

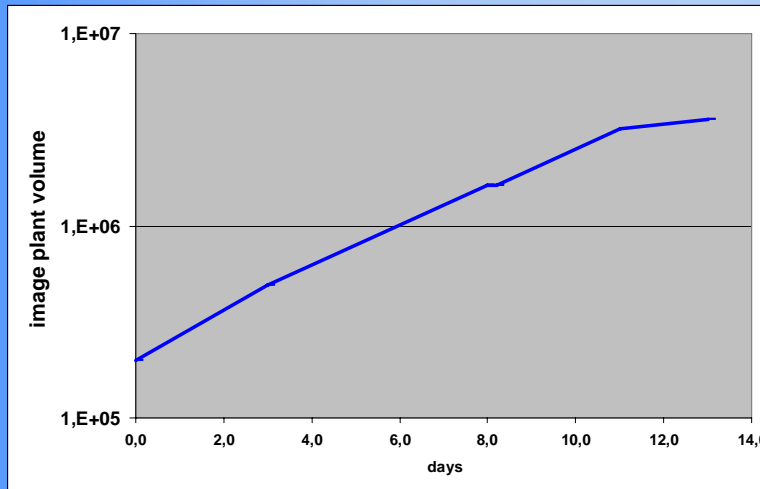
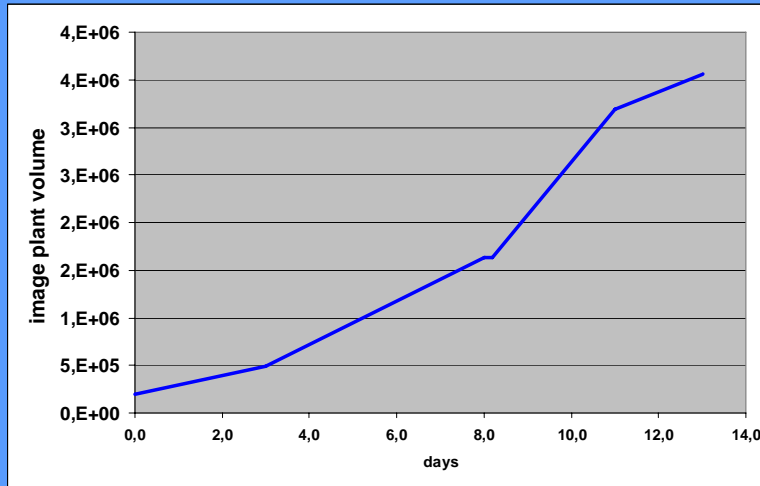


Get an impression of your picture!

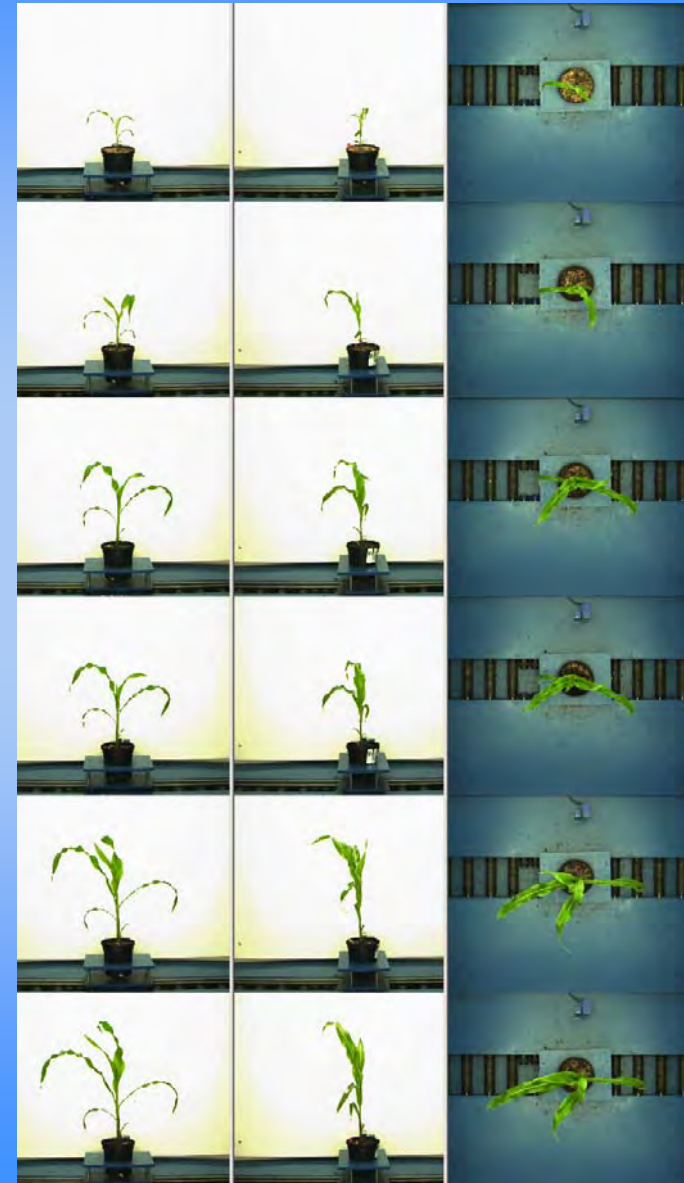
Interpretation of leaf areas of corn

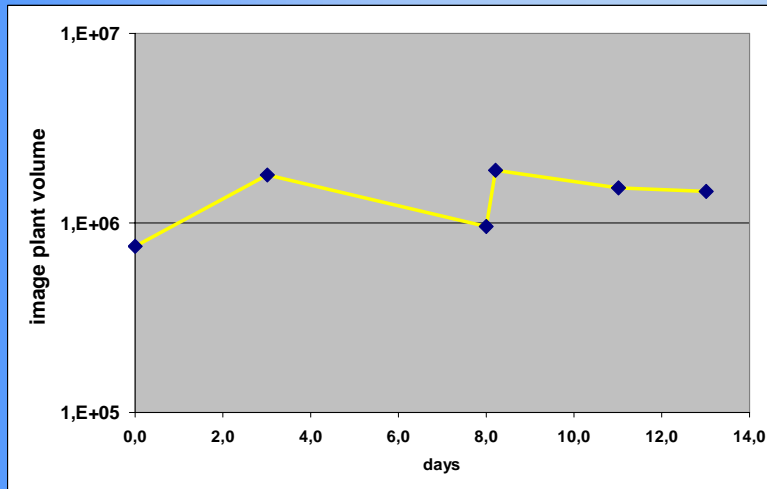
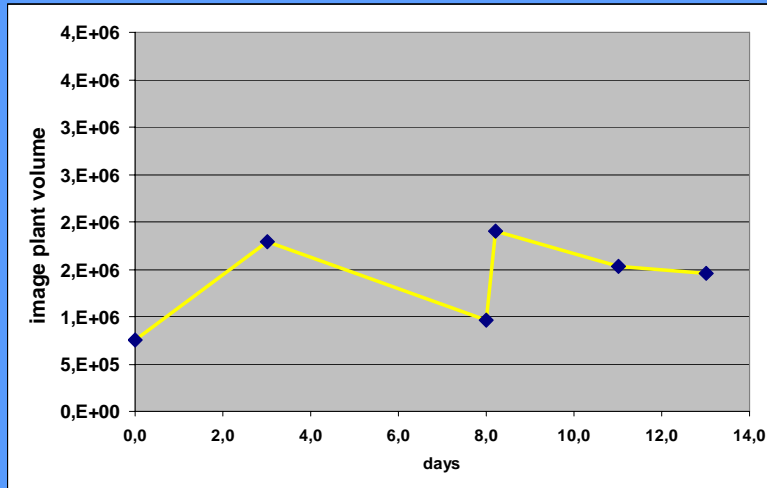


- side by side
- image plant volume: $(\text{sqr}(\text{side1} * \text{side2} * \text{top}))$
- ratio between different views e.g. top/side1



