



CEPLAS

Cluster of Excellence on Plant Sciences



CEPLAS

ANNUAL

REPORT



2014



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2014

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1.

General presentation

Summary

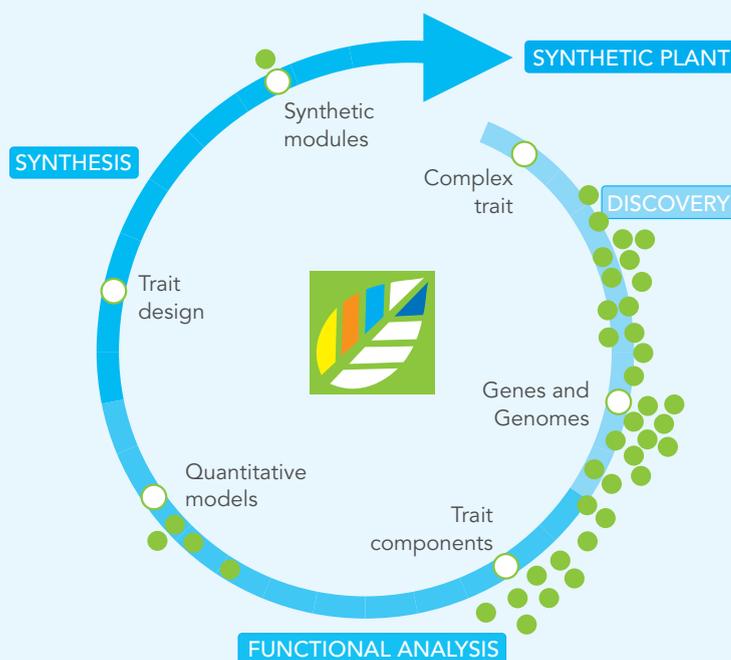
The global demand for plant products is increasing with unprecedented pace and it has been estimated that agricultural yields will have to double by the year 2050. However, global change, in particular altered precipitation and temperature patterns, is challenging the sustained production of crops and thus the agronomic base of human civilisation. Simultaneously, arable land is becoming scarce due to increased erosion and population pressure. Meeting the continuously increasing demand will require innovative strategies for crop improvement that aim at enhancing yield, without compromising increases in the use of water, nutrients, and soil, or diminished resistance to pests.

CEPLAS aims at achieving a fundamental understanding of the genetic mechanisms that enable plants to adapt to adverse environmental conditions and constraints. CEPLAS employs a research strategy that is driven by comparative evolutionary analyses in combination with modern synthetic biology. Specifically, the CEPLAS researchers investigate the mechanistic basis and genetic architecture of selected complex traits that have a crucial impact on adaptation to limited resources and are therefore of outstanding importance in designing and breeding the crops of the future:

- Annual and perennial life histories
- C₄ photosynthesis (photosynthetic carbon conversion efficiency)
- Plant/microbe interactions
- Metabolic interactions

Our vision

Through transformative science and innovative training, CEPLAS contributes to meeting the grand challenge of the 21st century, maintaining a sustainable food and energy base for human societies. CEPLAS raises the awareness for the importance of plant science for societal and economic progress and it serves as a platform for collaboration and interactions between academia and private entities. CEPLAS integrates over scientific disciplines to develop new paradigms in plant science.



At the 2014 Retreat, CEPLAS Young Researchers self-assessed their projects and allocated them to successive stages of the CEPLAS research programme.

It is expected that a significant portion of the ongoing projects will transition to the Functional Analysis phase in 2015.

Second year achievements

The first year of CEPLAS was dedicated mostly to recruiting the CEPLAS faculty (nine faculty positions) and research staff and to the implementation of the (infra-)structure and organisation of the cluster. We started our Graduate School and Postdoc Programme in summer 2013, which now involves around 50 young researchers working on CEPLAS-related research projects. The main focus of the second CEPLAS year was on implementation of the research programme and on establishing strong links between the research areas. In addition, thanks to advice by our Scientific Advisory Board, we began to refocus parts of our research programme. As a consequence, Research Areas A and B now initiated a joint programme focusing on the structure and function of leaves, at the interface between developmental genetics, biochemistry, and physiology. Research Areas C and D are jointly developing the research field metabolic and genetic plant-microbiota interactions. Considering our initial strategy of dividing the work programme into three distinct phases (discovery, functional analysis, and synthesis), we are glad to see that all research areas are now transitioning from the discovery to the functional analysis phase, with some project already entering the synthesis phase.

In 2014, we managed to fill two more W3 positions. *Dr. Gunther Döhlemann* (Terrestrial Microbiology) started in May; *Dr. Markus Pauly* (Cell Biology and Biotechnology) accepted the offer and will start in summer 2015. For the last open faculty position negotiations are still ongoing.

To intensify the translational aspects of research and the career promoting components of our training programmes, we enlisted expert support by *Dr. Günter Strittmatter* (formerly Head of Research and Development at KWS SAAT AG) as consultant for technology transfer and cooperation management. Spearheaded by *Dr. Strittmatter*, we organised a range of meetings with partners from industry over the past year, which have already resulted in joint research projects. In addition, the CEPLAS Training Committee and *Dr. Strittmatter* jointly organised excursions to industrial partners for our young researchers.

