



LemnaTec High Content Screening

CORN MORPHOLOGY – LEAF ROLLING

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Morphology of Corn Plants

The following application of the LemnaTec Scanalyzer 3-D for the assessment of leaf rolling shows which information can be derived from 3-D-imaging. The data collected with the LemnaTec 3-D-system and the Bonit HTS software only needs to be interpreted consistently from a biological point of view.

While some parameters, like leaf angles, leaf length and leaf colour, are derived from one image only, a wide range of parameters requires calculation of multiple images.

In general, three images are made each time a plant is imaged in the Scanalyzer 3-D. The turning of the plants to provide two-side view images in a 90° angle is performed automatically by a turning device within the imaging unit.



Image 1: Side view 1

Side view 2

Top view

From each single view one leaf area value is derived. These values may then be used in different ways:

1. as single values, describing, for example, the leaf area projection from top as a parameter for photosynthetic effective area need and plant extension;
2. as the image-based plant volume (square root (side 1*side 2*top)), reflecting a parameter surrogate for biomass estimations
3. as ratio between top view and side view 1, providing information on leaf rolling dynamics.

This very basic example shows that using one measuring parameter per image alone can already provide a wide range of different information, depending on how it is biologically interpretable. This is particularly true if the change of parameters is interpreted as development over time.

Image-based Plant Volume – Development over Time

For a healthy, well-watered plant image-based plant volume generally develops along similar lines as in the following figure. In the first weeks of development, plant volume is increasing faster from day to day.

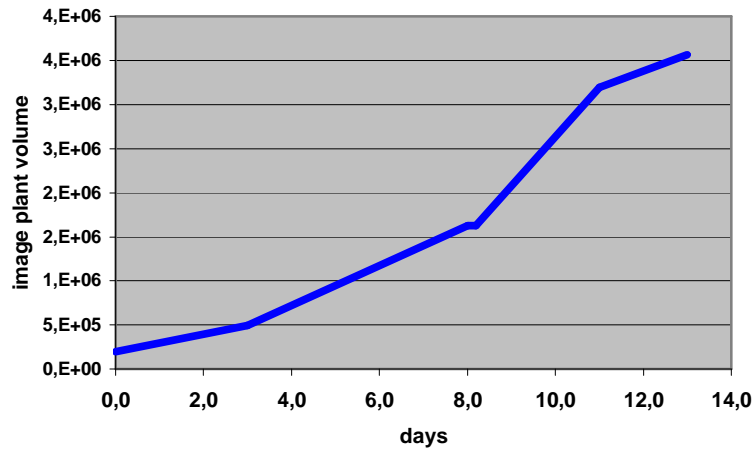


Fig. 1: Growth curve of a well-watered plant

In this case an external stressor slowed down growth during the last 2 days, bad weather (no sun) and cooler temperatures respectively.

Figure 2 shows that the growth pattern up to day 11 is very near to exponential, which is generally true for a wide range of younger plants.

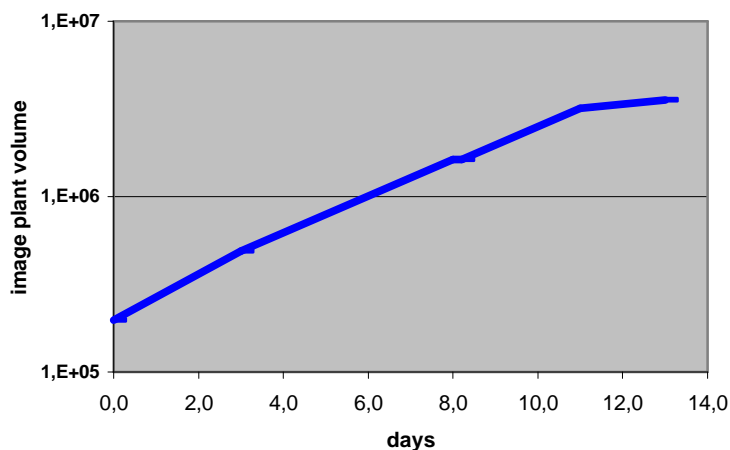


Fig. 2: Growth curve identical to fig. 1, but y-axis is logarithmic to display almost constant (linear line) growth rate (slope) up to day 11

For a plant drying out between day 6 and 8 and imaged twice at day 8 before and after watering to see if there is recovery, the correspondent figures are shown below. After watering at day 8 the plant dries out again till day 13.