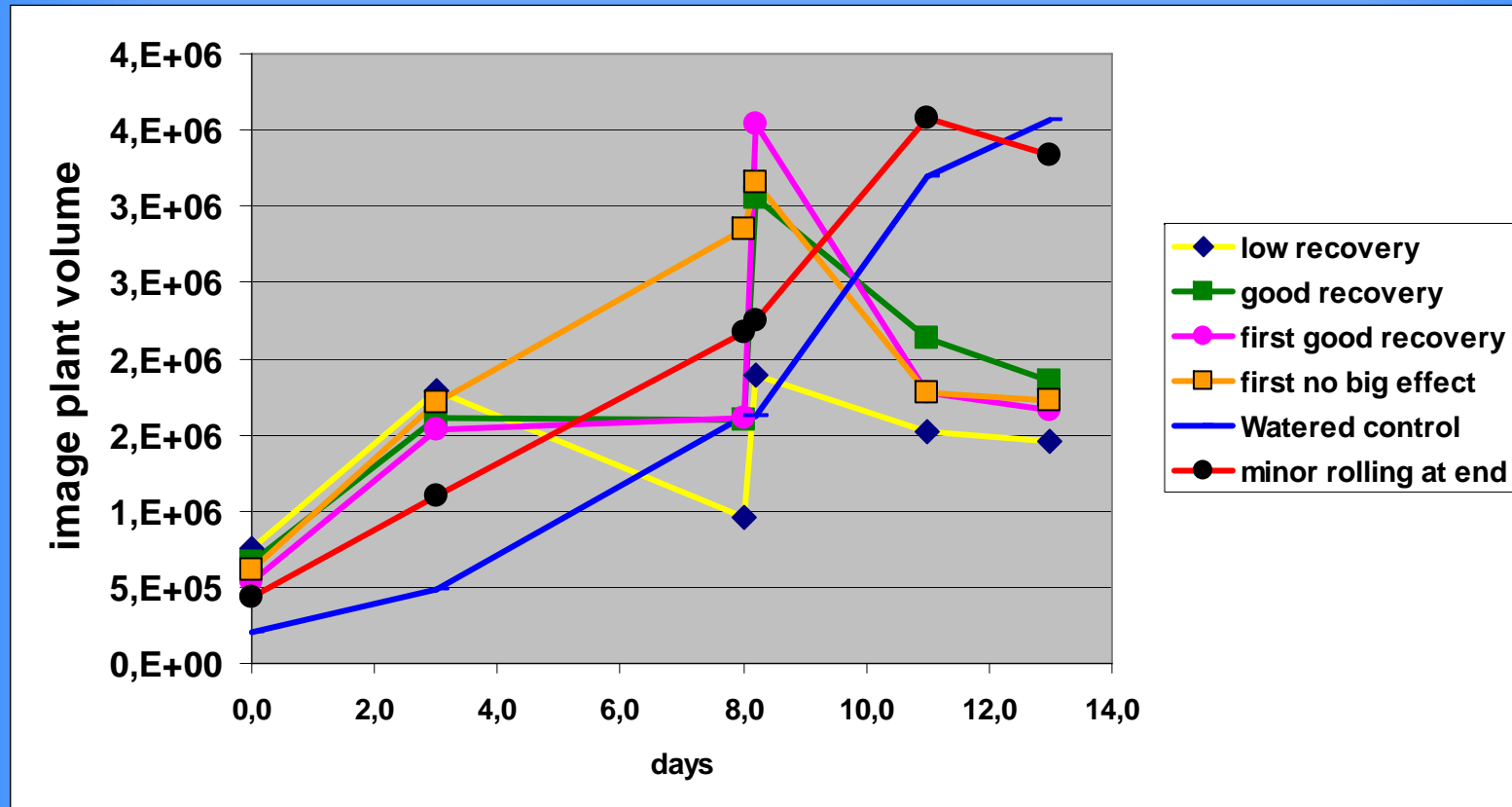
Near to exponential growth of a well-watered control plant





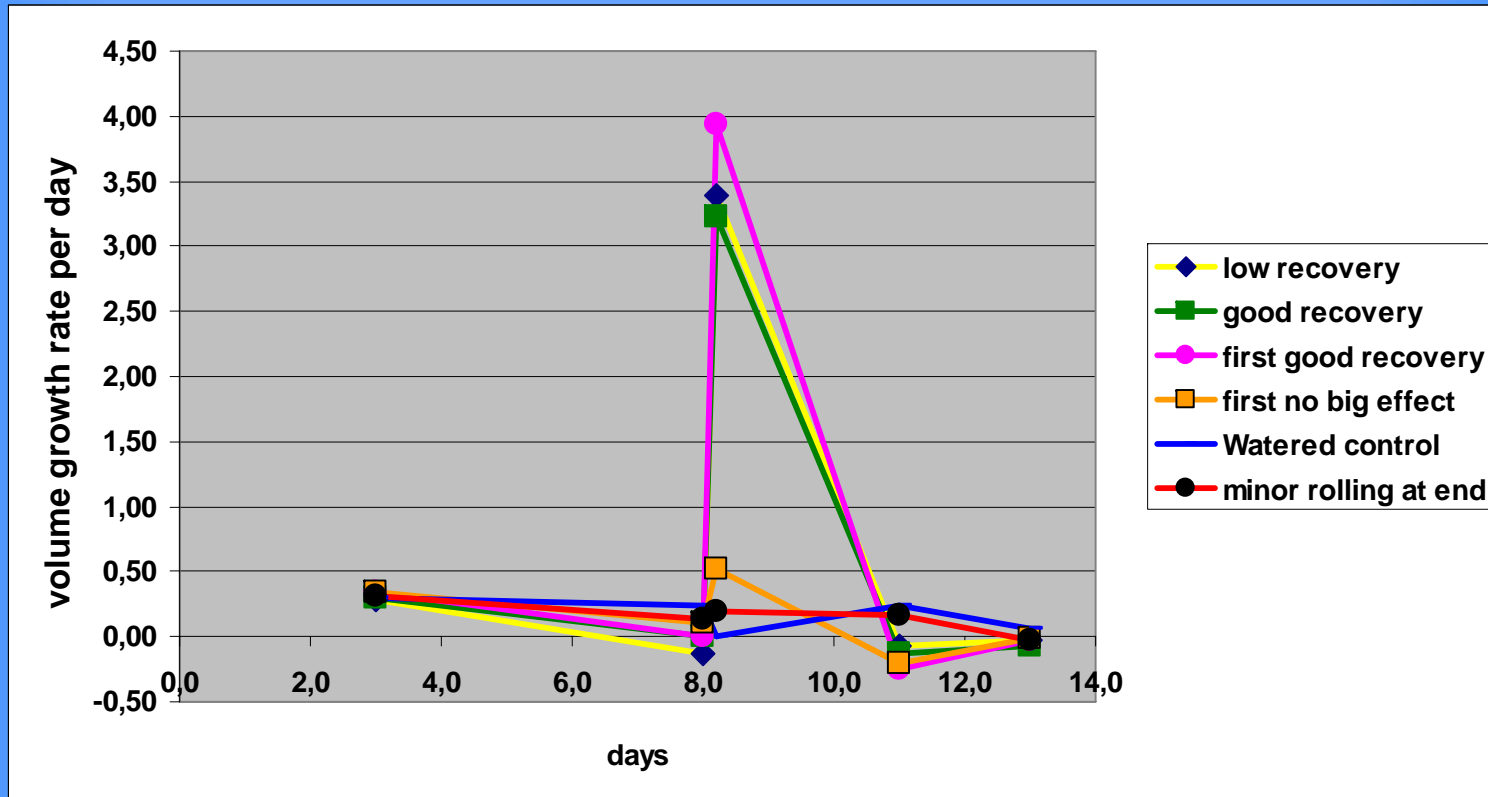
Heavily stressed plant with bad recovery at day 8 and 13

Imaged Plant volume



Different reactions of different plants to periodic stress

Plant volume growth rate

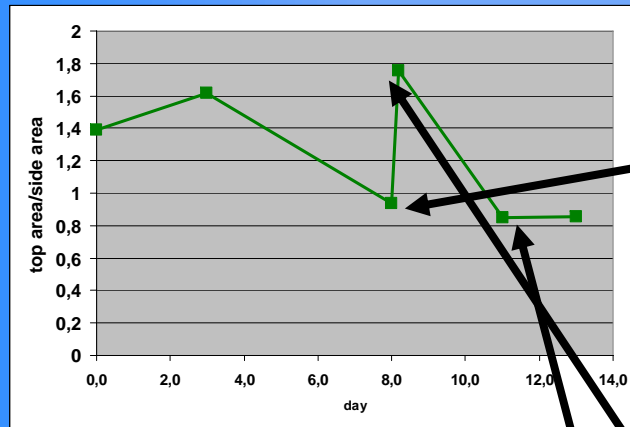


Growth rate of plant volume emphasises absolute recovery effects

- The less changes in growth rate during stress the better.
- The higher average growth rate the better