The curriculum of our Bachelor Programme in Quantitative Biology was finalised and approved by both universities. Accreditation by an external agency is ongoing and we expect to start in October 2015 with our first class.

As a major part of our public outreach programme we developed a public lecture series, a new Facebook page (<https://www.facebook.com/plantsoftomorrow>), and educational programmes. In response to comments by the Scientific Advisory Board, the CEPLAS web page was fully redesigned and has received more than 100,000 page hits since. The CEPLAS Young Researchers developed the Planter's Punch series (<http://ceplas.eu/en/communications/planters-punch-archive/>) to present each month a specific aspect of the CEPLAS research programme in an easy to understand format.

With the appointment of two platform leaders and the acquisition of new mass spectrometry instruments the CEPLAS Plant Metabolism and Metabolomics Laboratories were implemented. The platform leaders introduced the platform on CEPLAS Friday and conducted surveys and face-to-face meetings with CEPLAS-PIs to maximise the impact for the CEPLAS community. First experiments for CEPLAS members and external collaborators are ongoing. Moreover, the CEPLAS imaging platform headed by *Alga Zuccaro* was established at the Cologne Biocenter.

Major goals for 2015

- Finalise all faculty appointments
- Recruit the new cohort of CEPLAS postdoctoral fellows in an international selection workshop
- Intensify public outreach activities and outreach to policy makers
- Further integrate across research areas
- Strengthen translational aspects of CEPLAS activities
- Initiate the CEPLAS Bachelor Programme in Quantitative Biology



2.

Organisation

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Organisational structure

CEPLAS is partitioned in four **Research Areas**, each headed by a research area coordinator and co-coordinator.

All research area coordinators are part of the **CEPLAS Steering Committee**, together with the cluster speaker and deputy speaker, the equal opportunity representative and one representative of the young researchers. Additionally one representative of the Forschungszentrum Jülich is invited to the Steering Committee meetings.

All members of the Steering Committee were elected by the annual **General Assembly** (July 2012, October 2013 and 2014). The committee is responsible for the overall operation and development of the cluster, allocation of resources and preparation of site evaluations.

The **Executive Board** composed of speaker and deputy speaker is responsible for the implementation of the decisions made by the Steering Committee.

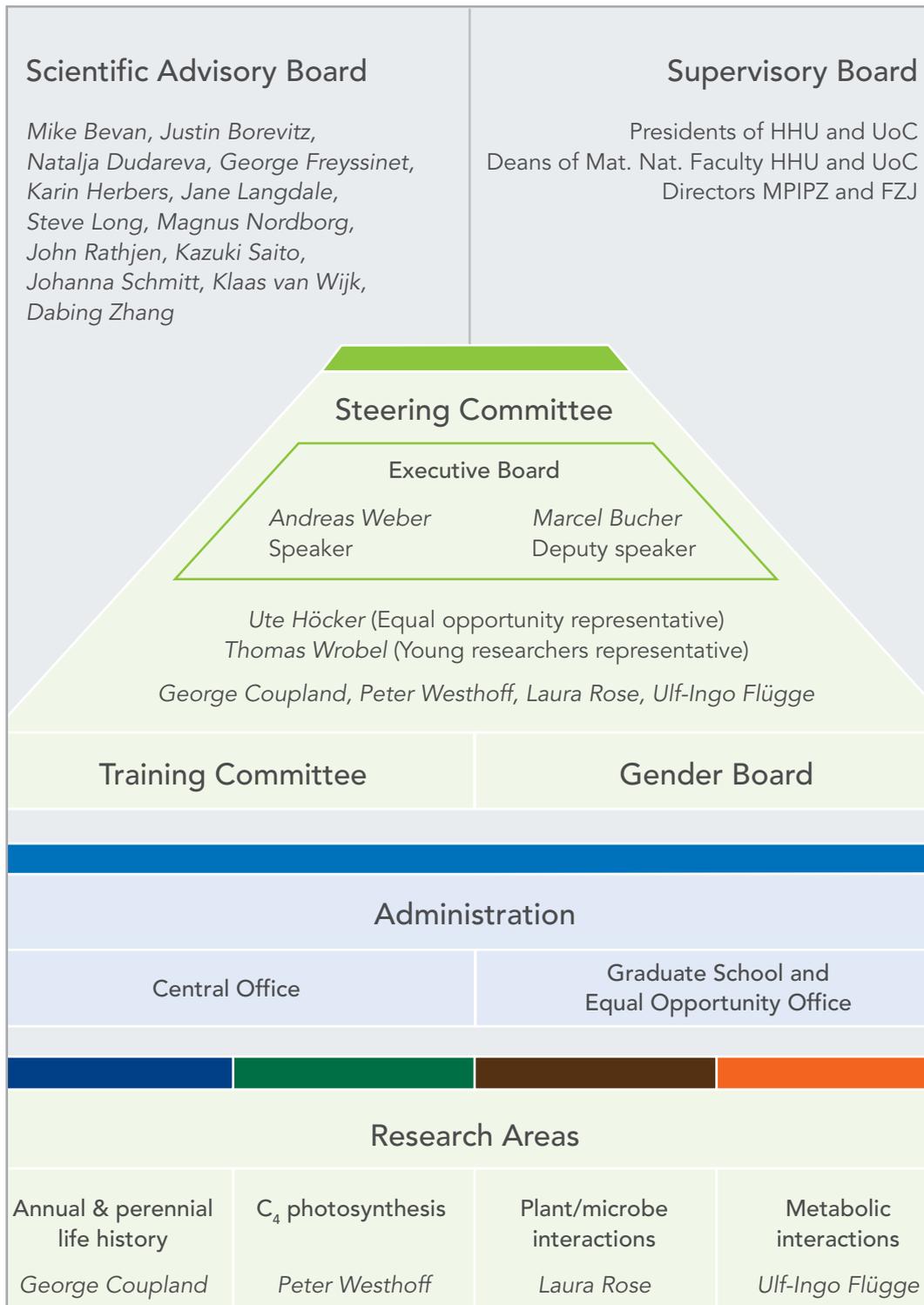
CEPLAS is consulted by a **Scientific Advisory Board** (SAB) that is composed of scientists from academia and industry, which meet on an annual basis. The SAB receives and reviews the annual report of the cluster and provides an assessment to the Supervisory Board. Moreover, the SAB is responsible for the external evaluation of the cluster (annual evaluation report, mid-term and comprehensive assessment) and provides advice on hiring decisions, scientific and structural planning, development of the training programmes, implementation of equal opportunity measures, and overall development of the cluster.

