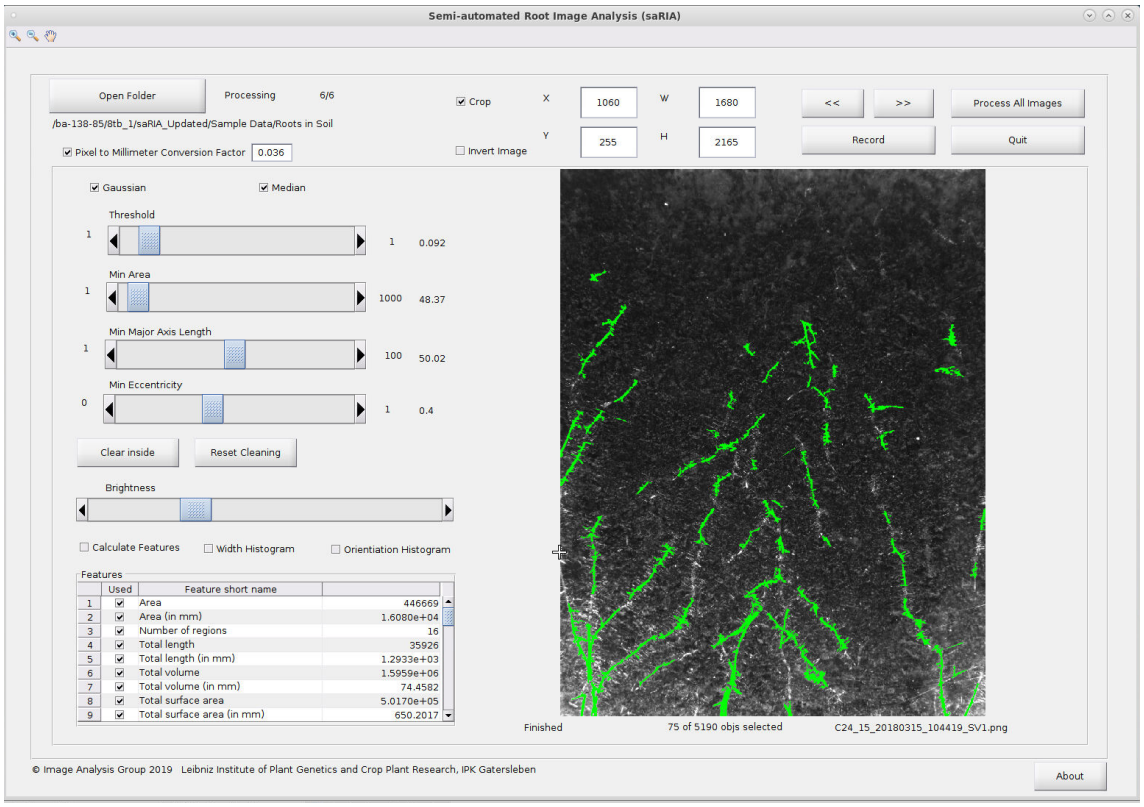
# Semi-automated Root Image Analysis
# saRIA v0.1 – Quick Guide

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# A semi-automated image analysis tool to extract root traits under different environments





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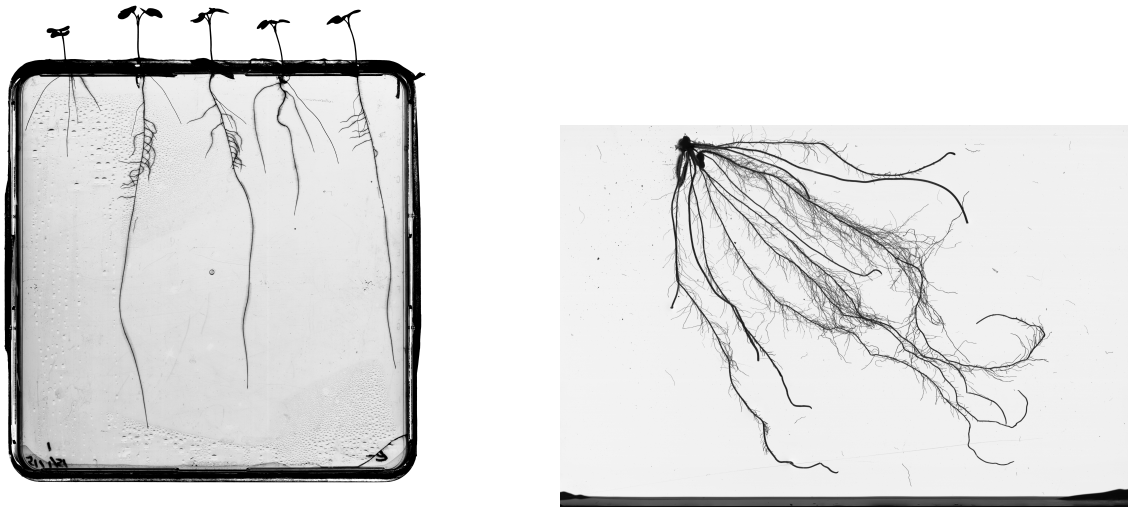