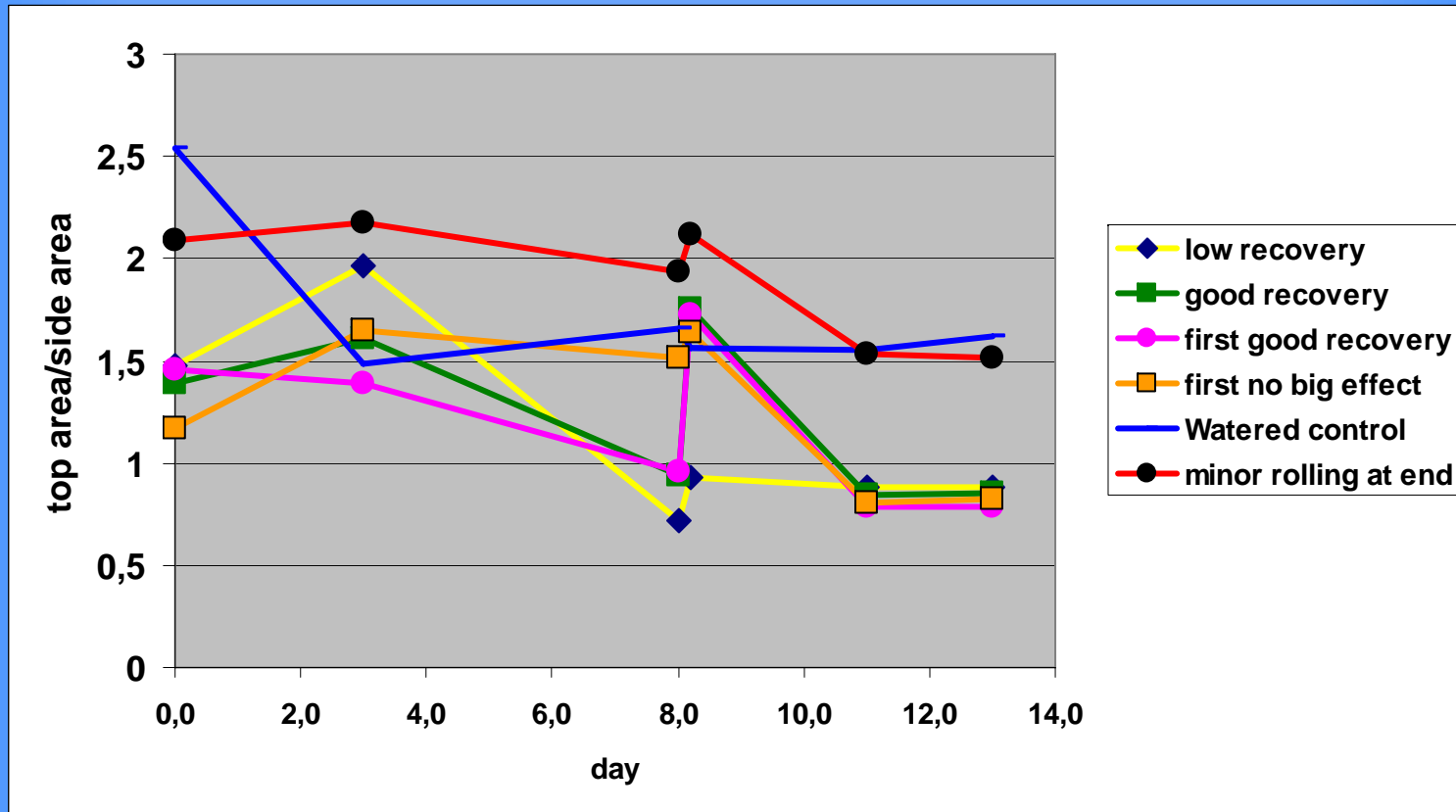
Direct quantification of leaf rolling

Ratio TopArea/Side1Area



While Side1Area is used for normalisation
TopArea reacts strongly on leaf rolling

Direct quantification of leaf rolling



- Changes in top area/side area provides immediate information on leaf rolling status
- Absolute value may contain important further information for screening

Summary Leaf-rolling

- **Already non-destructive leaf area measurements contain a high amount of important never before accessible information of growth dynamics including sensitive reaction e.g. on stressors like drought.**
- **Plant morphology including leaf angles, leaf length etc. provide additional information where biological information is presumed to be high but needs/allows specific interpretation based on screening aims.**

Arabidopsis growth phenotyping

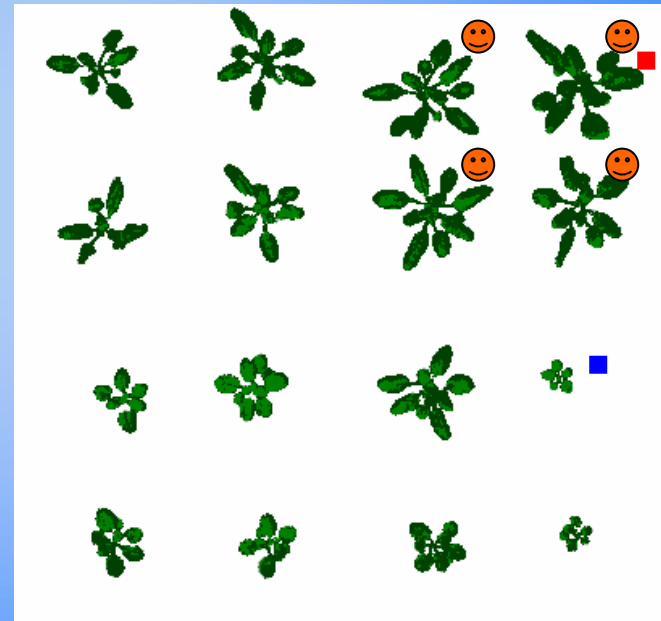
Single point assessment

Single point assessment allows e. g. estimation of plant size and colour at a specific time.



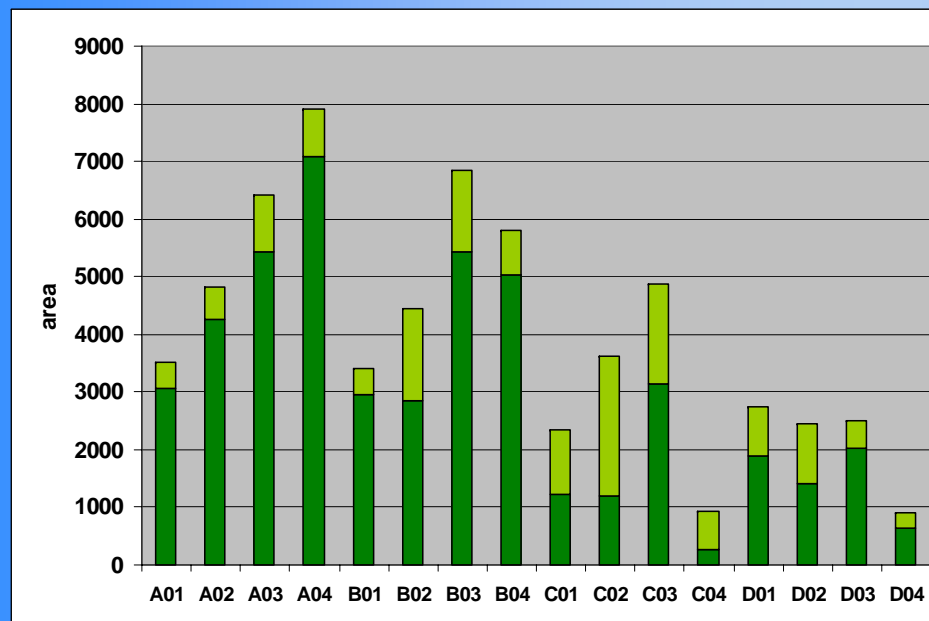
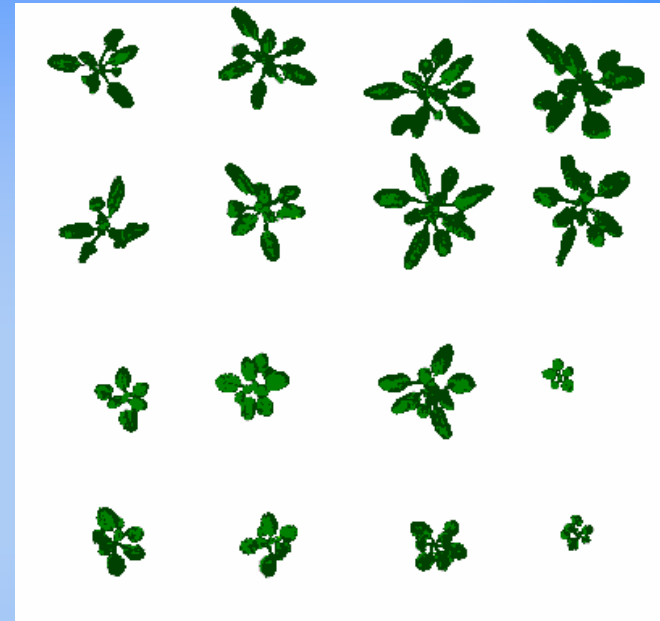
Visual assessment allows e.g. finding the **biggest** and **smallest** or a rough size classification.

