Hairy roots of beetroot (*Beta vulgaris*) as a model system for a structured growth model

Analysis of root growth morphology and architecture using automatic picture recognition for parameter acquisition

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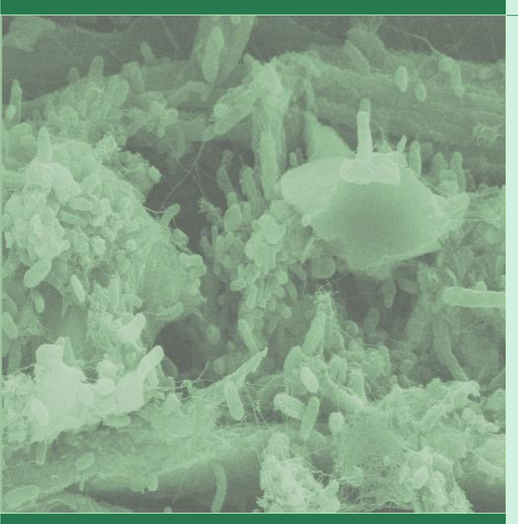

Introduction and Aims:

Fig. 1 *Beetroot* plant

Fig. 2 Hairy roots of *Beetroot*

Plant cells bare a wide range of nutritional-physiological and pharmaceutical relevant secondary metabolites. While usual suspended *in vitro* cultures need a constant level of different hormone concentrations, *Agrobacterium rhizogenes* induced Hairy roots can be cultivated in hormone free media. However the cultivation of these tissue cultures in bioreactors is difficult and several challenges exist [1].

In general the growth of these tissue cultures on agar plates, in shaking flasks or bioreactors for industrial use has been heavily investigated experimentally but only limited theoretical descriptions of the growth process exist [3].

In order to model the growth morphology and the distribution of secondary metabolites beetroot (*Beta vulgaris*) was chosen as a model system. It produces the red dye *Betanin* which is used as a food color and is also responsible for the red color of the root network. Therefore it can be used to identify the distribution of *Betanin* optically.

In a first step of the modeling process three parameters of growth were chosen and analyzed using an automatic picture recognition system to monitor spatial and temporal emergence of the root architecture over a cultivation time of up to 12 days.

The main aim is to generate data for calibration of the model system. Simulation results should be compared with experimental data.

Materials and Methods:

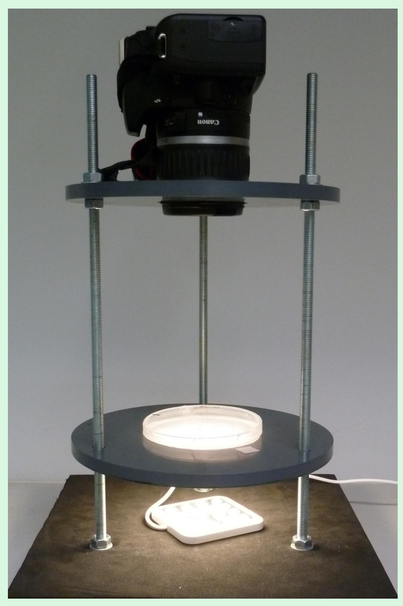

Fig. 3: photographing stand

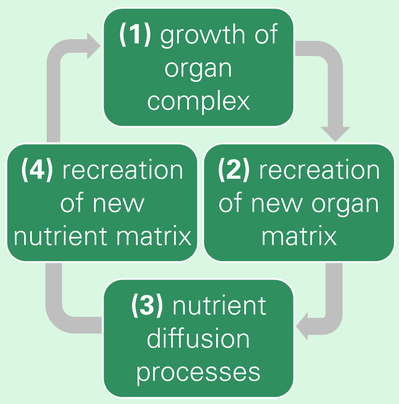

Fig. 4 growth model principle

The proposed structured model uses an individual-based matrix approach for growth simulations on agar plates [2]. It consists of a 2-dimensional **organ matrix** containing a vector with information about each state of a cell (e.g. age, size, metabolite concentrations) and a **nutrient matrix** which represents the composition of the nutrient media (e.g. carbon source, solved oxygen etc.) (see Fig. 4).

In dense root networks growth can be determined spatially at three different parts of the organ complex:

- tip movement / elongation
- branching
- overall biomass growth (secondary thickening)

For **model parameterization** all three growth processes have to investigated experimentally .