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# 1 Introduction

saRIA stands for the Semi-automated Root Image Analysis. The saRIA is a software tool to automate and facilitate the large-scale of root networks. It is designed to help sceinitsts or biologists quantify the plant root system architecture in agar, soil and washed roots, regardless of their prior training in mathmatics and coomputer sceince.

Like many useful peices of software, saRIA is originated from a question: how saRIA is different compared to existing tools? The optimistic answer is monitoring roots in natural environment is very important to quantitatively assess the development of the root system and its intraction with the soil. Since the the roots have low contrast and high level of noise in the soil background, it became one of the most challenging computer vision problem in plant sceince.

The saRIA tool was developed with in the scope of a PhD thesis aiming to quantifying the root sysytem architecture from natural environmnet like soil and also roots grown in artificial substrate (Ex: Agar roots) and scanned roots. In addition, this tool is capable of extracting root traits (like area, length, volume) for the plant biomass analysis. The example images of roots grown in agar, scanned roots are shown in figure 1 and roots grown in soil is shown in figure 2.



(a) Roots grown in agar

(b) Scanned Roots

Figure 1: Clearly visible roots

## 1.1 Key Features

The saRIA tool is implemeted to semi-automatocally process series of soil-root images (a set of images in a folder) or only one image at a time per user click in order to estimate root traits for each image.

- User defined values for image cropping (Crop)
- Invert image for roots grown in agar and scanned roots (Invert Image)


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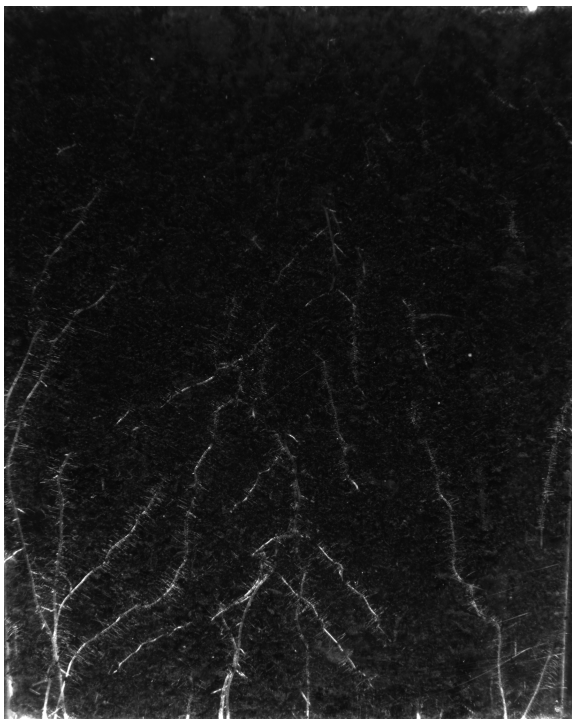


Figure 2: The Arabidopsis roots grown in soil environment.

- Deselection and undo of non-root pixles in the segmented image to erase root like noisy structures (Clear Inside and Reset Cleaning)
- The calculated root traits as follows:
  - Area : Total number of root pixels in the image
  - Number of regions: Total number of roots in the image
  - Total Length: Total length of roots (in pixels and millimeter) in the image
  - Total Volume: Total volume of roots (in pixels and millimeter) in the image for biomass calculation
  - Total Surface Area: The surface area of all roots (in pixels and millimeter) in the image.
  - Specific Root Length: The ratio of total length and total volume (in pixels and millimeter) of root pixels
  - Number of branching points and end points of the roots
  - Statisitcal analysis of geometrical distribution of roots
  - Statisitcal analysis of width and orientation of root pixels
- User defined value for the calculation of root traits in metric units (Pixel to Millimeter Conversion Factor)
- Calculation and storage of (done for each image - click Record button):


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- segmented, segmentation cleaned, feature binary image and color representation of roots on original image
- root traits of all images in a CSV file
- root widths and orientations histogram

The user can adjust diverse algorithmic parameters to influence the behaviour of the calculation.