Problems with reproducibility remain.



Single point assessment 2

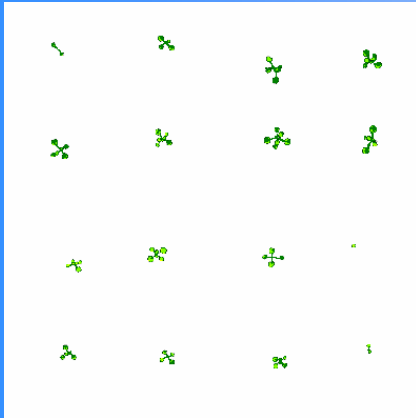
Image processing provides reliable quantitative data allowing e. g. any kind of statistics and a calculation of mean values and significances.



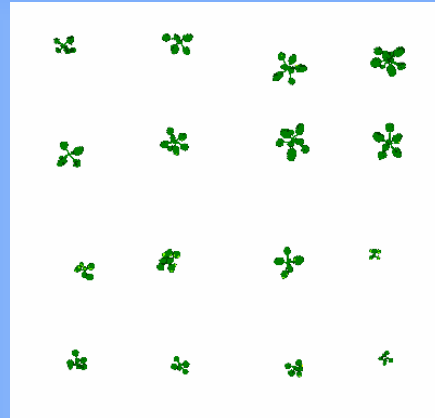
But information describing growth remains limited.

Imaging in time series

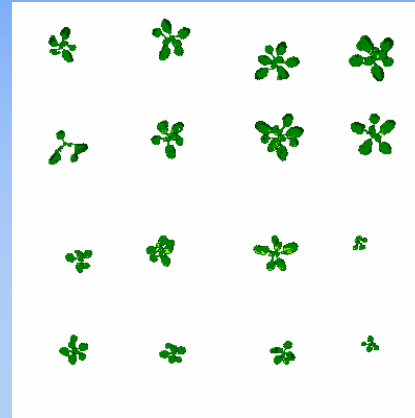
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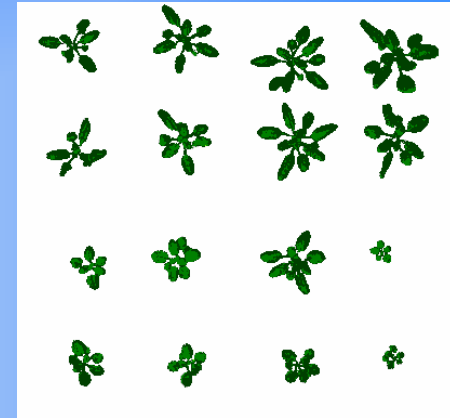
t = 4d



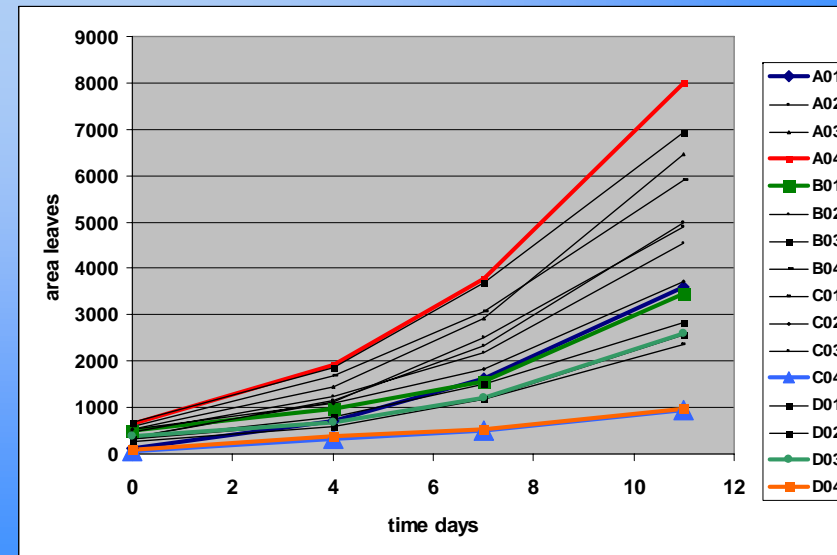
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t = 11d

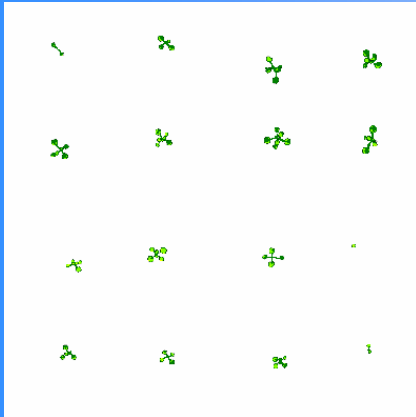


LemnaTecs image processing allows highly automated imaging in time series. But area based growth curves only show a minor part of the information available.

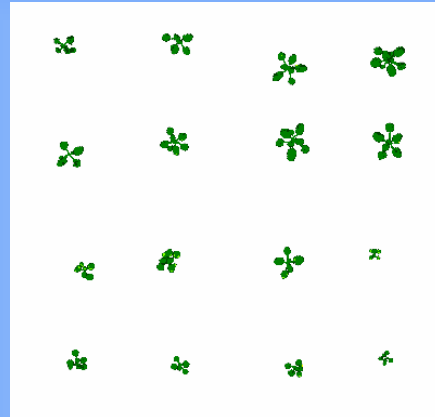


Imaging in time series

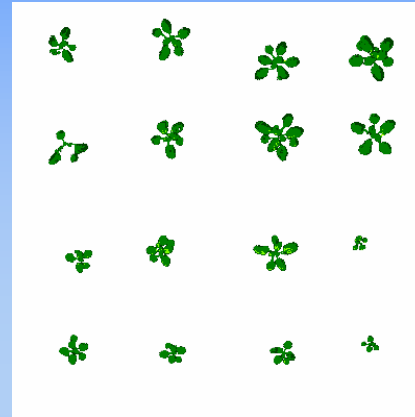
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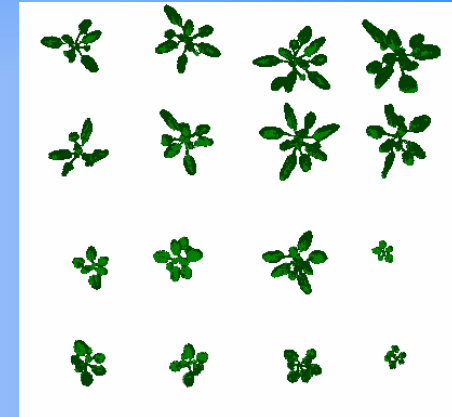
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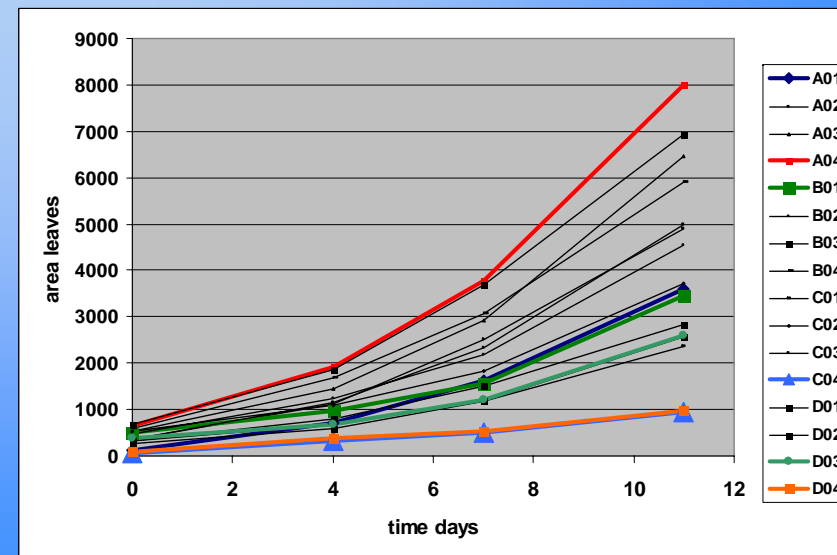
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t = 11d

