

structures are necessary. To date, investigations in this field have been limited by the asynchronous development of the symbiosis, leading to the presence of different developmental stages in the host root at a given timepoint and a lack of resolution, which has been restricted to whole root material.

To overcome these problems, we perform laser-capture-microdissection (LCM) of specific cell types in mycorrhized roots, using a PALM Microbeam (Zeiss). This technology provides the possibility to excise intact cells from complex tissues, so that homogenous cell pools for downstream analysis can be obtained.

Our goal is to isolate single cells or cell layers housing only one defined developmental stage of the AM fungus and to map the transcriptome of these, using *Medicago* GeneChips. The results should widen the knowledge about already known symbiosis genes, by providing information about the exact timepoint and place of activation, and will probably also lead to the identification of novel genes, which could not be detected in older approaches due to the lack of cellular resolution.

Finally, the function of selected genes in the AM symbiosis will be investigated. To this end, we use a RNAinterference (RNAi)-approach in *Agrobacterium*-derived hairy roots of *Medicago truncatula*. The knock-down level of the candidate genes is assessed by real-time RT-PCR and the performance of AM in transgenic roots is investigated using histological methods and real-time RT-PCR. The ultimate goal is to identify plant genes essential for the different developmental checkpoints of AM symbiosis.

## 6) Transcriptional and functional analysis of plant genes during the symbiotic interaction of the model legume *Medicago truncatula* with arbuscular mycorrhizal fungi

Lisa F. CZAJA<sup>1</sup>, Natalija HOHNJEC<sup>1</sup>, Raffaella BALESTRINI<sup>2</sup>, Paola BONFANTE<sup>2</sup>, Vivienne GIANINAZZI-PEARSON<sup>3</sup>, Million TADEGE<sup>4</sup>, Colby G. STARKER<sup>5</sup>,

Jörg D. BECKER<sup>6</sup>, Helge KÜSTER<sup>1</sup>

<sup>1</sup>Institute for Plant Genetics, Unit IV-Plant Genomics, Leibniz Universität Hannover, Hannover, Germany

<sup>2</sup> Instituto Protezione Piante, sez. di Torino – CNR and Dipartimento Biologia Vegetale – Università di Torino, Torino, Italy

<sup>3</sup> UMR INRA/CNRS/Université de Bourgogne Plante-Microbe-Environnement, Dijon, France

<sup>4</sup> Plant Biology Division, The Samuel Roberts Noble Foundation, Ardmore, USA

<sup>5</sup> Department of Plant Biology, University of Minnesota, Saint Paul, USA

<sup>6</sup> Affymetrix Core Facility, Instituto Gulbenkian de Ciência, Lisbon, Portugal

E-Mail: Lisa.Czaja@genetik.uni-hannover.de

Arbuscular mycorrhiza (AM) is a widespread symbiotic association between plants and fungi. AM fungi supply their symbiotic partner with nutrients and water from the soil, while they receive photosynthates from the plant in return. The successful establishment of AM requires mutual signaling of both symbiotic partners. Compared to the root nodule symbiosis, knowledge of the molecular basis of this signaling pathway is still limited. Thus, we are interested in uncovering plant genes involved in early symbiotic signaling.

To this end, candidate genes have been identified via extensive data mining of 72 *Medicago* GeneChip hybridizations covering (1) comparisons of different wild type plants with mutants impaired in early symbiotic signaling, (2) wild type plants inoculated with different AM fungi, and (3) wild type

plants inoculated with diffusible microbial signals; obtained in the frame of the TRUNCATULIX project. Candidate genes were selected according to their expression characteristics from the GeneChip experiments mentioned above, their expression profiles from the *Medicago truncatula* Gene Expression Atlas (MtGEA) and their annotation. Concerning the last point, we had a special focus on genes annotated as transcription factors and annotated as being involved in calcium signaling, since calcium signaling seems to play an important role in the closely related signaling pathway of the root nodule symbiosis.

To functionally characterize those candidate genes we currently design RNAi-constructs to determine whether the knock down of these genes has an impact on formation of AM symbiosis between *Medicago truncatula* and the arbuscular mycorrhizal fungus *Glomus intraradices*; and study the phenotype of *Tnt1* mutants in the genes concerned. In addition, promoter studies for selected candidate genes are planned to uncover novel expression markers that can be used to study microbial infection and plant responses to signals from AM fungi.

## 7) Ist *Piriformospora indica* eine Alternative zur arbuskulären Mykorrhiza?

Philipp FRANKEN

IGZ, Theodor-Echtermeyer-Weg 1, 14979 Großbeeren

E-Mail: franken@igzev.de

Neben den arbuskulären Mykorrhiza (AM) Pilzen gibt es eine Vielzahl anderer pilzlicher Organismen, die sich in der Wurzel tummeln. Einer davon ist der Basidiomyzet *Piriformospora indica*. Er besiedelt die Wurzeln aller bisher untersuchten Pflanzen und zeigt bei vielen von ihnen erstaunliche Wachstums-effekte. Gemeinsame Untersuchungen mit der Arbeitsgruppe von Professor Karl-Heinz KOGEL an der Universität Gießen haben ergeben, dass *P. indica* bei Gerstenpflanzen den Ertrag deutlich steigern und gleichzeitig die Pflanze schützen kann. Im Gegensatz zu den AM-Pilzen induziert er Resistenzen aber nicht nur gegen Wurzelpathogene, sondern auch gegen die Blatt-krankheit 'Echter Mehltau'. Dabei scheint keiner der bisher bekannten Signaltransduktionswege eine Rolle zu spielen. Stattdessen konnte eine Erhöhung verschiedener Antioxidantien beobachtet werden, die die Pflanze gegen das Pathogen schützen. Die Besiedelung der Wurzeln erfolgt nach einem gänzlich anderen Prinzip als bei den AM Pilzen. *P. indica* scheint den programmierten Zelltod der Wurzelzellen zu induzieren und so findet man ihn nur in toten Pflanzenzellen. Deshalb ist es nicht verwunderlich, dass man bei einer zu hohen Inokulumsdosierung auch negative Effekte erreichen kann. Am IGZ untersuchen wir nun, wie dieser Pilz im modernen Anbau von Tomaten eingesetzt werden kann.

## Literatur

FAKHRO, A., D.R. ANDRADE-LINAES, S. VON BARGEN, M. BANDTE, C. BÜTTNER, R. GROSCH, D. SCHWARZ, P. FRANKEN, 2010: Impact of *Piriformospora indica* on tomato growth and on interaction with fungal and viral pathogens. Mycorrhiza 20, 191-200.

WALLER, F., B. ACHATZ, H. BALTRUSCHAT, J. FODOR, K. BECKER, M. FISCHER, T. HEIER, R. HÜCKELHOVEN, C. NEUMANN, D. VON WETTSTEIN, P. FRANKEN, K.-H. KOGEL, 2005: The endophytic fungus *Piriformospora indica* reprograms barley to salt-stress tolerance, disease resistance, and higher yield. PNAS 102, 13386-13391.