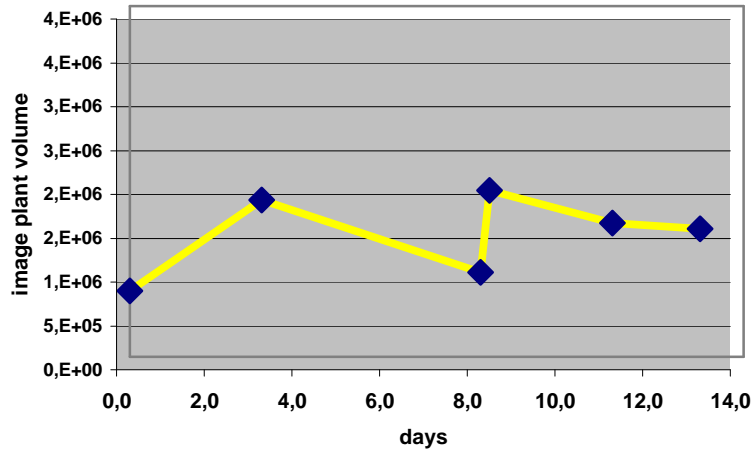


Fig. 3: Growth curve of a heavily drought-stressed plant, not watered between day 6 and 8, and after watering at day 8 (with double imaging) drying slowly out again

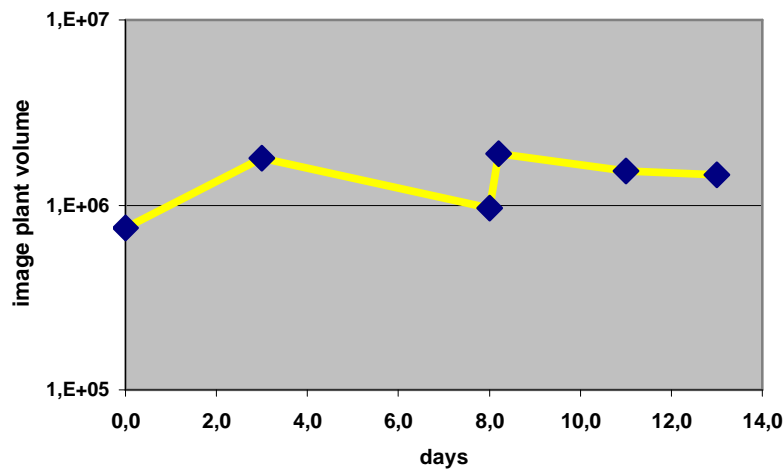


Fig. 4: Growth curve identical to fig. 3, but y-axis is logarithmic to display growth rate (slope) of image-based plant volume

The results show some recovery of plant volume after watering, but no further growth of the plant volume when drying out again.

Results for a Set of Different Plants

The following figures show a set of different development patterns, based on changes of the plant volume over time.

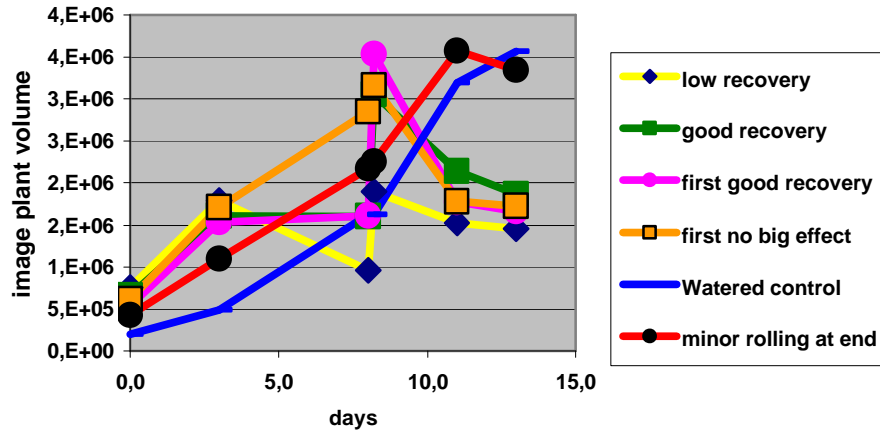


Fig. 5: Plant volume growth curve of a set of different plants with different treatments or different reaction patterns to drought

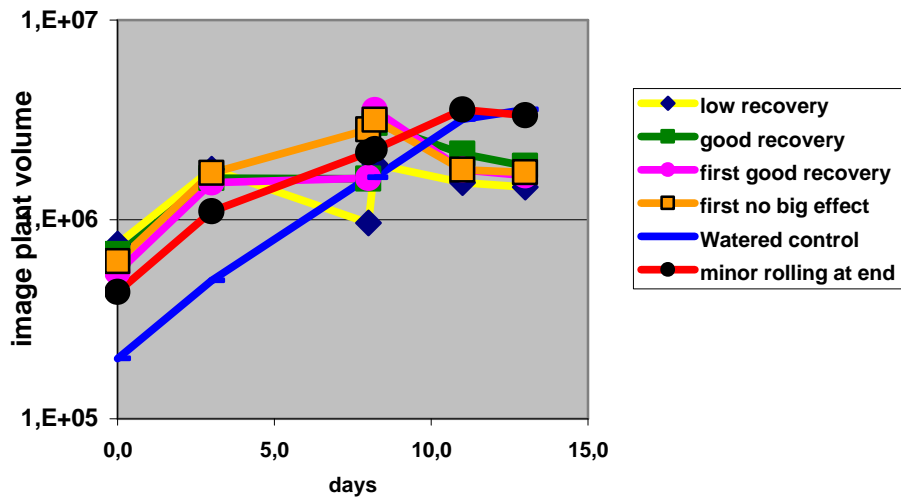


Fig. 6: Logarithmic plant volume growth curve of a set of different plants with different treatments or different reaction patterns to drought

