to these landmarks must not be deleted. These specific vertices are located in the upper and lower parts of the convex hull as the root system was

oriented in the binary image. Two horizontal bands of variable thickness - respectively called the upper and the lower exclusion masks - are introduced to preserve the characteristic landmarks during the convex hull reduction step. Only the vertices not belonging to these bands can be suppressed: they are called the *removable vertices*. The thickness of each band is a percentage of the convex hull height assimilated to the root system depth (cf. fig. 1).

The reduction of the convex hull is stopped when the number of removable vertices is inferior or equal to 2 (cf. fig. 2).

B. The granularity of holes

In order to study more deep fully the spatial distribution of the root densities and classify the different strategies of root growth [36], we propose to evaluate the spatial repartition of background regions with respect to their size and shape. For this purpose, we decompose the complex shaped background regions into a collection of simple structuring elements. We choose to use circles which correspond to the intuitive definition of holes. Circles ease the size appreciation and the structuration of background (cf. fig. 2).

The decomposition $\bar{D}(X)$ of a region X in circular elements $C(x,\rho)$ is built from the distance map $D(X)$ which associates to each point - or pixel - x of X its distance ρ to the closest border (pixel) [37][38].

Among all the possible configurations, the chosen decomposition is the subset of the larger-without-recovering circles (cf. fig. 3) which can be defined by:

$$\begin{aligned} \bar{D}(X) = \{ (x,\rho) \in D(X) \mid & \\ \forall (x',\rho') \in \bar{D}(X) \mid (x',\rho') \neq (x,\rho), & \\ C(x,\rho) \cap C(x',\rho') = \emptyset & \\ \forall (x'',\rho'') \in D(X), \exists (x',\rho') \in \bar{D}(X) \mid & \\ \rho' \geq \rho'', C(x',\rho') \cap C(x'',\rho'') \neq \emptyset \} & \\ \text{with } C(x,\rho) = \{ x' \in X \mid |x' - x| < \rho \} & \end{aligned} \quad (2)$$

Decomposing a region with a collection of large circles provides potentially several advantages. First, the algorithm based on sorting of circles by both the decreasing radius and the increasing positions makes the decomposition robust to errors in the silhouette acquisition. Second, the spatial density of the root system is directly quantified by the histogram of circle radii.

The classification of holes by size gives pertinent indicators about the root system status or the structural waste. When these holes are enough large, we talk about macro-holes; they correspond most often to a loss of major root axes or to the anchorage strategy of the plant. They are called micro-holes when they are small, and correspond rather to the nutrient recovery potential. An automatic repartition of radii by a 2-means clustering [41] (cf. fig. 3) is used to distinguish macro- and micro holes respectively.