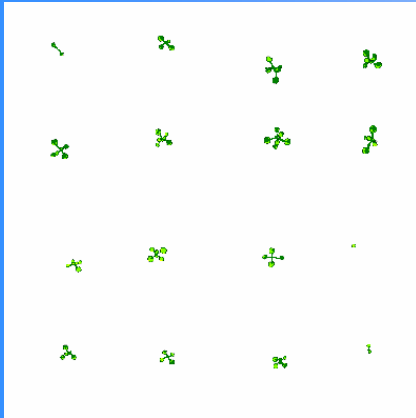


LemnaTecs image processing allows highly automated imaging in time series. But area based growth curves only shows a minor part of the information available. **How homogeneous was growth?**

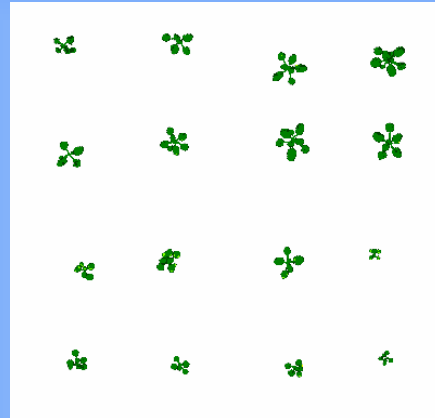


Imaging in time series

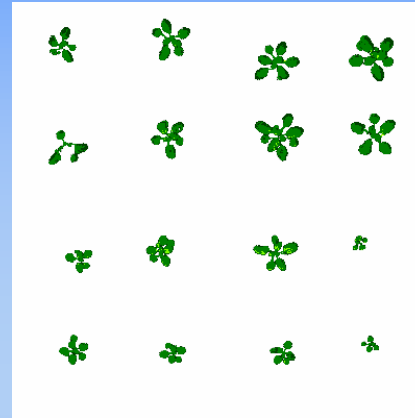
t = 0d



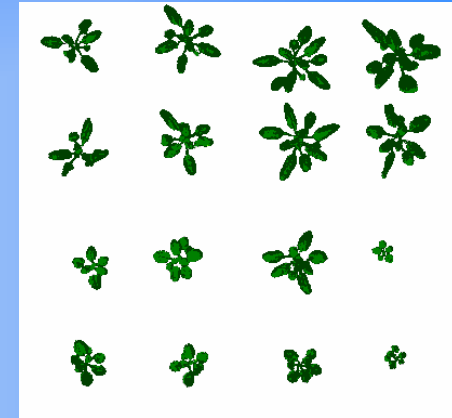
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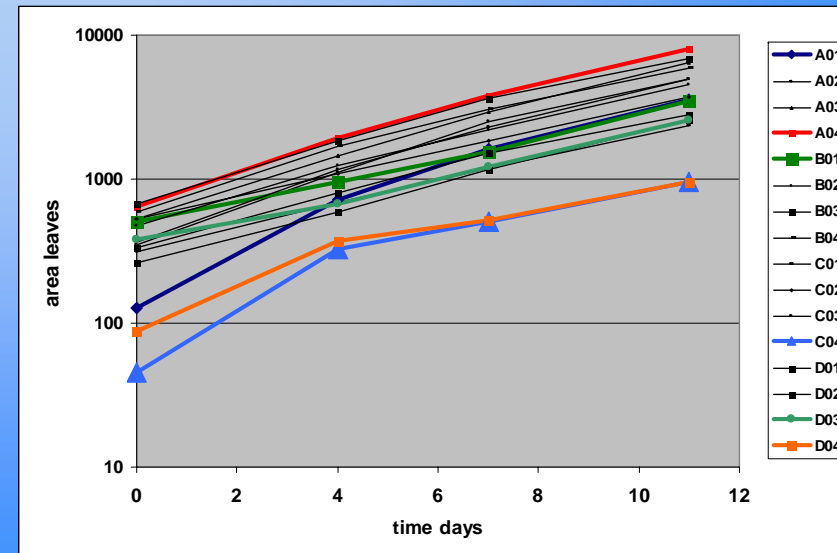
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t = 11d

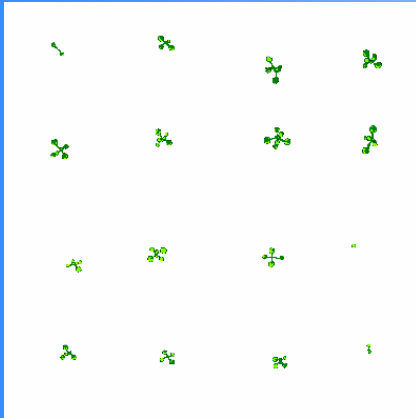


The slope of the curves displaying growth rates show that most plants grew at a relatively constant rate but just started at different sizes e. g. due to delay in germination.

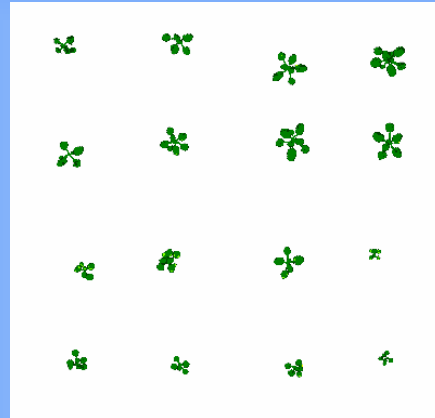


Imaging in time series

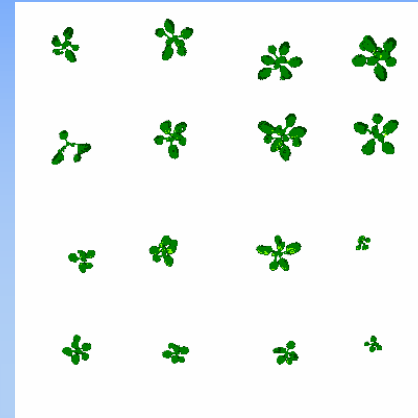
t = 0d



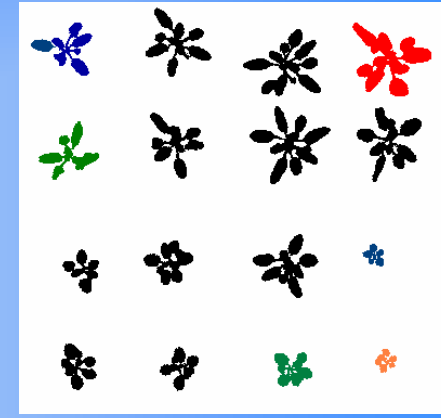
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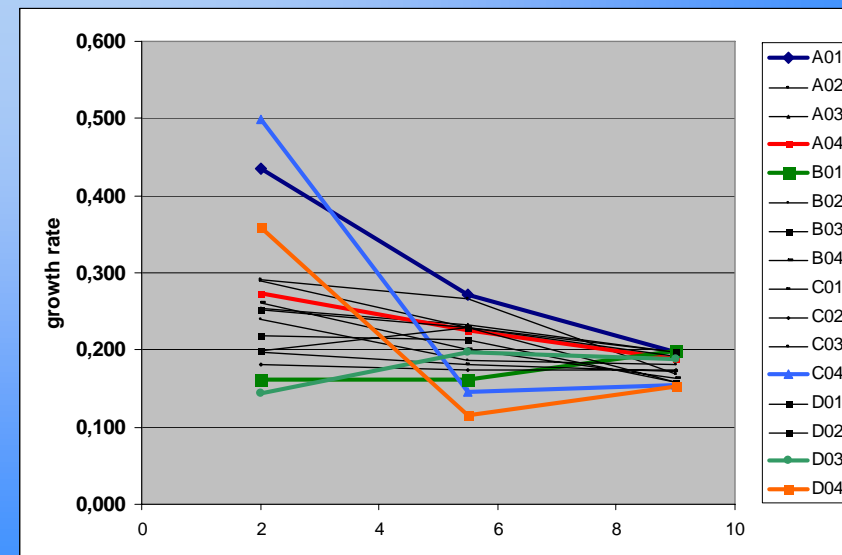
t = 7d



t = 11d

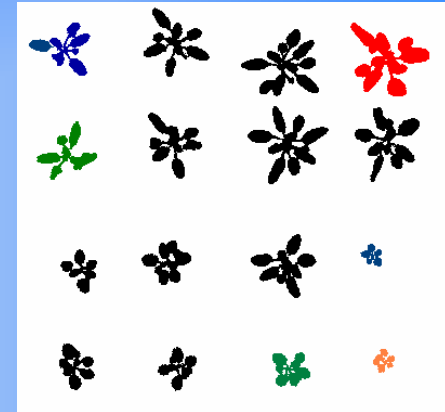


The growth rate graph shows more clearly that the **biggest** plants are not always the **fastest growing** during observation time. Some plants have **interesting growth patterns** and **slow growers** need not be the **smallest ones**.