The cluster administration, i.e., the **Central Office** and the **Graduate School and Equal Opportunity Office** are located at the interface between CEPLAS researchers and the Steering Committee.

The **Training Committee** is responsible for overseeing the development of the CEPLAS training programmes and its tasks include the general management of the training programmes, quality control of these programmes, evaluation of courses, and overseeing the recruitment procedure. The Training Committee is composed of the speaker of the Graduate School (*Ute Höcker*), the speaker of the Postdoc Programme (*Rüdiger Simon*), and a third PI representing the MIPZ (*George Coupland*) so that both universities and the MIPZ are represented as the majority of the CEPLAS members belong to these institutions. Further members are the representatives of the young researchers programmes, currently *Thomas Wrobel* as doctoral researcher representative and *Wilma van Esse* as postdoc representative. The coordinators of these programmes and the coordinator of the Bachelor Programme are members with an advisory vote.

The **Gender Board** consists of three PIs, i.e., the equal opportunity representative (*Ute Höcker*) and two other PIs (*Laura Rose*, *Jane Parker*), as well as the coordinator of the Equal Opportunity Office.

Details about the CEPLAS management structure and regulations between participating institutions are specified in the statutes of the cluster ("Clusterordnung") and the cooperation agreement between both universities and the non-university institutions. Both documents have been signed in November 2013.





3.

Research programme

3.1 Research Area A

Elucidation and manipulation of the mechanisms that differentiate annual and perennial life histories

Summary

In this research area we focus on understanding the regulation of traits that differentiate the annual and perennial life cycles. A major aim is to identify regulatory modules that diverged between annuals and perennials to confer phenotypic differences in meristem function, flowering behaviour, root growth or longevity.

We exploit two major model systems, which are closely related Brassicaceae species, particularly in the *Arabidopsis* and *Arabis* genera, as well as *Hordeum* species related to barley. A range of approaches are used including forward genetics, transcriptomics based on RNAseq, comparison of newly acquired genome sequences from phylogenetically closely related species, ChIPseq for interspecies comparison of transcription factor targets and development of algorithms for inferring gene regulatory networks from transcriptome data.

A major achievement this year was the completion and publication of the genome of *Arabis alpina*, our model perennial species. This sequence underlies application of forward and reverse genetic approaches in this species and facilitates inter-species comparisons with related annuals. In addition availability of the genome sequence allowed high resolution genotyping of progeny derived from inter-species crosses between *A. alpina* and its sister annual, *A. montbretiana*. The resolution of this mapping enabled us to confidently ascertain that much of the annual genome has been introgressed into the perennial parent. This genotyped material will form the basis of mapping loci contributing to traits of interest and will facilitate the construction of a library of near isogenic lines. A surprise came from the ChIPseq data and the comparison of transcription factor targets in different species. A much lower proportion of target



▪ Coordinator

George Coupland

▪ Co-coordinator

Rüdiger Simon

▪ Faculty

Maria Albani

Petra Bauer

Ute Höcker

Karl Köhrer

Markus Kollmann

Maarten Koornneef

Peter Nürnberg

Richard Reinhardt

Laura Rose

Ulrich Schurr

Klaus Theres

Miltos Tsiantis

Maria von Korff

Wolfgang Werr

genes of orthologous transcription factors was conserved between related plant species than had previously been expected. However, a useful outcome of this analysis was that the small proportion of conserved targets seem to be those that are particularly important in the core regulatory function of the transcription factor. We exploited this feature to identify key genes downstream of a MADS box transcription factor that is a major repressor of flowering in annuals and perennials.