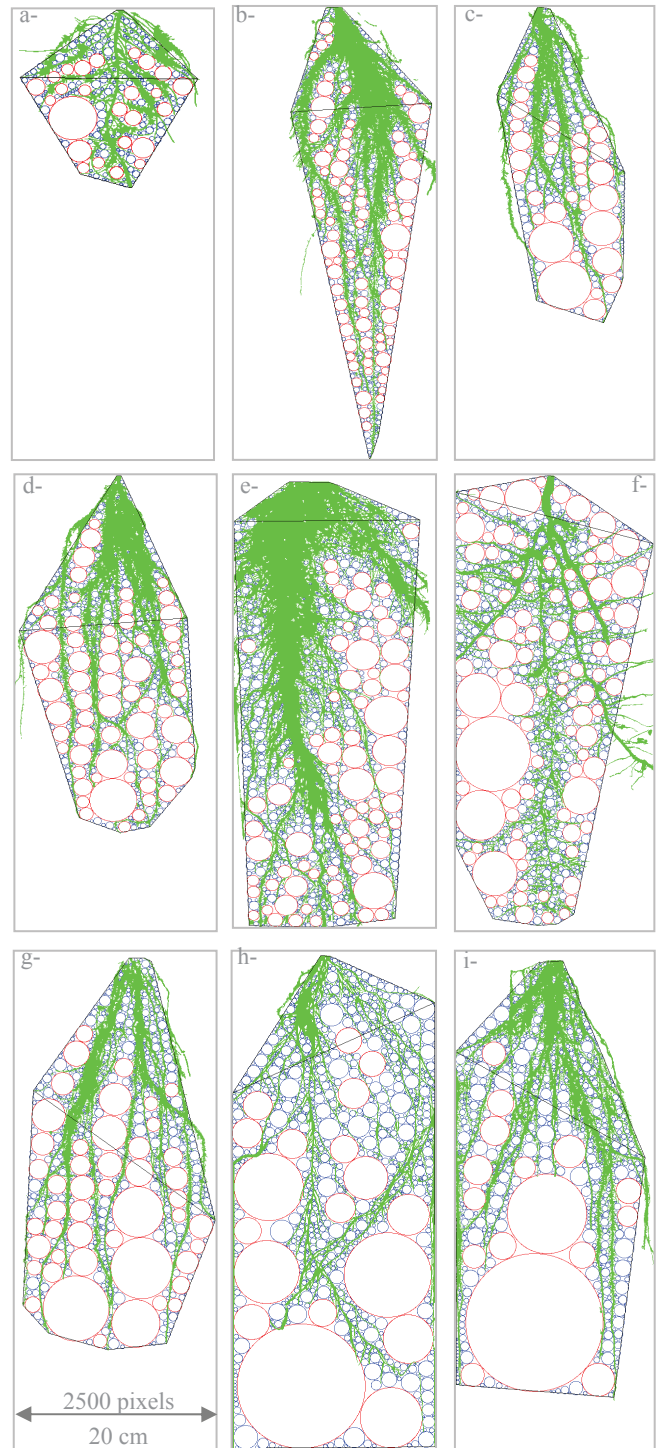


Fig. 3. “circular hole” distribution into the double-quadrangle-shaped polygon. The mature root system is represented in green, the micro-holes in blue and the macro-holes in red. a- 7-day-old *Arabidopsis thaliana*, b- 3-week-old Japonica rice (*Azucena*), c- 4-week old Indian rice, d- 4-week old wheat, e- 3-week-old sorghum, f- 6-week-old palm tree, g- 4-week-old eucalyptus, h- 6-week-old chickpea, i- 4-week-old Japonica rice (*Cicih Beton*).

C. Global features

Some geometrical characteristics can be easily measured from the double-quadrangle-shaped polygon (cf. fig. 4). UA and LA penetration angles are defined for the upper and lower parts of the root system from the polygon edges incident to the limit inter-parts. The width of the root system is approximated by the length of the inter-part segment considered as the basis of the quadrangles; each part is characterized by its surface US or DS, its height - given by the longest included segment perpendicular to the basis - and its hole size distribution.

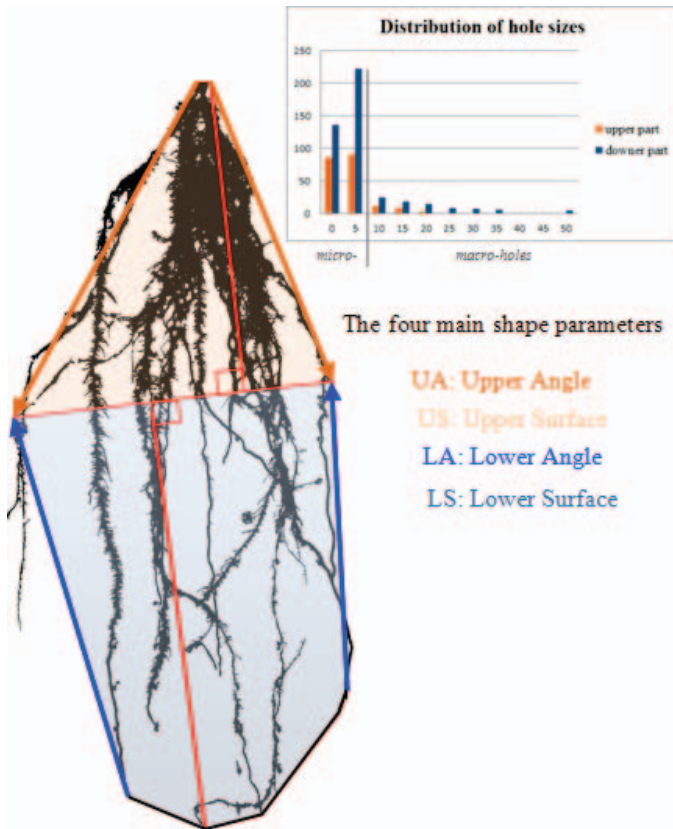


Fig. 4. Global features. The upper and lower penetration angles are respectively defined by the orange and blue arrows; width and heights are given by the red segments; the hole size distribution are described by an histogram in which the micro- and macro-holes are identified.

For the experimentation, we used a representative set composed of about 50 root system silhouettes of several species (and cultivars) – rice, wheat, maize, sorghum, chickpea, eucalyptus and palm tree – at different stages of growth – from 1 to 4 weeks. The data set has been doubled by left-to-right reversing these images. The plants were grown in Rhizoscope, a Cirad’s phenotyping platform developed for understanding the adaptation mechanisms of the cereal root system architecture under environmental constraints [42]. This platform is composed of several instrumented containers in which are immersed two-dimensional hydroponic-based systems called rhizoboxes; a rhizobox is a sandwich of two 50 cm x 20 cm x 2 cm Plexiglas plates filled with glass beads of 1.5 mm diameter that mimic soil resistance when root system is

growing (cf. fig. 5). Root growth can be examined and quantitative parameters can be collected after removing the glass beads. When the root systems are mature, they are photographed. The 15 mega-pixel high-quality colour images are automatically processed by GT-RootS [36], a dedicated software which can provide root system silhouette outputs.