Fig. 6 already shows quite clearly that the plant named "first no big effect" displays some stress symptoms well before day 8, while this is not so obvious from the non-logarithmic fig. 5.

The following figure shows the growth rate values emphasising changes in growth rate.

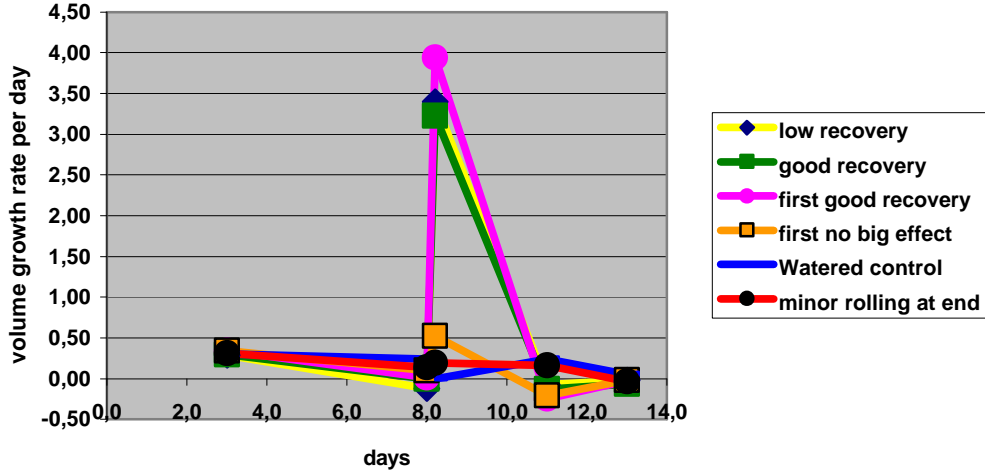


Fig. 7: Growth rates based on plant volume of a set of different plants with different treatments or different reaction patterns to drought

Control plants or such that show no major effect of drought for other reasons have an almost constant growth rate, while plants showing any recovery show a big peak at day 8 before and after watering. As recovery is a relative value, there is no significant difference between the “low recovery” plant and the “good recovery” plant as both recover, but on quite different levels. The “good recovery” plant never reduced its volume to such a great extent and recovers to a much higher absolute level than the “low recovery” plant (see fig. 5).

The graphs show clearly how much information is inherent in the parameters once they are finally displayed. And the data calculated from total leaf areas of different sides represent just a small part of all the parameters that the LemnaTec Scanalyzer 3-D can provide.

While all these parameters have a clear biological interpretation, none of them represents the very basic visual classification criterion leaf rolling as a stress symptom at the first glance.

Thus further analysis of the calculated images is always necessary.

Taking a look at some plant images, it becomes clear that the side leaf area is not changing dramatically when the leaves start rolling.

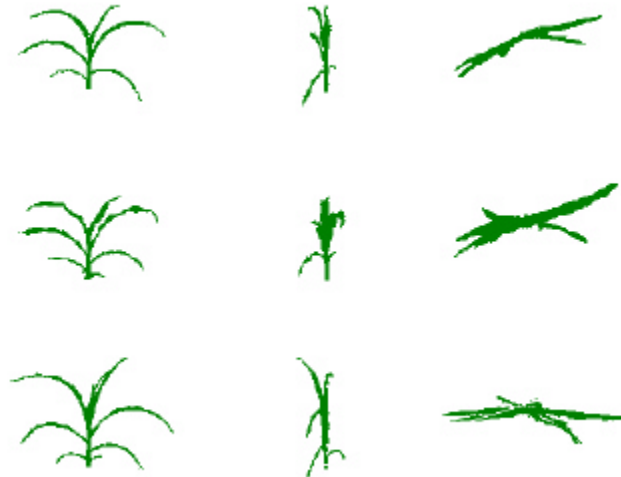


Image 2: The same plant is imaged after leaf rolling (top row), after recovery (2nd row) and after another leaf rolling (3rd row)

By contrast, the top leaf area changes dramatically under leaf rolling conditions. Based on this observation, the ratio between both areas was calculated. The side leaf area was thus used as a normalising parameter, because plants may have different absolute sizes when rolling their leaves. Leaf area index values lower than 1 are indicators for strong rolling.