Every 12 hours a picture of the respective root network on an agar plate has been taken using a special photographing stand (see Fig. 3) with subjacent LED lighting for maximal brightness and contrast.

After uploading the images to servers running a **picture recognition software**, mean tip movement was determined and branching points as well as mean overall biomass accumulation get identified.

All three growth processes have statistical values and variances and are therefore subject to investigation on a broad scale. Identified segments are numbered and their characteristic parameters are summed up in a machine-readable format (MS Excel file) and as graphics (see Fig. 5 - 7).

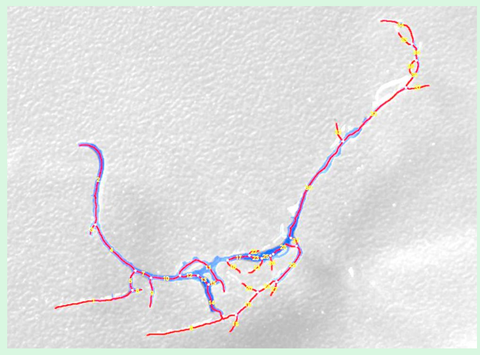

Fig. 5 segment recognition

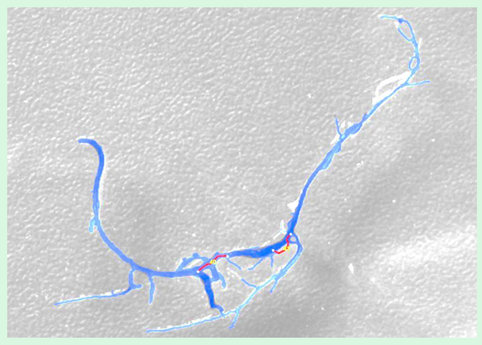

Fig. 6 overlapping segments

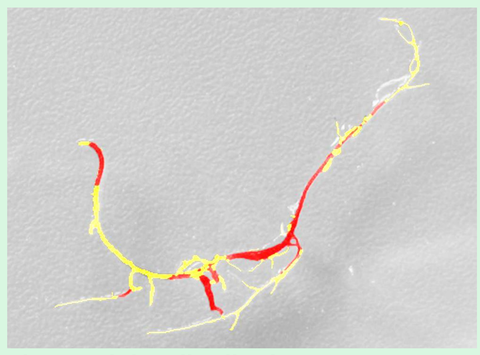

Fig. 7 red dye distribution

Results and future prospects:

A model grid for the simulation of Hairy root growth was established and experiments to gain three relevant growth parameters have been conducted using an automatic picture recognition software. Several other characteristical parameters can be calculated with this data and model calibration is currently under development. The number of **total branching points** (see Fig. 8) as well as **total root length** (see Fig. 9) follow an exponential function with a regression >90%. The custom picture recognition solution will be developed further.

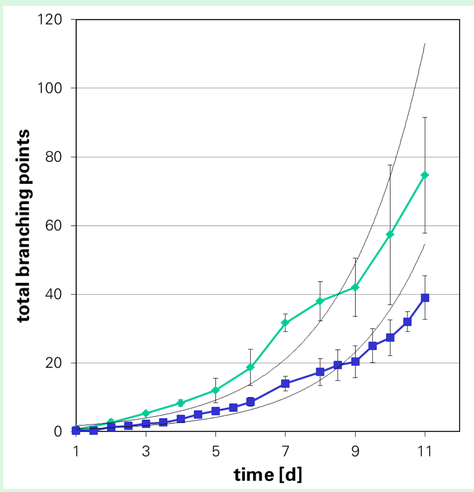

Fig. 8 total branching points over time

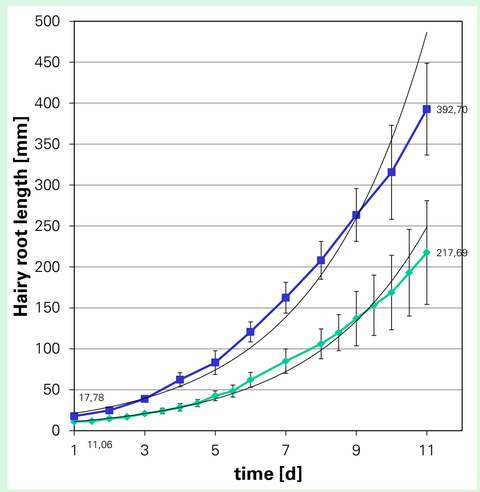

Fig. 9 root length increase over time

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