# 2 Quick Start

## 2.1 How to install?

After unpacking the zip archive following two folders will be generated:

```
root
├── saRIA
└── quickGuide
```

The saRIA tool folder contains the pre-compiled executable of the computer program, example image files (sample data), a readme- and a license file. Please, read both text files carefully before starting the program. The *quickGuide* folder contains a copy of this file.

## 2.2 How to run?

The saRIA tool comes compiled in two versions, one for Linux- and one for Windows-based operation systems, respectively. To run the program the user has to install the MATLAB Runtime Environment. Since the saRIA tool was developed, tested and compiled under MATLAB 2018b, we recommend to install exactly the same version, i.e. MCR 2018b, which can be downloaded from the official MATLAB side Install and Configure the MATLAB Runtime.

### 2.2.1 Linux

Under Linux-based operation systems one has to open a terminal and switch to the folder which contains the saRIA tool . Then type

```
./run_saRIA.sh /path/to/your/MATLAB/Runtime/v95
```

where */path/to/your/MATLAB/Runtime/v95* specifies the path to the locally installed MATLAB Runtime Environment (version 2018b - v95).

### 2.2.2 Windows

To run the program under Windows double-click on the icon of the provided executable in the Windows file explorer or start it with its name from the command line.


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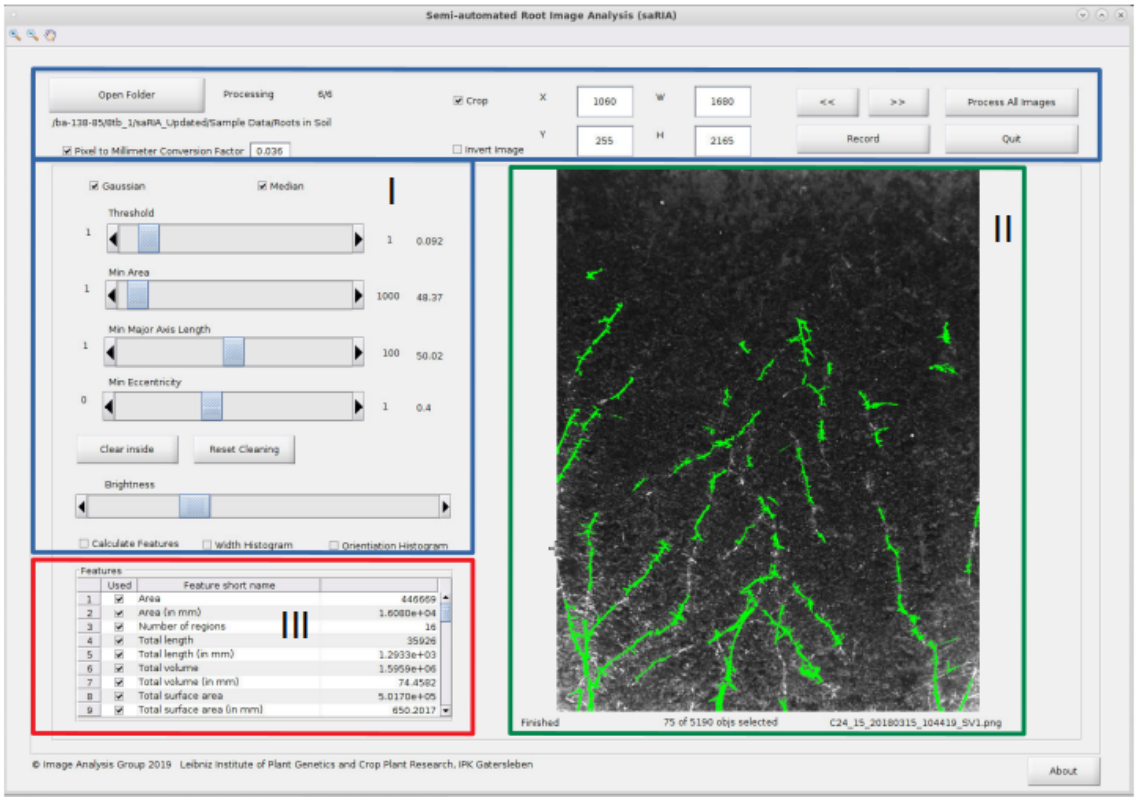



Figure 3: The graphical user interface (GUI) of the saRIA tool .