In addition, the *Hordeum* system was developed significantly by obtaining a more detailed phenotypic description of traits of interest, particularly tillering behaviour in annuals and perennials, and this was extended to a molecular analysis through RNAseq. Strong interactions were established across the collaborating institutions within the research area. The comparative system based on *A. alpina*, *A. montbretiana* and *A. thaliana* was used by groups at the MPIPZ, HHU and UoC to analyse differences in flowering behaviour, root growth and for genomic comparisons. Also the development and outgrowth of axillary meristems was studied both in the Brassicaceae and the *Hordeum* systems enabling comparisons at the phenotypic and molecular levels. This focus builds on long-term interests of the Theres group at the MPIPZ working on the Brassicaceae and now is extended by the von Korff group at HHU into the *Hordeum* system.

Interactions across research areas have developed. Notably, we have planned an extension of Research Area A into metabolic regulation and particularly how perennials store metabolites between growing seasons. The topic will be led by Petra Bauer at HHU and has strong connections with Research Area D, mainly at UoC. In addition, we have interactions with Research Area C where the bacterial and fungal microbiota of *Arabis alpina* roots have been analysed by groups at MPIPZ and UoC, respectively. This analysis provides the possibility of analysing how the microbiota change with time over the longer life cycle of a perennial. There are also technological connections to Research Area B, particularly in the analysis of RNAseq data and its exploitation to deduce regulatory networks utilising algorithms developed by Markus Kollmann's group.

The work of the research area is making a scientific impact in defining model systems with all of the required phenotypic description, genomic information and technological methodology to understand the divergence of annual and perennial life histories. Publications have been widely recognised, such as the one describing the *Arabis alpina* genome, which was published along with a News and Views article in *Nature Plants*. Also members of the research area have received many invitations to speak at international meetings presenting the data obtained in these projects. Discussions are ongoing with several companies to consider application of results obtained, notably in areas associated to flowering such as how to overcome juvenility.

A major goal for 2015 is to further develop the introgression material by characterising near isogenic lines and to complete the genome sequence of *Arabis montbretiana* to allow detailed comparison to annual and perennial sisters. In addition, we expect to exploit our CRISPR/CAS system for reverse genetic analysis in *Arabis alpina*, and thereby to use reverse genetics to test specific hypotheses related to flowering of perennials. In the Brassicaceae system we aim also to start metabolic analysis to define the location and identity of storage compounds in perennials, and to design RNA profiling experiments to investigate differences in developmental senescence patterns between annual and perennial sister species. In the *Hordeum* system we expect to map in detail loci associated with tillering and to characterise at the molecular level some of the candidate genes.

Project A1 Genotypic and phenotypic analysis of interspecific hybrids between *A. montbretiana* and *A. alpina*

Researchers

Christiane Kiefer
René Richter

Project leader

George Coupland

Project type

Postdoc project

Project start

01.01.2013

Cooperations

(other participating
CEPLAS and non-CEPLAS
researchers)

Maria Albani
Marcel Bucher
Laura Rose
Rüdiger Simon
Korbinian Schnee-
berger (MPIPZ)

Publications

Willing EM, Rawat
V, Mandáková T et
al. (2015) *Nature
Plants* 1, doi 10.1038/
nplants.2014.23

Aim of the project

This project exploits plants generated by intercrossing annual *Arabis montbretiana* to perennial *Arabis alpina* to identify genes that confer phenotypic differences between the species.

Context within CEPLAS

Genotyped plants will be provided to other groups to map traits of interest. Notably the Simon group to study roots, and with the Rose group we study genes that differ in activity between the species.

First results

We used genotyping by sequencing to genotype around 500 plants derived from the original cross. Around 20,000 polymorphic markers were identified and scored. Most of the *A. montbretiana* genome has been introgressed into *A. alpina*. Plants that acquired competence to flower in response to vernalization earlier than *A. alpina* were identified.

Scientific/commercial impact

This material is unique in allowing mapping of interspecies traits associated with annual/perennial divergence in the Brassicaceae.

Project A2 Comparative transcriptome profiling of meristem states in annual and perennial *Arabis* and *Arabidopsis* species and hybrids

Researcher

Helge Pallakies

Project leader

Rüdiger Simon

Project type

Postdoc project

Project start

01.05.2013

Aim of the project

The aim of this project is the definition of regulatory gene networks contributing to either annualism or perennialism in root development. We therefore perform comparative phenotypic and transcriptome analyses in two closely related *Arabis* species, the perennial *Arabis alpina* Pajares and the annual *Arabis montbretiana*.

Context within CEPLAS

Both the phenotypic and transcriptomic description and comparison of the root development of *A. alpina* and *A. montbretiana* provides the basis for future studies in the next steps of the project: The phenotypic observations are the key to screens for annual/perennial specific root growth traits in *A. alpina*/*A. montbretiana*-hybrid populations. The characterisation of the transcriptomes of both species provides a database to identify annual/perennial specific candidate genes for subsequent functional analyses.