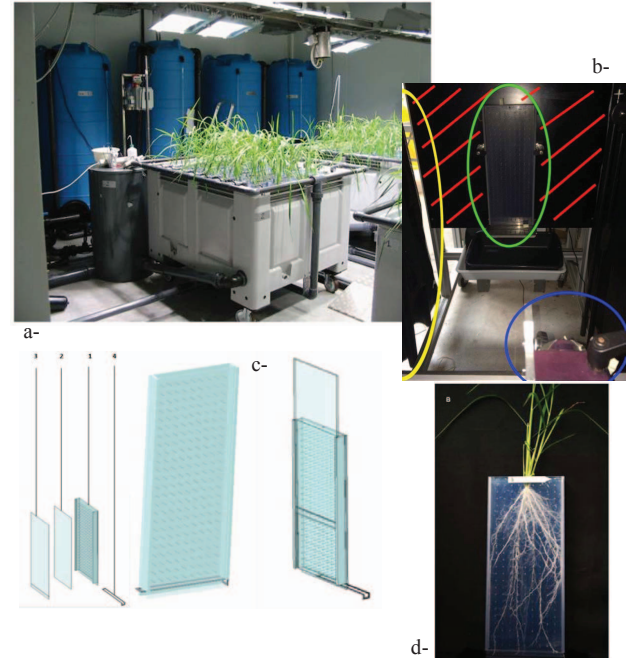


Fig. 5. General view of the Rhizoscope phenotyping system [40]. a- the growth chamber is composed of large hydroponic tanks where are immersed two-dimensional hydroponic-based boxes called rhizoboxes. b- the digitizing unit is a 1m³ black chamber composed of a rigid frame and dark panels or curtain in yellow: each rhizobox (in green) is positioned in front of a black background (in red); the camera (in blue) is fixed on the rigid frame. c- the rhizobox is composed of several movable elements which define a volume filled with glass beads: Plexiglas plates, grid of nails which holds the root system in place after bead removal, opening trap. d- colour image of a rhizobox and its mature rice root system.

III. RESULTS

This part is focused on the sensitivity of the double-quadrangle-shaped polygon to both the setting of the exclusion masks and the peripheral variations of root silhouettes. The study is interested in following the four main shape parameters: the upper and lower penetration angles *UA* and *LA*, and the upper and lower quadrangle surface *US* and *LS*.

A. Preamble: root penetration angle and area

The root penetration angulation (RPA) is a major but subjective parameter which can have different meanings or correspond to various geometric features (cf. fig. 6). It can be defined either as the width-based angle (WB) between vertical and the most external left and right crown roots (before roots reach the rhizobox sides and change direction)[40], either the angle between the Random Sample Consensus (Ransac) fit line at 10% depth of the root system and the horizontal soil line [27] or the angle defined by lateral edges of the trapezoid

covering the root upper compartment in the house-shaped polygon (HSP)[36] or in the double-quadrangle-shaped polygon (DQSP).

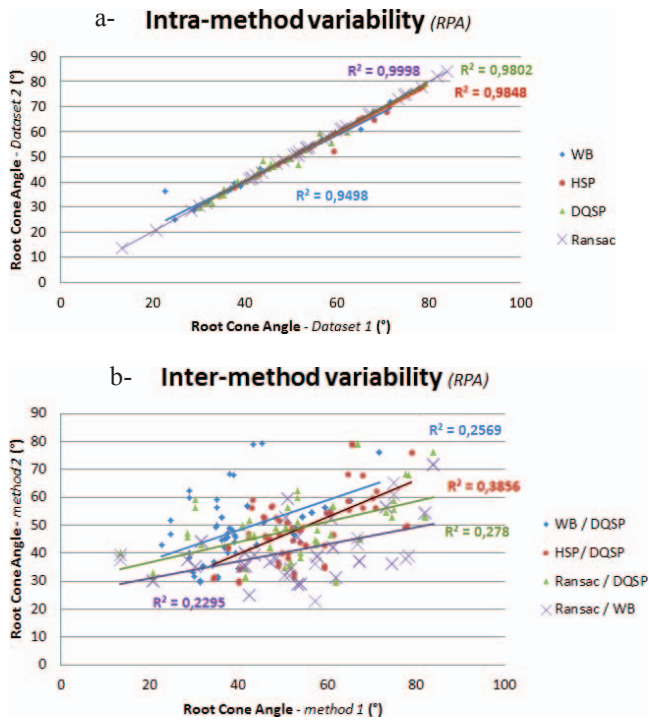


Fig. 6. Bias and variability of the root cone angle. *a-* For each method, we compare the root penetration angle obtained from the original images (dataset 1) and its mirrored image (dataset 2). With coefficients of determination closed to 1, the methods can be considered as invariable and repeatable. *b-* The cross-method comparisons do not enable to identify significant correlations between the different methods: the root penetration angle takes different meaning which corresponds to various geometric features.

The root angle penetration is difficult to formalize as shown in Figure 6. Different definition can be proposed, none of which is better. But, only the double-quadrangle-shaped polygon gives the possibility to evaluate a root closing angle to qualify the behaviour of the deep roots.

The volume of the global root system form is another key parameter for the study of the growth of mature system. This measure is different of the cumulated root volume as proposed in classic RSA analysis. In bi-dimensional study, the covering form of the root system will be characterized by its area; we call it the “root system form area” (RSFa). Figure 7 shows the comparison between areas of the different forms proposed to encapsulate the root system, especially the convex hull area (CHA), the house-shaped area (HSA) and the double-quadrangle-shaped area (DQSA).

Figure 7, the areas of root system form are compared with the whole dataset. The different methods are well correlated showing the trends are respected. The house-shaped polygon under-estimates the root system form area due to its squared

inferior compartment; the convex hull gives the reference area even if this method is well-known for its high spatial-sensitivity: a light offset in of the points of the convex hull leads to strong variation of area. The double-quadrangle-shaped polygon remains closed to the convex hull estimation as shown by a coefficient of determination greater than 0.97; due to its building process, it is also more stable than the convex hull.