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## 2.3 The Interface Layout

The major elements of the software interface include an **Input-area** at the upper part and an **Output-area** at the lower part of the GUI as shown in Fig. 3.

At the input area (Fig. 3 - I, blue color rectangle boxes), the user can select the folder containing the input images, define the underlying crop parameters and set the algorithmic parameters. Also, user can select/deselect the Invert Image (root image type), calculation of root traits, pixel to millimeter conversion factor and generation of width and orientation histogram. By clicking the Process All Images button, all images of the input folder are processed according to the defined settings.

The example image for color overlay of roots on original image and processed binary images named segmented, segmentation cleaned and features image are displayed by single user click on grid layout in GUI (Fig. 3 - II, green color rectangle box). Also, name of the image, number of detected objects among segmented objects and a live feedback at the lower part of the GUI (Fig. 3 - II) are given during the whole process of calculation. The calculated root traits are displayed on features table in the GUI (Fig.3- III, red color rectangle box).

## 2.4 First Run

The typical steps to analyse an experiment (a series of images) are:

1. select an input folder (containing a time series of images)
2. (optional) If the input image is agar or scanned roots, select invert image checkbox
3. (optional) adapt the parameter settings
4. run the analysis
5. define the cut-off values according to the given preview data (to get rid of false results)
6. (optional) still non-root pixels are present in final segmented image, click on Clear Inside button and select region on image to remove non-root pixels.
7. (optional) preview of segmented, segmentation cleaned and feature binary image can be seen using right-click on image as shown in figure 4, 5, 6, 7.
8. accept or withdraw these calculations; when accepted (Record) the results are stored within the used created target folder (results), if not the user has possibility to go back to step 5 to define new cut-off values

To run the program, the user has to select the input folder of the images. Once the folder was found and successfully imported, the images of the selected folder are automatically analysed to calculate the root traits with the used configuration. The user now has the chance to adapt some algorithmic parameters. These parameters are:

**Threshold** Threshold is an image segmentation parameter which ranges from 0 to 1. Depending on image intensity and noise level, this parameter need to be adapted dynamically for efficient results. The preview of the selected image is automatically updated after every change.


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**Min Area** Area defines the number of pixels of the binary object. To avoid false detections, the binary objects with more than minimum number of pixels are considered for root trait extraction. It ranges from 0 to 1000 on scale.

**Min Major Axis Length** Major axis length defines the length of the binary object. This parameter avoids the small false structures like roots for root trait extraction. It ranges from 0 to 100 on scale.

**Min Eccentricty** Eccentricty defines structure of the binary object. In genral, circle objects tends to have low value of eccentricity (approximately zero). By adapting this parameter, the non-root structures can be avoided for root trait calculation. It ranges from 0 to 1 on scale.

In addition to the parameter selection, the user can also select the required traits from the output feature list. In particular, the orientation of roots requires parallel processing units for the computation. Therefore, the user can deselect this option (if unnecessary) for the fast computation of other root traits in the feature list. Moreover, the user can load the stored parameter configuration of previously segmented images for further improvement of root traits.

## 3 Provided example data

The saRIA tool comes with three types of example images with different folder names in parent folder named Sample Data.