First results

The characterisation of early root development revealed differences in root length and emergence of lateral roots. From the first day up to 14 days after germination, *A. alpina* exhibited

smaller roots compared to *A. montbretiana*. Furthermore, lateral roots emerged at later stages in *A. alpina* compared to *A. montbretiana*.

The observation of later developmental stages up to initiation of flowering in *A. montbretiana* revealed differences in growth speed and root system expansion.

Furthermore, differences in lateral root angles caused by differences in gravitropic responses are indicated. Experiments with both species and wild variations (collected by S. Woetzel, MPIPZ) aiming at gravitropic responses are ongoing.

The transcriptome analysis with replicates for three early root meristem stages for both species is in progress. RNA has been successfully isolated, libraries could be synthesised and sequenced. Mapping on the basis of genome data from K. Schneeberger / E.-M. Willing is in progress and differential expression within each species and across both species will follow.

Scientific/commercial impact

Our approach will deepen our understanding of the factors that allow perennial plants to develop an expanded root system. We will later try to use the gene modules identified here to extend the root system of an annual plant.

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

George Coupland

Christiane Kiefer

Fabio Fiorani (Jülich Plant Phenotyping Centre, FZJ)

Kerstin Nagel (FZJ)

Korbinian Schneeberger (MPIPZ)

Eva-Maria Willing (MPIPZ)

Stefan Woetzel (MPIPZ)

Publications

Pallakies H & Simon R (2014), *Molecular plant* 7(11):1619-1636.

Project A3 Comparison of annual-perennial species pairs across the Brassicaceae

Aim of the project

Comparison of annual-perennial species pairs across the Brassicaceae.

Context within CEPLAS

We are using comparative genomic methods to determine the genes underlying perennial/annual life history transitions. Our analysis covers four species pairs consisting of an annual and a perennial in each pair (*Arabidopsis thaliana*/*Arabidopsis lyrata*, *Arabis montbretiana*/*Arabis alpina*, *Draba nemorosa*/*Draba hispanica* and *Arabis verna*/*Aubrieta canescens*) plus the annual *Capsella rubella*.

First results

Orthologous genes that have been deleted or become nonfunctional in all annual species are potential candidate genes and we aim to find these genes. After grouping the proteomes of all species into families based on sequence similarity, we determined those families where genes were present in perennials but missing in all annuals.

Scientific/commercial impact

Gene families matching this description include a number of potential regulatory genes including transcription factors and kinases. These genes are candidates for future experimental evaluation and validation within Research Area A.

Researcher

Andrew Heidel

Project leader

Laura Rose

Project type

Postdoc project

Project start

01.04.2013

Project A4 Genotypic and phenotypic analysis of tiller development in cultivated (*Hordeum vulgare*) and wild barley species (*H. v. spp. spontaneum*, *H. bulbosum*)

Researcher

Wilma van Esse

Project leader

*Maria von Korff
Schmising*

Project type

Postdoc project

Project start

01.07.2013

Cooperations

(other participating
CEPLAS and non-CEPLAS
researchers)

*George Coupland
Maarten Koornneef
Rüdiger Simon
Laura Rossini
(University of Milano,
Italy)
José Jiminez Gomez
(MPIPZ)*

Aim of the project

Shoot architecture is one of the primary morphological structures that impact crop productivity. The aim of the project is to study barley mutants impaired in tillering to identify the genetic and molecular basis of axillary meristem initiation and outgrowth.

Context within CEPLAS

The project directly contributes to the Research Area A as it aims at deciphering the molecular basis of axillary meristem development in barley.

First results

Wilma van Esse has successfully conducted macroscopic and microscopic phenotyping of AXM development in low and high-tillering mutants. She has carried out RNA-sequencing analyses on crown tissue of these tillering mutants to a) map the mutant loci using introgression lines and b) characterise expression changes in these mutants.

Scientific/commercial impact

Understanding the genetic basis of tillering is important for targeted manipulation of plant architecture and yield in temperate cereals.

Project A5 Transcriptome data analysis and optimal experimental design

Researcher

Armin Sadat Khonsari

Project leader

Markus Kollmann

Project type

Ph.D. project

Project start

01.04.2013

Aim of the project

Establish a network inference algorithm that is
(i) unbiased,
(ii) can identify links that are in principle non-inferable because of lack of information, and
(iii) can operate in the $p > n$ limit.

Context within CEPLAS

Inference of gene regulatory networks is an essential step in generating synthetic plants, such as generating perennial plants from annual plants or C_4 plants from C_3 plants.

First results

We have developed the algorithm, tested it for synthetic data, and showed that it outperforms other state of the art algorithms. An immediate application is to estimate the number of required transcription data to reconstruct a certain percentage of a gene regulatory network with a given false positive rate.