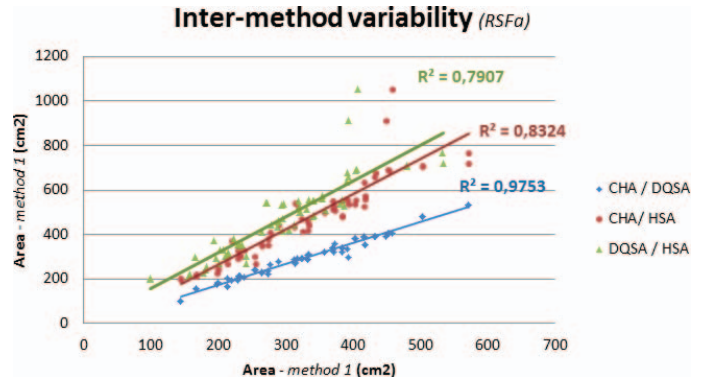


Fig. 7. Bias and variability of the root system form area.

B. Polygon sensitivity and exclusion mask

The double-quadrangle-shaped polygon construction needs masks to preserve the most significant geometrical landmarks of the root system. In this part, we explore the mean impact of their respective thickness onto (the variability of) the geometrical characteristics of the covering polygon.

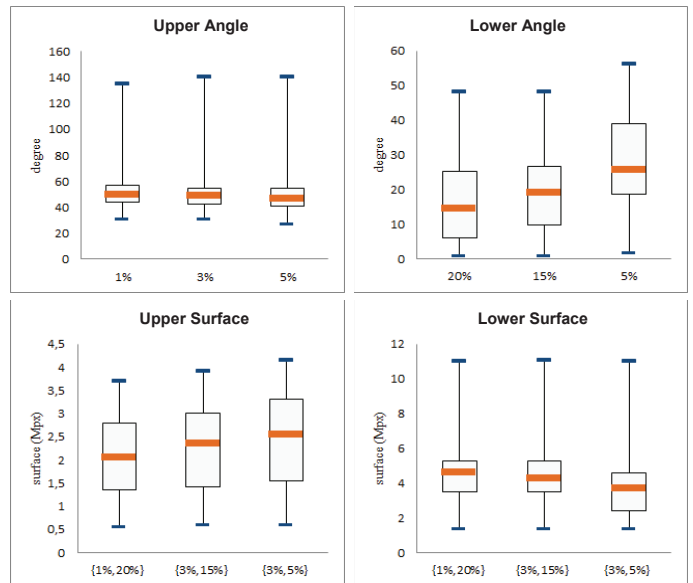


Fig. 8. Thickness sensitivity. The boxplot with whiskers illustrates the global behaviour of the data set with respect to the thickness variation of the upper and lower exclusion masks. The median value is represented in orange, the minimal and maximal values in blue, and the 2nd and 3rd quartile values define the grey rectangle. Significant variations appear, suggesting the sensitivity of form parameters relative to the thickness, except for the Upper Angle of penetration

The UA, LA, US and LS parameters are evaluated for each silhouette, the upper band thickness set respectively to 1%, 3% and 5% and the lower band thickness to 20%, 15% and 5%. The boxplots of Figure 8 integrate the measures evaluated on all the silhouettes of the data set.

UA: the variations observed in the top-left graph of Figure 8 are not significant. The lower, higher and median values are very close in the three experimentations; the distributions of the 2nd and 3rd quartile values are similar. The parameter can be considered as insensitive to the thickness of the upper band. This relative invariance is of course the consequence of the weak thickness amplitude between the three experimentations. But the upper band is narrow by nature because it allows just preserving the local shape of the root neck.

LA, US and LS: the graphics of the Figure 8 show significant differences in the measure distribution between the three experimentations. The size and position variations of the 2nd and 3rd quartile values illustrate the thickness sensitivity of the construction method. The geometrical comparisons between root systems should be done at constant setting of the horizontal masks.

C. Polygon sensitivity and silhouette smoothing

In this part, we explore how the smoothing of the root system silhouette impacts the shape of the double-quadrangle-shaped polygon and its geometric characteristics.

A Gaussian blur (*also known as Gaussian smoothing*) is applied to the binary image in order to reduce image noise – by example the isolated points – and reduce detail – by example the root hair. An automated threshold algorithm [41] is then used to divide the points of the 8-bit blurred image into two classes so that their combined spread (intra-class variance) is minimum. The result is a *de-noised* and smoothed silhouette of the root system as shown in Figure 9.

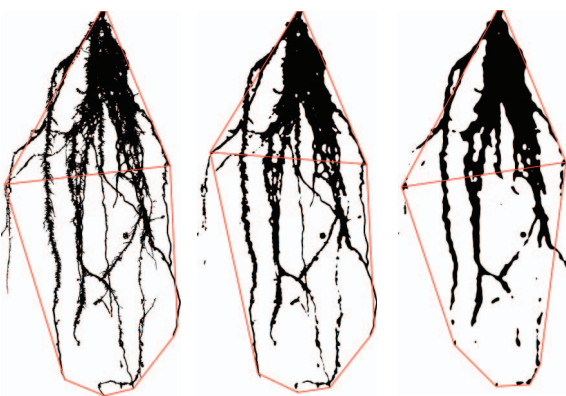


Fig. 9. Smoothed silhouette. The smoothing degree is determined by the standard deviation σ of the Gaussian distribution used for calculating the transformation to apply to each pixel of the binary image. The silhouette of the Figure 1 is progressively blurred with σ respectively set to 2, 10 and 20. The double-quadrangle-shaped polygon is represented in red.