Conclusions – Growth dynamics

- Comprehensive data on growth dynamics are now readily available for research and screening by non-invasive measurement with the LemnaTec Scanalyzer units.
- Growth rates allow to eliminate assessment artifacts due to inevitable differences in plant size at the start of the test.
- Using quantitative, reliable data and growth rate based calculations reduces „biological data variability“ and raises data quality for any kind of later assessment or statistics
- Data provided can offer new insights in growth dynamics especially under stress and recovery test-designs.





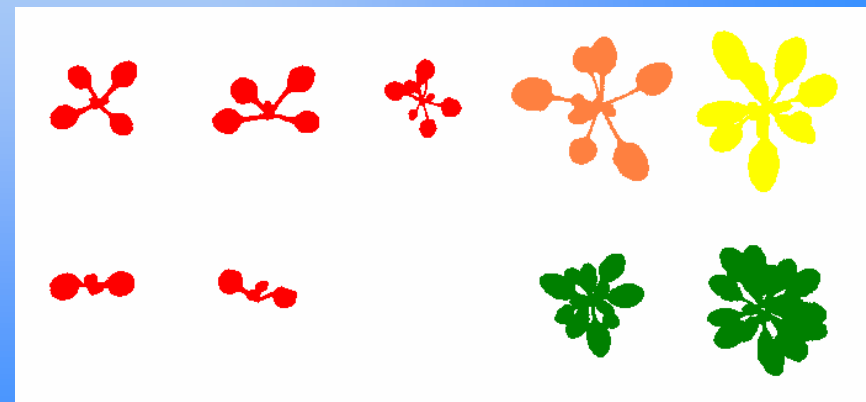
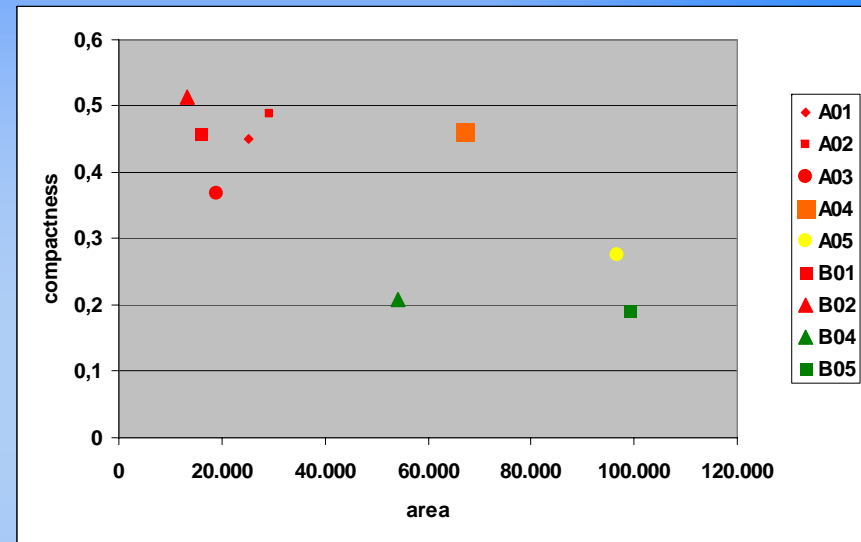
Morphological assessment

- LemnaTecs new Morphological Assessment Tool (MAT) provides an almost unlimited set of morphological parameters to be correlated with biological effects
- The high number of different parameters quantified automatically allows a comprehensive high content screening and statistics on morphological patterns and changes of patterns in time.
- The following examples are parameterised for arabidopsis but similar approaches are available for a wide range of biological applications.

Morphological assessment

- compactness -

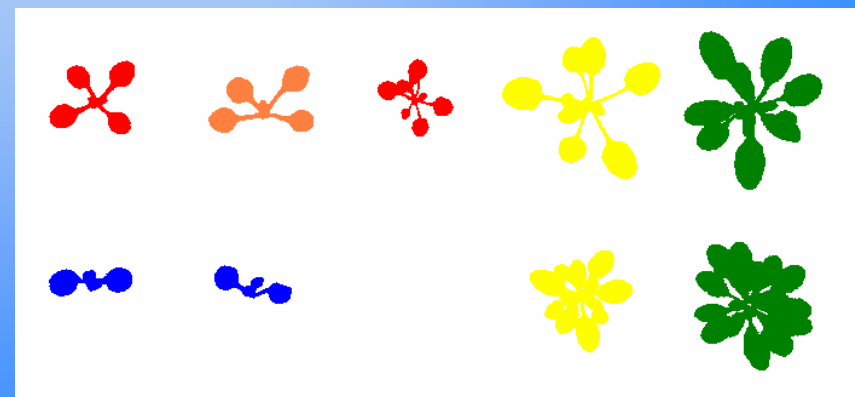
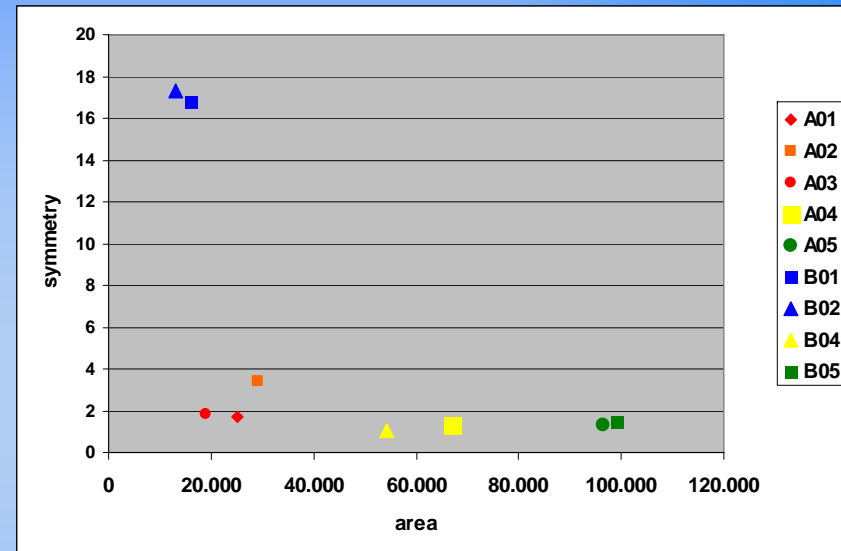
- compactness is calculated based on the size independent rotational momentum of the plant
- it describes if the leaves are nearer around the centroid or farther outside e. g. showing longer stipes



Morphological assessment

- rotational symmetry -

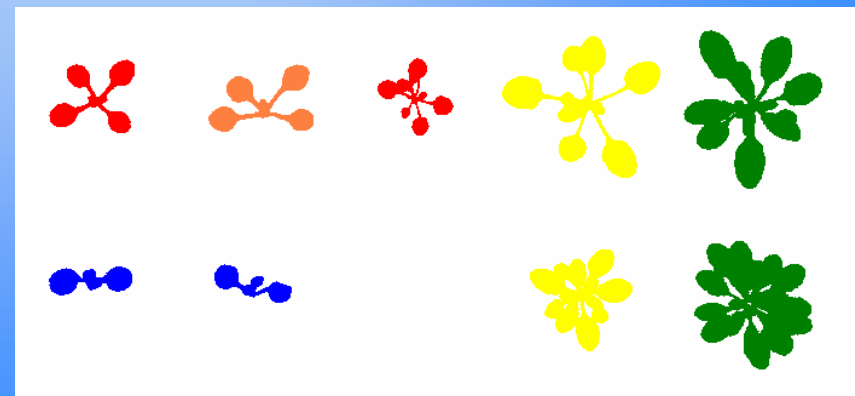
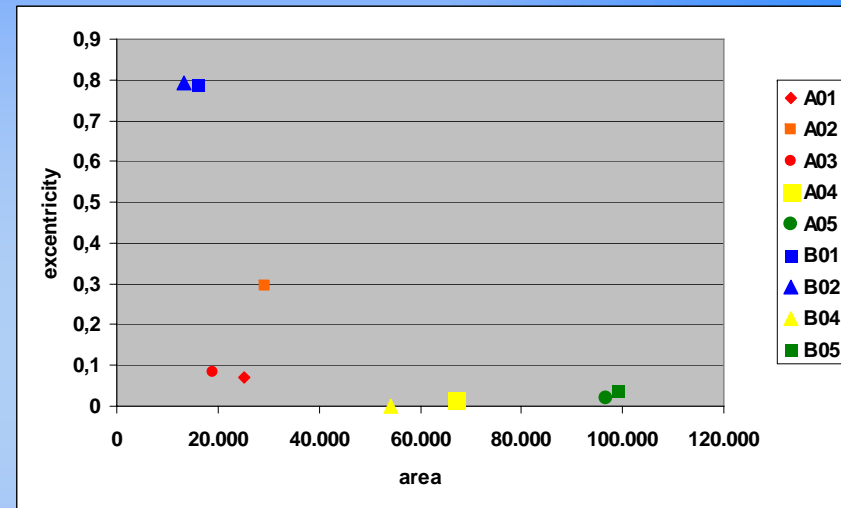
- rotational symmetry is calculated based on the size independent 2nd moment principal axis ratio
- it describes in how far the leaves altogether show a symmetric plant
- this may lead to completely other grouping than before-mentioned compactness