Scientific/commercial impact

The first paper is entitled 'Inferability of complex networks' and will include among biotechnological applications also a section of how much data ecommerce cloth retailer have to accumulated such that they can estimate both the true body sizes of their customers and the true size of their offered goods.

Project A6 Analysis of the roles of PERPETUAL FLOWERING 1 direct target genes in perennial flowering and their divergence in sister annual species

Aim of the project

To use ChIPseq and RNAseq to identify the target genes of the PEP1 transcription factor in perennial *Arabis alpina* and to compare these with the targets of the orthologous protein FLC in annual *Arabidopsis thaliana*.

Context within CEPLAS

Differences in transcription of PEP1 and FLC is a major driver of different seasonal flowering patterns between annuals and perennials. Here we address how similar the functions of the two proteins are.

First results

Target genes of PEP1 were identified. There are similarities and differences between this set and FLC targets. Most flowering gene targets are conserved, and the binding sites recognised by the transcription factors are present in both species. However there are differences in other classes of target.

Scientific/commercial impact

So far the targets of orthologous transcription factors have not been compared in plants. This provides a novel data set of wide interest.

Researcher

Vicky Tilmes

Project leader

George Coupland

Project type

Ph.D. project

Project start

01.11.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Martin Hülskamp

Julieta Mateos

(Fundación Instituto Leloir, Buenos Aires, Argentina)

Eva Willing (MPIPZ)

Project A7 Competence for axillary meristem formation: analysis of the lateral suppressor pathway in *Arabidopsis*

Aim of the project

To investigate the role of axillary meristem initiation in *Arabis alpina* and to identify the targets of *LATERAL SUPPRESSOR (LAS)*.

Context within CEPLAS

The analysis of axillary meristem formation in *A. alpina* and the elucidation of the role of genes involved in this process will help us to better understand the diversification of annual and perennial life histories.

First results

- The pattern of axillary bud formation in *A. alpina* during vegetative and reproductive development was analysed using a stereomicroscope and RNA *in-situ* hybridisation. Our preliminary data suggest that axillary buds that remain vegetative are produced before vernalisation, whereas axillary buds that flower are produced after the onset of vernalisation. We detected a zone near the shoot tip, where axillary meristem formation seems to be inhibited.
- Knock-down constructs targeting the axillary meristem regulator *LATERAL SUPPRESSOR* have been introduced into *Pajares* and *pep1* plants.
- The INTACT method was established to isolate nuclei from the *LAS* expression domain. These nuclei will be used for transcriptome analysis.

Scientific/commercial impact

Better understanding of axillary meristem formation in perennial plants.

Researcher

Udhaya Ponraj

Project leader

Klaus Theres

Project type

Ph.D. project

Project start

01.10.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Maria Albani

George Coupland

Christiane Kiefer

Project A8 Light control of plant growth and development in seasonal and perpetual *Arabis alpina*

Researcher

Panpan Jiang

Project leader

Ute Höcker

Project type

Ph.D. project

Project start

01.10.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Maria Albani

George Coupland

Aim of the project

- To understand the effect of day length on flowering time and inflorescence development and their possible interactions with the vernalisation pathway/perennialism in perpetual and seasonal *A. alpina* accessions.
- To analyse photomorphogenic responses in *A. alpina* (seedling deetiolation, shade avoidance)

Context within CEPLAS

This project helps achieve the main goal of Research Area A, i.e., to understand perennialism in *A. alpina*.

First results

- We found that *A. alpina* responds to simulated shade very differently from its relative *A. thaliana*. Morphological shade avoidance responses in *A. alpina* appear limited to internode elongation and do not include seedling or leaf parameters.
- Screening of an EMS-population of *A. alpina* has identified putative light signalling mutants.
- Experiments on the effect of day length on *A. alpina* are established and are ongoing.

Scientific/commercial impact

Provide the grounds for modifications in plant architecture and life history.

Project A9 Genetic dissection of natural variation in tiller development in cultivated and wild barley

Researcher

Agatha Walla

Project leader

Maria von Korff

Schmising

Project type

Ph.D. project

Project start

01.02.2014

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Maarten Koornneef

Rüdiger Simon

José Jimenez Gomez

(MPIPZ)

Frank Blattner

(IPK Gatersleben)

Aim of the project

The objective of this project is to use natural genetic diversity between cultivated and wild annual barley and between annual and perennial *Hordeum* species to decipher the genetic control of tillering.

Context within CEPLAS

The project directly contributes to the goals of Research Area A as it aims at understanding the genetic differences in AXM development within and between annual and perennial *Hordeum* species.

First results

Agatha Walla has scored the development of annual and perennial barley accessions. She has generated re-sequencing data for candidate genes in annual and perennial barley species. In addition, she has analysed RNA-sequencing data of a natural high-tillering mutant in barley.

Scientific/commercial impact

Perennial cereals are a long-standing breeding goal as they have improved root systems and are adapted to low-input agricultural systems.

Project A10 Analysis of an evolutionary conserved module regulating root system development in monocot and dicot species

Aim of the project

Characterise barley homologues of genes known to be important for root development in *Arabidopsis*, maize and rice.

