

## USING MICROFLUIDICS TO STUDY PROGRAMMED CELL DEATH: A NEW APPROACH

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This project focuses on applying microfluidic tissue culture for electrochemical or optical measurements during programmed cell death (PCD) in barley aleurone layer to increase understanding of the underlying mechanisms of PCD in plants.

Microfluidic tissue culture enables *in vitro* experiments to approach *in vivo* conditions. Microfluidics also allow implementation of a wide range of electrochemical or optical assays for online, real-time, parallel analysis of important parameters such as redox activity, O<sub>2</sub> and H<sub>2</sub>O<sub>2</sub> concentration, extracellular pH, cell viability and enzyme activity<sup>1,2</sup>.

Currently, we are optimising an intracellular whole-cell redox activity assay<sup>3</sup> that detects changes in redox activity in barley aleurone layer during PCD. The assay uses a double mediator-system to electrochemically measure redox activity via changes in the NADP:NADPH ratio. Initial experiments show that the redox activity changes depending on phytohormone activation or inactivation of aleurone layer metabolism and subsequent PCD. This is similar to H<sub>2</sub>O<sub>2</sub> concentration changes observed recently by Ishibashi *et al.*<sup>4</sup>.

We have also successfully detected PCD induced by phytohormones in barley aleurone layer using a double-fluorescent probe-system also used by Fath *et al.*<sup>5</sup>.

Future challenges include integrating both these systems into a microfluidic device for plant tissue culture.

### References

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- <sup>3</sup> Heiskanen *et al.*, *Anal Biochem*, 2009, 384, 11-19, 2009
- <sup>4</sup> Ishibashi *et al.*, *Plant Physiol*, 158, 1705-1714, 2012
- <sup>5</sup> Fath *et al.*, *Plant Physiol*, 126, 156-166, 2001