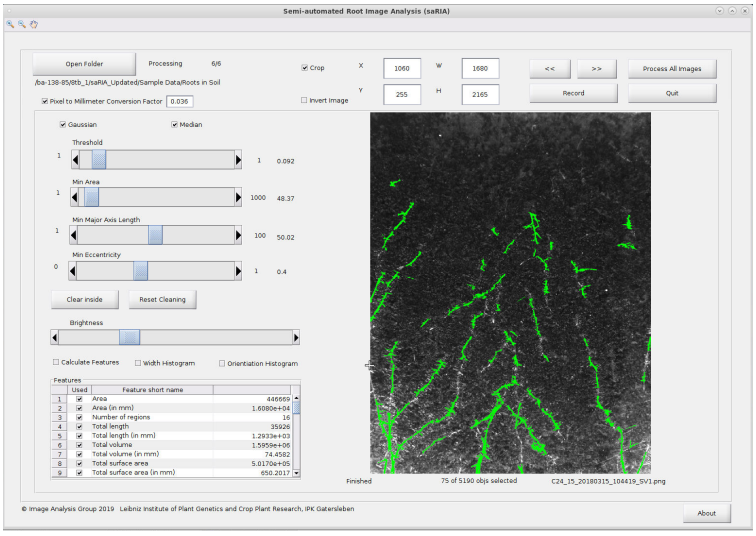



Figure 4: The feature extraction of an Arabidopsis plant roots grown in soil.

For any queries please contact us at address mentioned on project page https://ag-ba.ipk-gatersleben.de/saRIA.html


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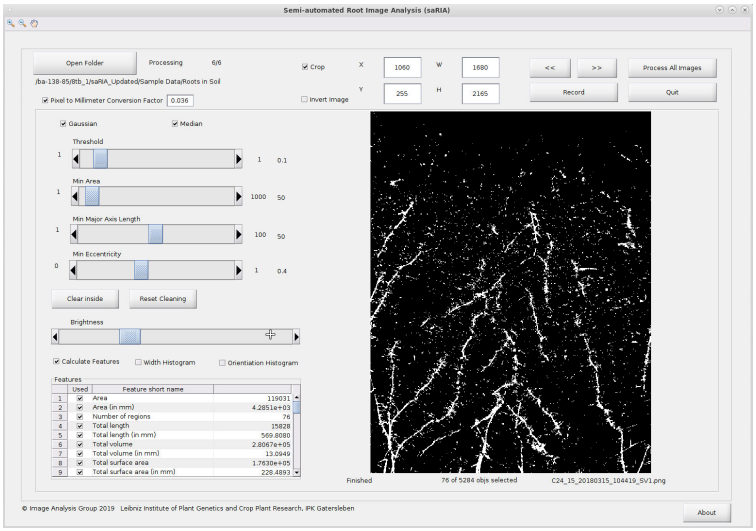



Figure 5: The segmented binary image of an Arabidopsis plant.

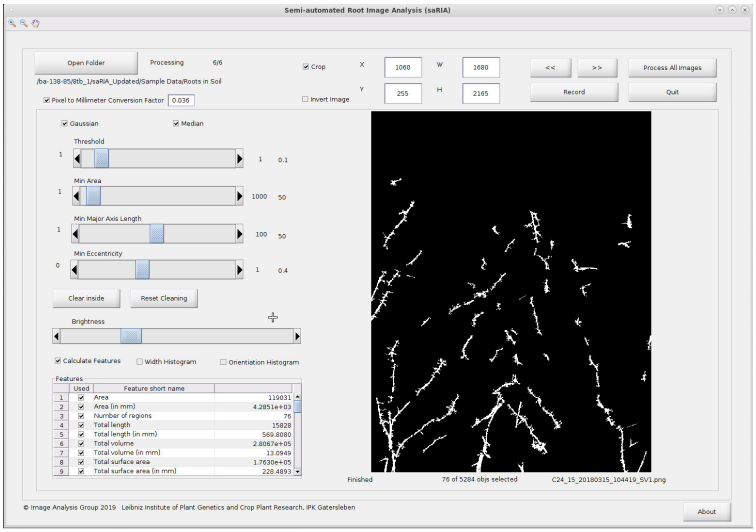



Figure 6: The cleaned segmented binary image of an Arabidopsis plant.