Context within CEPLAS

Perennial plants grow a deeper and more widespread root system than annual plants. To understand how they are able to do that, it is important to know the key regulators of root growth not only in model plants but also in the crop plants.

First results

First gene homologues and promoter regions were cloned and characterised by transient expression in tobacco leaves.

Scientific/commercial impact

Results could enable to grow plants with a root system that is more resistant to draught, water stress and could prevent soil erosion.

Researcher

Gwendolyn Kirschner

Project leader

Rüdiger Simon

Project type

Ph.D. project

Project start

01.12.2013

Cooperations

Maria von Korff
Schmising

Project A11 The control of adventitious root formation in the perennial *Arabis alpina*

Aim of the project

Study the process of adventitious root formation in the perennial life cycle of *A. alpina*.

Context within CEPLAS

Perennial species are more prone to clonal propagation than annuals. Understanding of this process will contribute to the aims of RA A to characterise perennial traits.

First results

n.a. - Project start 1.10.2014

Scientific/commercial impact

Understanding of the molecular mechanisms regulating this process might have commercial impact as horticultural plant industries depend on the successful production of adventitious roots in stem cuttings.

Researcher

Priyanka Mishra

Project leader

Maria Albani

Project type

Ph.D. project

Project start

01.10.2014

Project A12 Nutrient recycling in the perennial plant *Arabis alpina*

Aim of the project

Perennialism relies on an optimised soil nutrient use efficiency based on the recycling and storage at the end of the vegetation period, often involving the formation of lateral storage branches. We will investigate differences in the signalling of nutrient recycling and storage branch formation that sustain the perennial/annual lifestyles during senescence.

Context within CEPLAS

Nutrient recycling upon senescence was likely a target during the evolution of annuals from perennials. Annuals disperse nutrients with seeds, while perennials retain them in storage organs. This hypothesis will be tested with regard to selected nutrients.

First results

n.a. - Project start 1.02.2015

Researcher

Anna Sergeeva

Project leader

Petra Bauer

Project type

Ph.D. project

Project start

01.02.2015

Cooperations

Maria Albani
George Coupland
Stanislav Kopriva
Klaus Theres

3.2 Research Area B

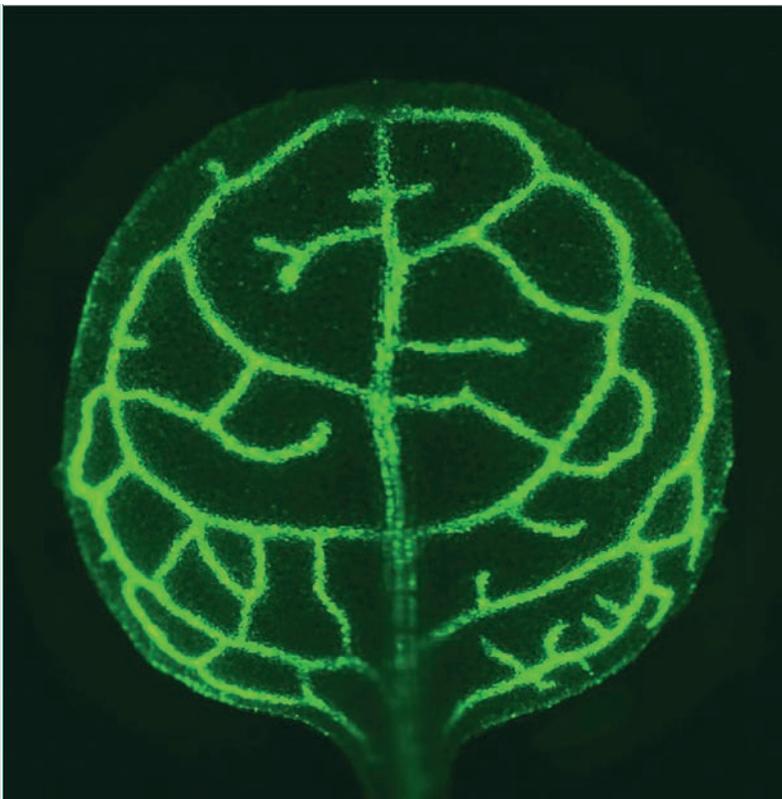
Decoding function and development of a C₄ leaf

Summary

This research area aims to understand the evolutionary transition from C₃ to C₄ photosynthesis in sufficient detail to create a synthetic C₄ plant from a C₃ species. For identifying the genetic blueprint of C₄ photosynthesis the research area focuses primarily on evolutionary C₃-to-C₄ transitions in dicot genera and uses the C₃ plant *Arabidopsis thaliana* for forward-genetics and experimental evolution approaches as well as chassis for synthetic biology. CEPLAS funds are complemented by funds raised in the framework of the 3to4 project (EU), the International Research Training Group 1525 (DFG), the Research Group 1186 (DFG) and the Priority Programme Adaptomics (DFG).