The UA, LA, US and LS parameters are respectively evaluated onto two more or less smoothed silhouettes with Gaussian- σ respectively set to 2 and 10; the lower value addresses the impulsive noise, the higher of the root hair. The boxplots of Figure 10 integrate the measures evaluated on all the silhouettes of the data set.

The variations shown in Figure 10 are not significant. The lower, higher and median values between experimentations are very close; the distributions of the 2nd and 3rd quartile values are similar. Some differences are found, in particular a slight variation of minimum, median or maximum values: but the amplitude of these variations is too weak to constitute a clear trend. The heterogeneity of the data set has likely an equalising effect. Therefore, the shape parameters can be considered as insensitive to the smoothing of the root system silhouette.

The study was extended to the density parameter, and more exactly to the macro-hole distribution (cf. fig. 4 & 11). The decomposition of the background regions and the disk clustering are evaluated for two smoothing levels as previously. But here, the results are analysed plant-by-plant.

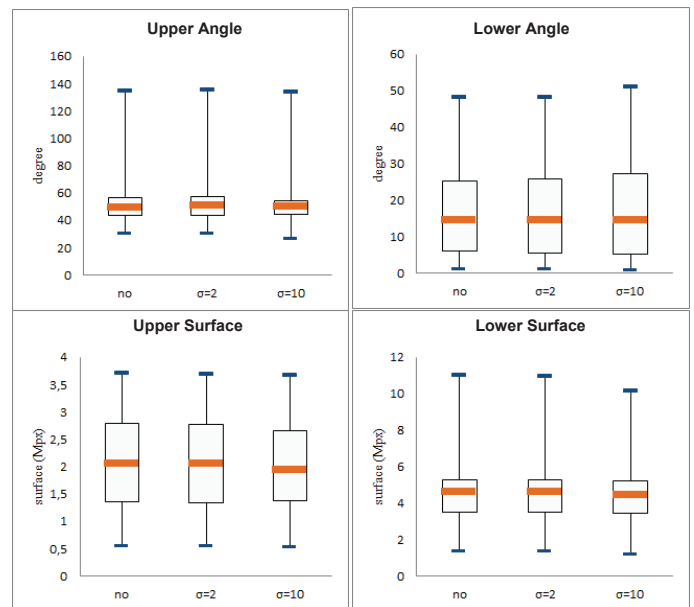


Fig. 10. The smoothing sensitivity of the shape parameters. The boxplots illustrate the impact of the Gaussian blurring on the shape parameters computed from the data set. Each parameter is re-evaluated on more and more smoothed root silhouette, the Gaussian- σ respectively set to 2 and 10. No significant variation appears.

The restitution of results concerns only the macro-hole classes. The micro-hole distributions are indeed not enough discriminant between the upper and lower parts of the root systems to be really informative.

Figure 11 shows significant impacts of smoothing degree on the macro-hole distribution: minimum, median and

maximum values change according to the value of the Gaussian- σ . The macro-hole distribution is weakly sensitive to the cleaning process applied to the root silhouette. The appreciation of the smoothing-sensitivity depends on the considered root system as illustrated in Figure 11: the two root systems present smoothing-sensitivities which may seem different preventing to draw any conclusion. But, the main point is that this sensitivity is significantly the same in the upper and lower parts of the root system. The observed variations in each part are following the same trend. The comparison between the upper and lower macro-hole distributions is so an insensitive indicator which could be used as signature of the root system density.

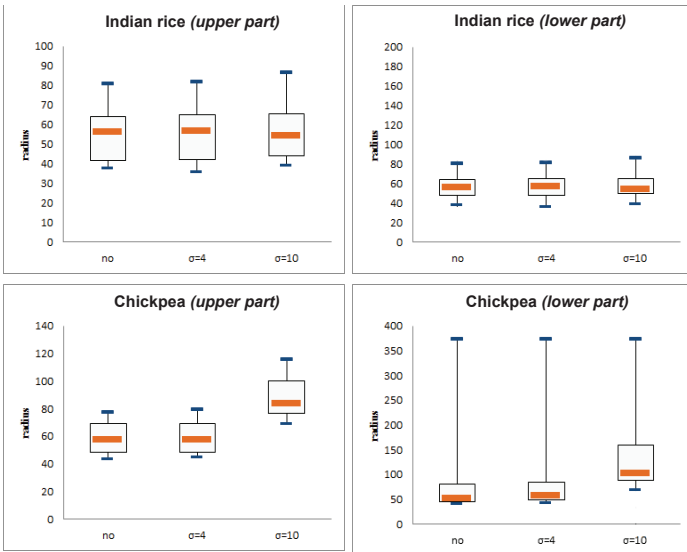


Fig. 11. The smoothing sensitivity of the macro-hole distribution. *Boxplots illustrate the impact of Gaussian blurring onto macro-hole distribution of Indian rice (cf. fig. 3.d) and Chickpea (cf. fig. 3.h). The macro-hole distribution is re-evaluated on more and more smoothed root silhouette, the Gaussian- σ respectively set to 4 and 10. Significant variations appear but trends are similar in upper and lower parts of the root system.*

IV. DISCUSSION

The idea is to progressively reduce the convex hull of the root system to a double-quadrangle-shaped polygon by iterative suppression of least significant vertices to only keep the major features of the global form. For this, we combine the root recovery rate and the surface loss rate. Horizontal exclusion masks are introduced to conserve the main geometrical landmarks of the root system, especially around the principal root neck and meristems. The resulting form is a closed polygon which best covers the root system.