Morphological assessment

- excentricity -

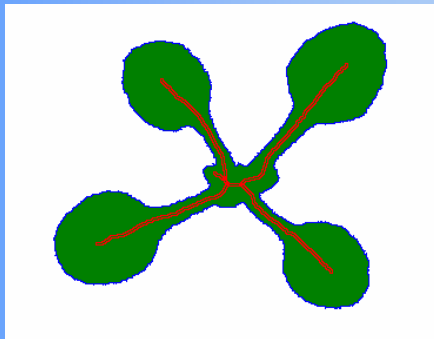
- while calculated with a different algorithm excentricity provides here quite similar grouping results as rotational symmetry
- nevertheless plant A02 shows more significant distance to A01 and A03 than with rotational symmetry



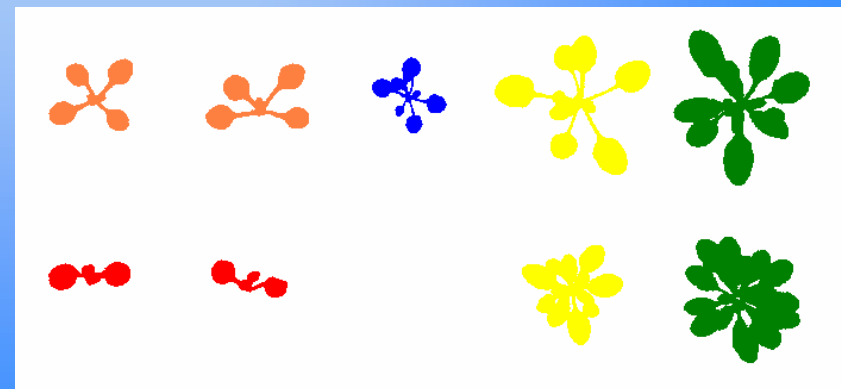
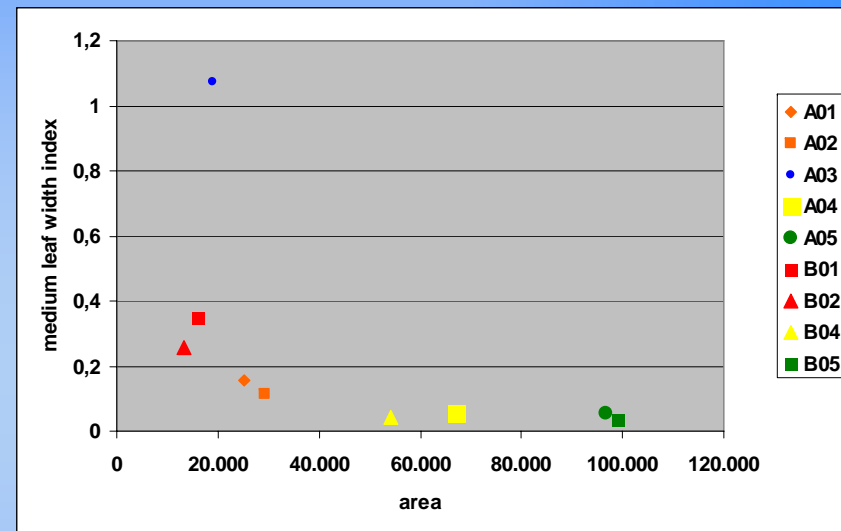
Morphological assessment

- medium leaf width index -

- the medium leaf width index is calculated from the square length of the plant skeleton divided by the leaf area



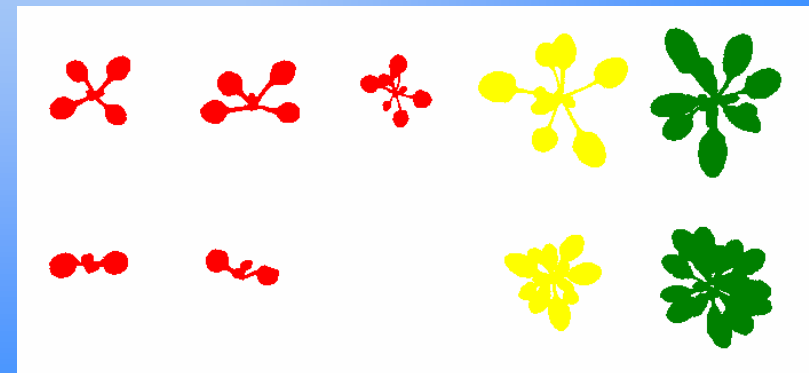
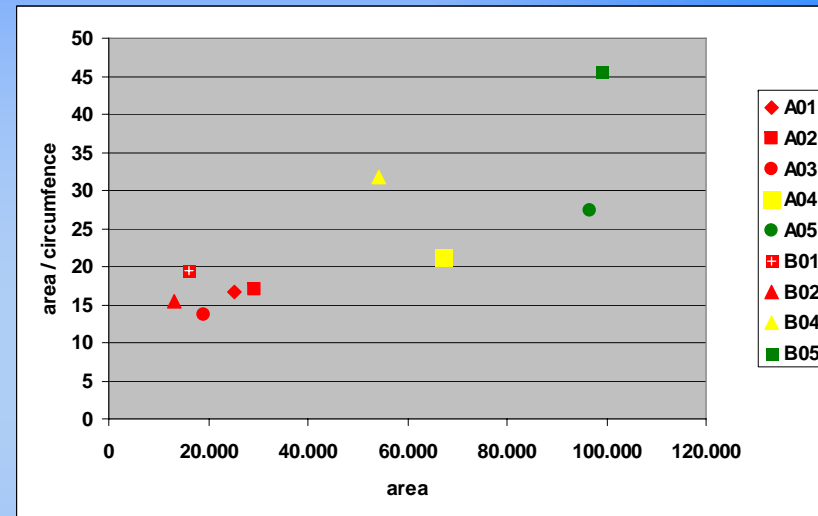
- it describes size independent differences in „leaf width“ integrating stipes, leaves and overlapping effects.



Morphological assessment

- area/circumfence -

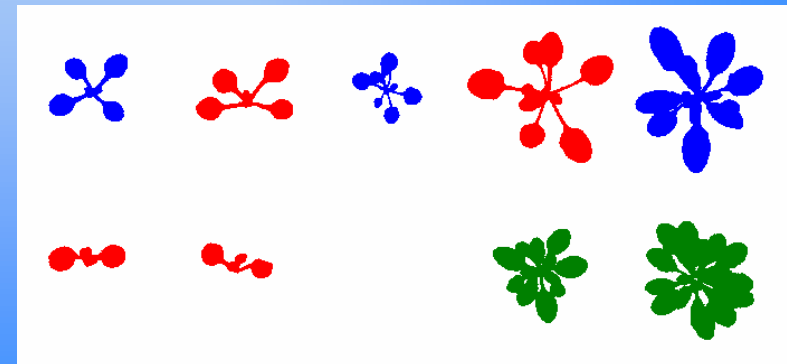
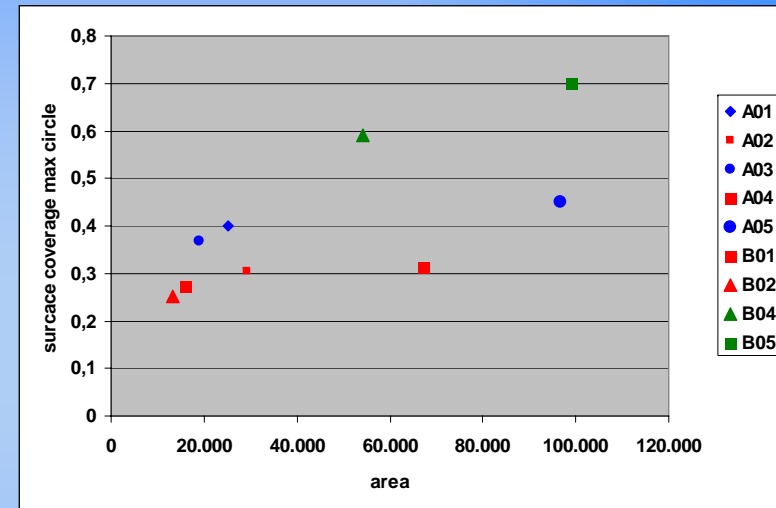
- while having some size dependency left the ratio of leaf area divided by plant circumference may allow additional classification of morphological traits.
- as with many parameters each one provides new aspects for grouping plants which may be especially suitable for statistical QTL analysis