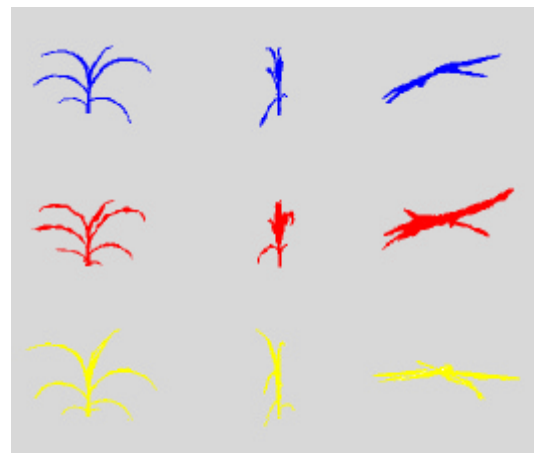
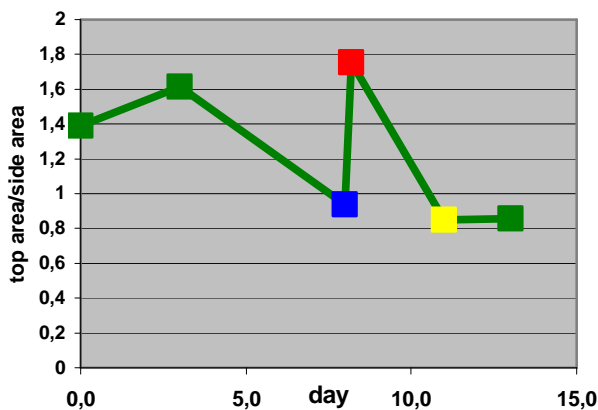


Figure 8: Relation between leaf rolling index and visual impression. Leaves were rolled for the blue and the yellow plant

When calculating the leaf rolling index of the above shown plants for all measured times, data can be visualised as in figure 9.

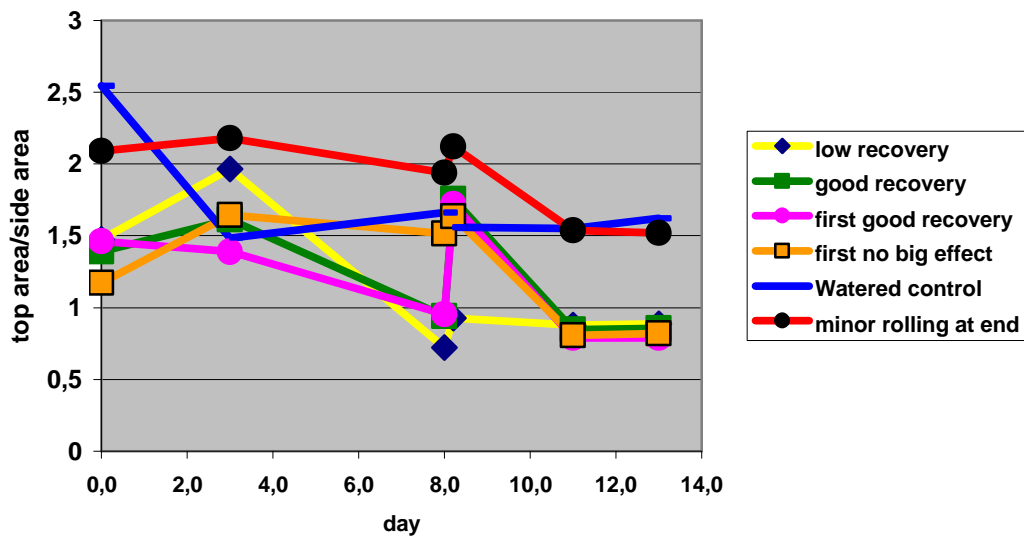


Figure 9: Leaf rolling index for a set of plants reacting differently to drought or various other treatments

By just looking at the graphs it becomes quite obvious why the different plants were named accordingly. While the “watered control” plant has an almost constant index from day 2 on, the “low recovery” plant does not roll its leaves significantly, whereas plants showing good recovery reraise the index to a level over 1.5 after watering. The plant “minor rolling at end” shows as a rule a significantly different pattern, growing with an index of about 2, which is only reduced to the index value the well-watered plant showed during the entire test.

Conclusion

Biological interpretation of only one value per image (leaf area), provided by the LemnaTec Scanalyzer 3-D, already allows for a comprehensive interpretation of drought reaction patterns. This example demonstrates the immense informational treasure the great number of different parameters hold, if interpreted in the appropriate biological context. All these useful parameters can be provided by the Scanalyzer 3-D.

For further information, please do not hesitate to contact

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