This double-quadrangle-shaped polygon reflects implicitly the strategy of the root system growth: its geometrical properties leads to a significant differentiation between the topsoil root systems and the deep root systems. The major interest of the covering polygon is to classify the root system behaviours based on a very little number of parameters: penetration angulation and root level heights. This reduced

description is in opposition with the RSA approaches which are rather based on many accurate and fine measurements of the root axes [15][16][27] as mean or cumulated lengths, distribution of root diameters, and profile of root densities. These parameters characterize mainly the functional capability of the plant and not directly its growing strategy. Moreover, unlike our method, RSA approaches are limited to the study of scattered root systems as they are based on a structural decomposition of the root system.

Researchers who study occupation of the root volume often use the convex hull to delimit the root system space. But it is particularly sensitive to small peripheral geometrical variations of the root system. In our method, the mechanism of reduction of the convex hull leads to a more robust covering contour. This stability is illustrated by the small variation of measures after progressive degradations of the root system (Figures 9 and 10).

The double-quadrangle-shaped polygon defines a robust area where root density can be estimated. For this, we propose not to use directional approaches as [36] but to study the distribution of holes which is independent of any privileged orientations of the container where grows the root system.

The spatial distribution the root density is addressed from the spatial repartition of background regions with respect to their size and form. We decompose the complex shaped background regions into a collection of simple structuring elements. We chose to use circles which correspond intuitively to hole notion. An automated repartition of circles by 2-means clustering was introduced for a best appreciation of macro- and micro-holes, respectively associated to the anchorage and nutrient recovery potentials. The originality of the method arises from the way of considering holes. Hole is considered here as neither a form nor as an area but as a sum of circular structuring elements. This very local vision results in the localization of micro-holes which correspond to dense root mesh and of macro-holes which emphasize large root mesh, as shown in Figure 3. The distribution of the circular element diameters is a good indicator of the root system compactness and probably of the plant ability to explore soil and find nutrients.

Lastly, Figures 6 and 7 illustrate the difficulty to define some notions, especially the penetration angular. Several definitions have been proposed as [27] [36][40] but none is really adopted by the scientific community. Ours has the advantage to be easy to understand because it is defined from (the slopes of) the double-quadrangle-shaped polygon and is easy to display graphically.

The double-quadrangle-shaped polygon is well-adapted in measuring the global feature of dense plant root system form. The proposed definitions are easily understandable and parameters can be displayed on the root system which facilitates the validation or invalidation of numerical measures.

Two aspects will be developed in further work.

Firstly, some local indicators should be introduced, especially the usual measurements of root axes: length and diameter classes and orders. This requires a structural

segmentation in which each root axis is identified. Back-tracking a root from its deepest level to its origin is possible and would make it possible to build the root system topology. Such information could significantly increase the accuracy of deep root counting, and perhaps address some questions about root architecture units [20] or about morphology and functioning aspects.

Secondly, the distribution of densities – or macro-holes – is a too global indicator. Characterizing the spatial distribution of macro-holes remains a major challenge to detect and measure the heterogeneity of the root architecture. The recursive decomposition of the quadrangles should refine the root system signature.

V. CONCLUSION

In this paper, we proposed a new global approach to define the growth area of the root system from a covering polygon composed of two superimposed quadrangles to evaluate geometry features or to classify root systems. We presented also an original method based on the decomposition of background regions in circular structuring elements to estimate the root system porosity.

The global parameters / features of the root system form are evaluated from the main geometry and density properties of the double-quadrangle-shaped polygon. Upper and lower penetration / closing angles or macro- and micro-hole distributions were evaluated for characterizing the plant strategy and ability of soil exploration.

Experimental studies were focused on the geometry-sensitivity of the double-quadrangle-shaped polygon. Several experimentations were realized using a representative set composed of 100 root system silhouettes of several species – *rice, wheat, maize, sorghum, and palm tree* – at different stages of growth. We demonstrated that the major parameters were enough invariant to be used as signature of the root system.

ACKNOWLEDGMENT

We wish to thank Christophe Jourdan and Hervé Rey for their constructive exchanges about root system architecture, Audrey Dardou for its expertise required for the comparative study. Lastly, we should like to thank the reviewers for their detailed comments and suggestions for improving the paper. This work was carried as part of the framework of the RoSoM project. This project is supported by Agropolis Fondation under the reference ID 1202-073 through the “Investissements d’avenir” programme (Labex Agro: ANR-10-LABX-001-01).