Morphological assessment

- surface coverage -

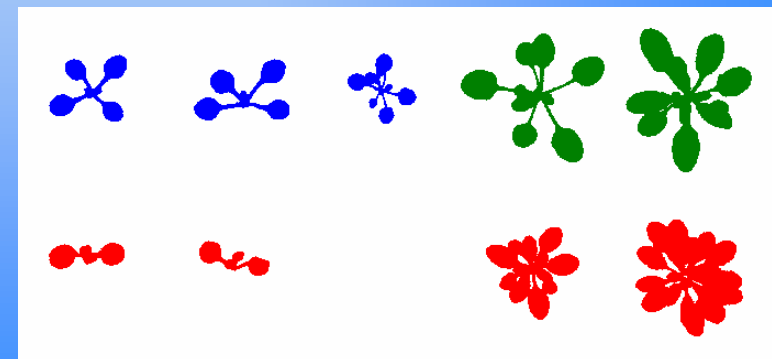
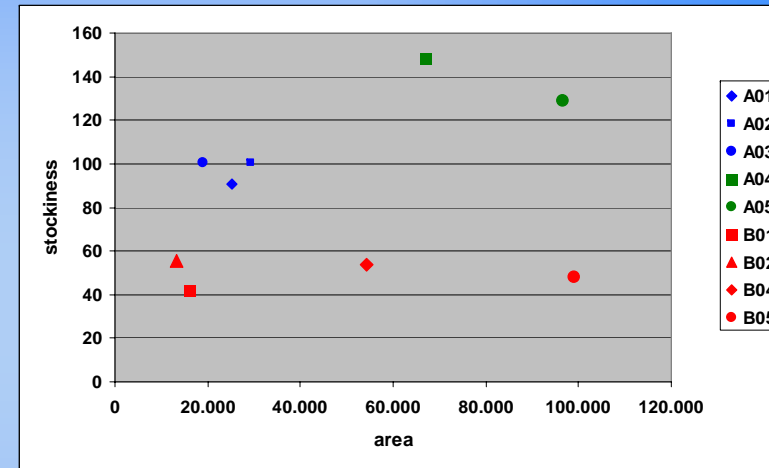
- surface coverage compares the measured plant area to the area of a circle covering the whole plant
- this parameter is intended to provide a calculation how dense the plant covers the soil in its immediate growth area



Morphological assessment

- stockiness -

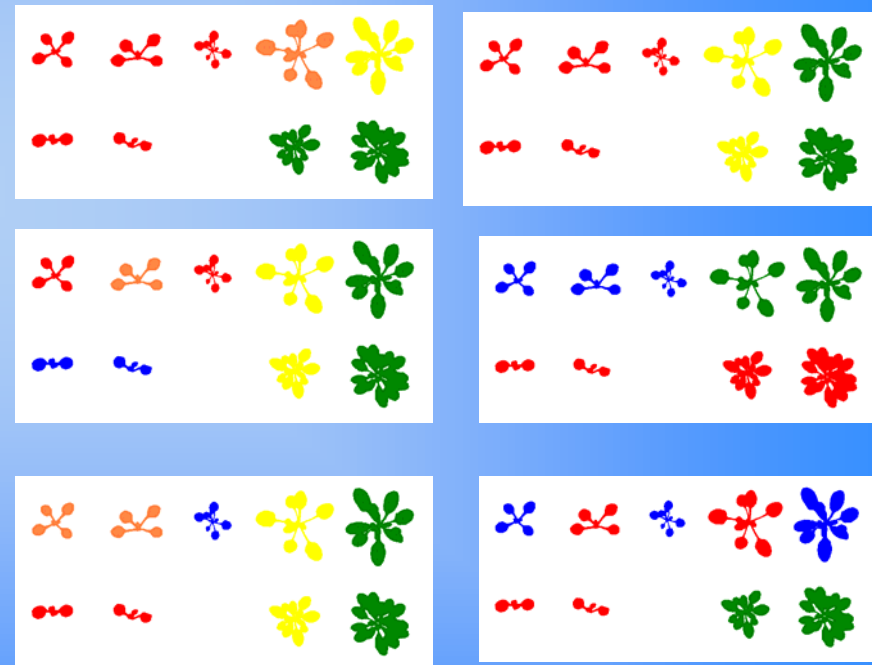
- stockiness is mathematically the description of roundness.
- applied to Arabidopsis images it separates plants with invisible or relatively short broad stipes from plants with long small stipes



Morphological assessment

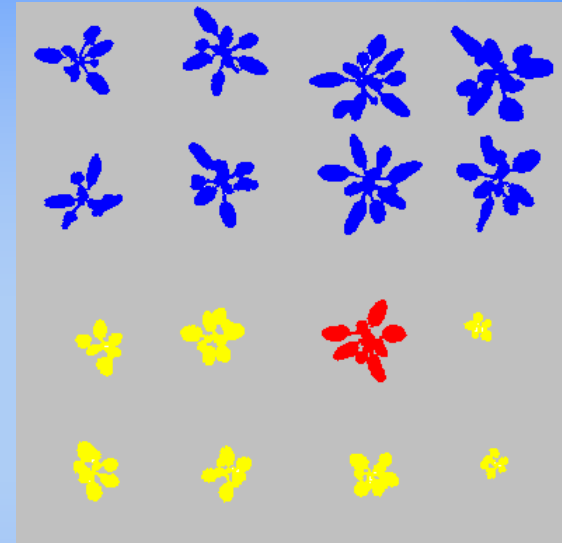
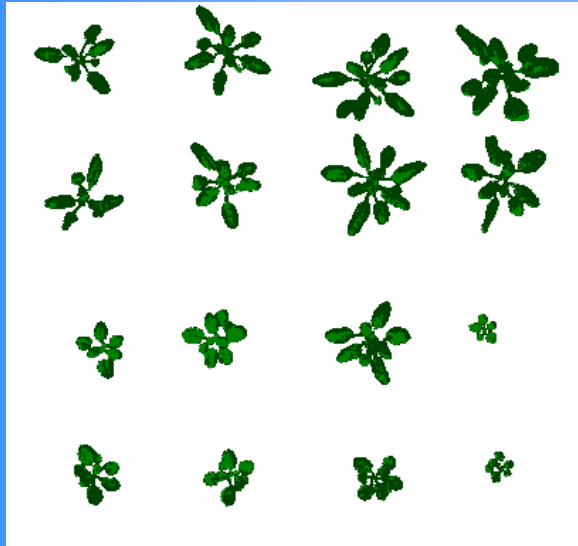
- conclusions -

- With MAT LemnaTec can provide very powerful tool to assess a wide range of morphological parameters for plants
- each quantitative parameter refers to a different morphological trait and may be used in high content screening e. g. for QTL analysis
- different parameters allow reproducible different groupings of the plants
- the examples shown here are just a selection parameters to be individually defined based on the traits to be separated

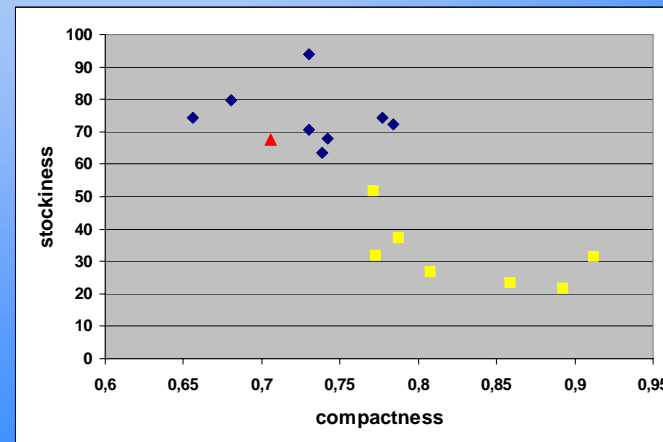


Morphological assessment

- applications -



To show that different parameters can be used for the identification of specific traits, the example here shows classification based on compactness and stockiness





It works!