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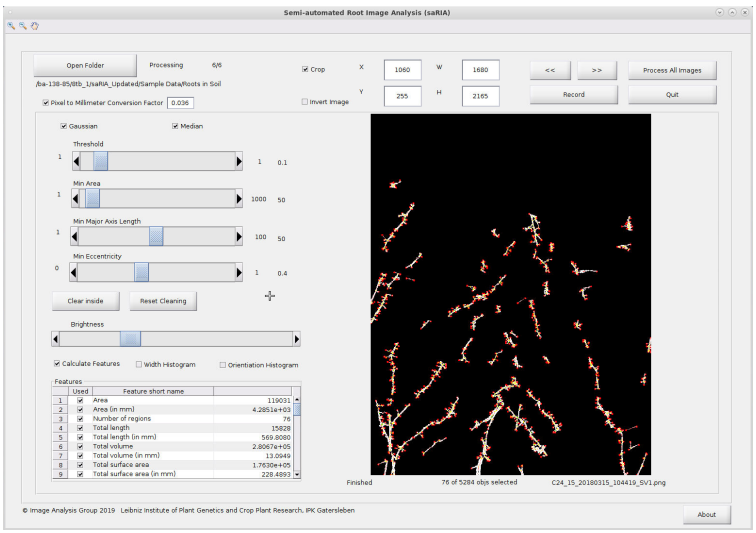



Figure 7: The root feature image of an Arabidopsis plant.

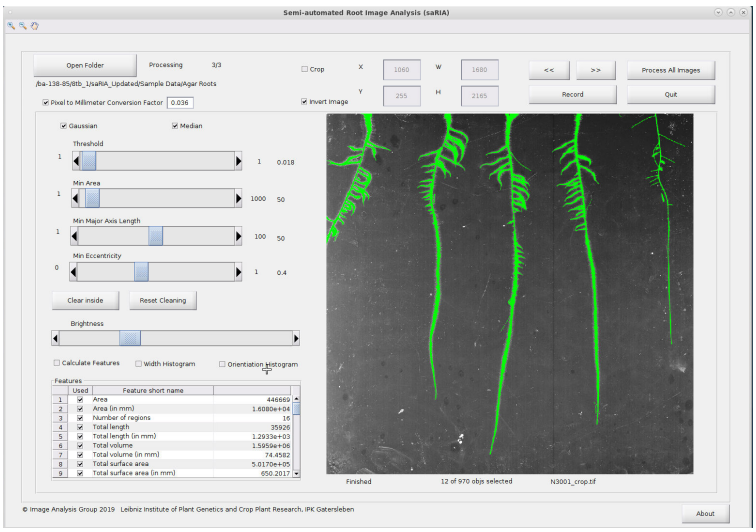



Figure 8: The feature extraction of roots grown in agar gel.

# A Funding

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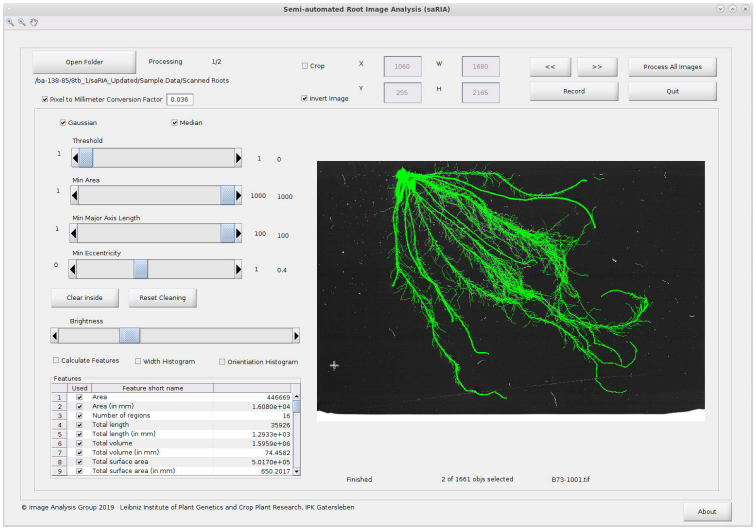



Figure 9: The feature extraction of scanned roots.

## B Acknowledgments

We would like to thank Molecular Gentetics Group, Leibniz Institute of Plant Genetics and Crop Plant Research (IPK Gatersleben) for providing image data for the analysis.

## C References

[1] Henke, M., Narendra, N., Gladilin, E., paper: todo, *in preparation*

## D Terms of use

1. The saRIA tool and the example image data are distributed for non-commercial usage WITHOUT ANY WARRANTY under the terms described in the EULA license. See the included *EULA.txt* file for details.

2. The user manual is intellectual property of the Image Analysis Group of the IPK Gatersleben. The user may download and use the tool and information available on our web site.

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