REFERENCES

- [1] M.L. Parry, C. Rosenzweig, A. Iglesias, M. Livermore, and G. Fischer, “Effects of climate change on global food production under SRES emissions and socio-economic scenarios”, *Global Environmental Change*, 14(1), 53-67, 2004.
- [2] C. Lesk, P. Rowhani, and N. Ramankutty, “Influence of extreme weather disasters on global crop production”, *Nature*, 529(7584), 84-87, January 2016.
- [3] P.J. Gregory, A.G. Bengough, D. Grinev, S. Schmidt, W.B. Thomas, T. Wojciechowski, and I.M. Young, “Root phenomics of crops: opportunities and challenges”, *Functional Plant Biology*, 36(11), 922-929, 2009.
- [4] L.H. Comas, S.R. Becker, V.C. Von Mark, P.F. Byrne, and D.A. Dierig, “Root traits contributing to plant productivity under drought”, *Ecophysiology of root systems-environment interaction*, 18, 2014.
- [5] J.L. Araus and J.E. Cairns, “Field high-throughput phenotyping: the new crop breeding frontier”, *Trends in Plant Science*, 19(1), 52-61, 2014.
- [6] C. Granier and D. Vile, “Phenotyping and beyond: modelling the relationships between traits”, *Current Opinion in Plant Biology*, 18, 96-102, 2014.
- [7] L. Li, Q. Zhang, and D. Huang, “A review of imaging techniques for plant phenotyping”, *Sensors*, 14(11), 20078-20111, 2014.
- [8] J.F. Humplik, D. Lazár, A. Husičková, and L. Spichal, “Automated phenotyping of plant shoots using imaging methods for analysis of plant stress responses—a review”, *Plant methods*, 11(1), 1, 2015.
- [9] A. Skirycz, K. Vandenbroucke, P. Clauw, K. Maleux, B. De Meyer, S. Dhondt, A. Pucci, N. Gonzalez, F. Hoerberichts, V.B. Tognetti, M., Galbiati, C. Tonelli, F. Van Breusegem, M. Vuylsteke, and D. Inzé, “Survival and growth of Arabidopsis plants given limited water are not equal”, *Nature biotechnology*, 29(3), 212-214, 2011.
- [10] S. Tisné, Y. Serrand, L. Bach, E. Gilbault, R. Ben Ameer, H. Balasse, and G. Chareyron, “Phenoscope: an automated large-scale phenotyping platform offering high spatial homogeneity”, *The Plant Journal*, 74(3), 534-544, 2013.
- [11] L. Mathieu, G. Lobet, P. Tocquin, and C. Périlleux, “Rhizoponics: a novel hydroponic rhizotron for root system analyses on mature Arabidopsis thaliana plants”, *Plant Methods*, 11(1), 3, 2015.
- [12] A.S. Iyer-Pascuzzi, O. Symonova, Y. Mileyko, Y. Hao, H. Belcher, J. Harer, J.S. Weitz, and P.N. Benfey, “Imaging and analysis platform for automatic phenotyping and trait ranking of plant root systems”, *Plant Physiology*, 152(3), 1148-1157, 2010.
- [13] R.T. Clark, A.N. Famoso, K. Zhao, J.E. Shaff, E.J. Craft, C.D. Bustamante, S.R. Mccouch, D.J. Aneshansley, and L.V. Kochian, “High-throughput two-dimensional root system phenotyping platform facilitates genetic analysis of root growth and development”, *Plant, cell & environment*, 36(2), 454-466, 2013.
- [14] M.O. Adu, A. Chatot, L. Wiesel, M.J. Bennett, M.R. Broadley, P.J. White, and L.X. Dupuy, “A scanner system for high-resolution quantification of variation in root growth dynamics of Brassica rapa genotypes”, *Journal of experimental botany*, eru048, 2014.
- [15] S.M. Rich, and M. Watt, “Soil conditions and cereal root system architecture: review and considerations for linking Darwin and Weaver”, *Journal of experimental botany*, 64(5), 1193-1208, 2013.
- [16] L.V. Kochian, “Root architecture”, *Journal of Integrative Plant Biology*, 58(3), 190-192, 2016.
- [17] J. Lynch, “Root architecture and plant productivity”, *Plant Physiology*, 109, 7-13, 1995.
- [18] W. Böhm, “Methods of studying root systems”, (33), 2012. Springer Science & Business Media.
- [19] R.W. Zobel, and Y. Waisel, “A plant root system architectural taxonomy: a framework for root nomenclature”, *Plant Biosystems*, 144(2), 507-512, 2010.
- [20] C. Jourdan, and H. Rey, “Architecture and development of the oil-palm (*Elaeis guineensis* Jacq.) root system”, *Plant and Soil*, 189(1), 33-48, 1997.

- [21] G. Bodner, D. Leitner, A. Nakhforoosh, M. Sobotik, K. Moder, K., and H.P. Kaul, "A statistical approach to root system classification", *Frontiers in Plant Sciences*, 292(4), 2013.
- [22] L. Kutschera, and E. Lichtenegger, "Bewurzelung von Pflanzen in Verschiedenen Lebensräumen", *Stapfia*49, Guttenberg/Linz: Land Oberösterreich, 1997.
- [23] A. French, S. Ubeda-Tomás, T.J. Holman, M.J. Bennett, and T. Pridmore, "High-Throughput Quantification of Root Growth Using a Novel Image-Analysis Tool". *Plant Physiol.* 150: 1784-1795, 2009
- [24] P. Armengaud, K. Zambaux, A. Hills, R. Sulpice, R.J. Pattison, M.R. Blatt, and A. Amtmann, "EZ-Rhizo: integrated software for the fast and accurate measurement of root system architecture", *The Plant Journal*, 57(5), 945-956, 2009.
- [25] T. Galkovskiy, Y. Mileyko, A. Bucksch, B. Moore, O. Symonova, C.A., Price, C.N. Topp, A.S. Iyer-Pascuzzi, P.R. Zurek, S. Fang, J. Harer, P.N. Benfey, and J.S. Weitz, "GiA Roots: software for the high throughput analysis of plant root system architecture", *BMC plant biology*, 12(1), 116, 2012.
- [26] T. Alberda, "Growth and root development of lowland rice and its relation to oxygen supply", *Plant and soil*, 5(1), 1-28, 1953.
- [27] A. Das, H. Schneider, J. Burrige, A.K.M. Ascanio, T. Wojciechowski, C.N. Topp, J.P. Lynch, J.S. Weitz, and A. Bucksch, "Digital imaging of root traits (DIRT): a high-throughput computing and collaboration platform for field-based root phenomics". *Plant methods*, 11(1), 1, 2015.
- [28] M.P. Pound, A.P. French, J.A. Atkinson, D.M. Wells, M.J. Bennett, and T. Pridmore, "RootNav: navigating images of complex root architectures", *Plant Physiology*, 162(4), 1802-1814, 2013.
- [29] J. Pace, N. Lee, H.S. Naik, B. Ganapathysubramanian, and T. Lübberstedt, "Analysis of maize (*Zea mays* L.) seedling roots with the high-throughput image analysis tool ARIA (Automatic Root Image Analysis)", In *Plos One* DOI: 10.1371/journal.pone.0108255, 2014.
- [30] R. Rellán-Álvarez, G. Lobet, H. Lindner, P.L. Pradier, J. Sebastian, M.C. Yee, Y. Geng, C. Trontin, T. LaRue, A. Schrager-Lavelle, C.H. Haney, R. Nieu, J. Maloof, J.P. Vogel, and J.R. Dinneny, "GLO-Roots: an imaging platform enabling multidimensional characterization of soil-grown root systems", *Elife*, 4, e07597, 2015.
- [31] D. Ristova, U. Rosas, G. Krouk, S. Ruffel, K.D. Birnbaum, and G.M. Coruzzi, "RootScape: a landmark-based system for rapid screening of root architecture in *Arabidopsis*", *Plant Physiology*, 161(3), 1086-1096, 2013.
- [32] J. Diener, P. Nacry, C. Périn, A. Diévar, X. Draye, F. Boudon, A. Gojon, B. Muller, C. Pradal and C. Godin, "An automated image-processing pipeline for high-throughput analysis of root architecture in OpenAlea. 7th International Conference On Functional-Structural Plant Models, Saariselkä, Finland, 85-87, 2023.
- [33] D. Leitner, B. Felderer, P. Vontobel, and A. Schnepf, "Recovering root system traits using image analysis exemplified by two-dimensional neutron radiography images of lupine". *Plant Physiol* 164: 24–35, 2014
- [34] J. Cai, Z. Zeng, J.N. Connor, C.Y. Huang, V. Melino, P. Kumar, and S.J. Miklavcic. "RootGraph: a graphic optimization tool for automated image analysis of plant roots". *J. Exp. Bot.* 66 (21): 6551-6562, 2015
- [35] A. Bucksch, J. Burrige, L.M. York, A. Das, E. Nord, J.S. Weitz and J.P. Lynch, "Image-based high-throughput field phenotyping of crop roots". *Plant Physiology*, 166(2), 470-486, 2014.
- [36] P. Borianne, G. Subsol, F. Fallavier, A. Dardou, and A. Audebert, "GT-RootS, Global Traits of Root System: an integrated software for an automated root system measurement from high-throughput phenotyping platform images", *JXBot*, 2016, unpublished.
- [37] R. Kimmel, N. Kiryati, A.M. Bruckstein, "Sub-pixel distance maps and weighted distance transforms", *Journal of Mathematical Imaging and Vision*, 6(2-3), 223-233, 1996.
- [38] A. Hajdu, L. Hajdu, L., and R. Tijdeman, "Approximations of the Euclidean distance by chamfer distances", *arXiv preprint arXiv:1201.0876*, 2012.
- [39] E.W. Forgy, "Cluster analysis of multivariate data: efficiency versus interpretability of classifications", *Biometrics* 21: 768–769, 1965.
- [40] B. Courtois, A. Audebert, A. Dardou, S. Roques, T. Ghneim-Herrera, G. Droc, J. Frouin, L. Rouan, E. Gozé, A. Kilia, N. Ahmadi, and M. Dingkuhn, "Genome-wide association mapping of root traits in a japonica rice panel." *PLoS ONE* 8(11): e78037, 2013.
- [41] N. Otsu., "A threshold selection method from grey scale histogram", *IEEE Trans. on Syst. Man and Cyber.* 1, 62-66, 1979.