Following the general CEPLAS research strategy, the gene discovery phase was started two years ago with extensive transcriptome comparisons in the genera *Cleome* (Cleomaceae/Brassicales) and *Flaveria* (Asteraceae). These genera are characterised by the concomitant occurrence of C₃ and C₄ species and, in the case of *Flaveria*, by a large number of C₃-C₄ intermediates. The comparisons of total leaf transcriptomes (Bräutigam et al, 2011; Gowik et al, 2011; Mallmann et al, 2014) of C₃, C₄ and C₃-C₄ intermediate species of *Cleome* and *Flaveria* allowed to define a core set of genes whose expression domains and strength was altered during the transition from C₃ to C₄ photosynthesis in both species. These data were instrumental for developing a predictive evolutionary model of the origin of C₄ photosynthesis (Heckmann et al, 2013; Mallmann et al, 2014). The model predicts that evolution proceeded by individually adaptive steps on a Mount Fuji-shaped fitness landscape (Heckmann et al, 2013).

The model was further refined and extended by showing that the photorespiratory CO₂ pump present in all C₃-C₄ intermediates represents a pre-adaptation for the C₄ system by providing important components for the establishment of the C₄ cycle (Mallmann et al, 2014).



▪ Coordinator

Peter Westhoff

▪ Co-coordinator

Martin Hülskamp

▪ Faculty

Oliver Ebenhöf

Georg Groth

Ute Höcker

Karl Köhrer

Markus Kollmann

Maarten Koornneef

Martin Lercher

Veronica G. Maurino

Peter Nürnberg

Richard Reinhardt

Laura Rose

Ulrich Schurr

Kai Stühler

Miltos Tsiantis

Andreas P. M. Weber

The leaf transcriptome studies were complemented by developmental transcriptomics in a phylogenetic framework. The differentiation of leaves, from the primordia up to fully matured leaves, was followed in C₃/C₄ pairs of *Cleome* (Külahoglu et al, 2014) and *Flaveria* (Billakurthi et al, unpublished). Interestingly, in both cases leaf development in the C₄ species is lagging behind that of the C₃ species. This causes a delay in mesophyll differentiation and possibly also allows a more dense venation pattern (Külahoglu et al, 2014).

Beyond the recently published data, the research area has generated scientific impact by providing large comparative RNA-Seq atlases of C₃ and C₄ species pairs.

In addition, jointly with the Schranz (Wageningen), Hibberd (Cambridge), and Zhu (Shanghai) groups, the genome of the C₃ species *Taranaya hassleriana* has been sequenced and published; sequencing of the C₄ Cleomaceae *Gynandropsis gynandra* has been completed and will be publically released and published in 2015.

Since the characteristic feature of leaf anatomy of C₄ plants, a photosynthetically active bundle-sheath, is already found in many C₃ angiosperms, among them *Arabidopsis*, this plant was used for genetic approaches to discover genes involved in bundle-sheath ontogeny and general leaf anatomy. Stable mutant phenotypes affecting bundle-sheath size have been detected and are currently being mapped by sequencing. In parallel to the mutational approaches the natural variation of *Arabidopsis* for leaf anatomical traits was explored by using existing MAGIC-line type populations.

Since a photosynthetically active bundle-sheath is an anatomical prerequisite of C₄ evolution, the identification of the genetic network defining Kranz anatomy is of absolute priority. Understanding the regulatory peculiarities of the C₄ biochemistry and enzymology is the second major challenge to be overcome. Finally, the engineering of C₄ anatomical and biochemical/enzymological submodules into C₃ species requires quantitative models explaining the behaviour of these submodules. We envisage that through intensifying the interactions with the developmental geneticists of Research Area A, CEPLAS will provide a unique opportunity for achieving significant contributions in understanding and engineering C₄ photosynthesis in the near future.

Project B1 Mesophyll and bundle-sheath-specific transcriptomes of C₃/C₄ species pairs

Researcher

Stefanie Schulze

Project leader

Peter Westhoff

Project type

Postdoc project

Project start

01.04.2013

Aim of the project

Identification of differentially expressed genes in mesophyll and bundle sheath cells of C₃ and C₄ species of the genus *Flaveria*.

Context within CEPLAS

The knowledge of yet unknown genes that are differentially expressed in the two leaf cell-types of C₄ photosynthesis will be instrumental in understanding the transition from C₃ to C₄ photosynthesis. This will help to identify new players for the transformation of a C₃ into a C₄ species.

First results

Epitope-tagged ribosomal protein RPL18 was specifically expressed in the mesophyll and bundle sheath cells of the transformable C₄ species *Flaveria bidentis* and is used to compare the transcriptomes of these two cell types. In a complementary approach laser micro dissection is being optimised to provide mesophyll and bundle sheath tissue fractions of *Flaveria robusta* (C₃) and *Flaveria bidentis* (C₄) for the analysis of their transcriptomes.

Scientific/commercial impact

Transcriptome/translatome patterns of mesophyll and bundle sheath cells of dicotyledonous C₄ plants are not yet available nor have transcript profiles of mesophyll and bundle sheath cells from closely related C₃ and C₄ species been compared.

Project B2 Circadian control of transcriptomes and proteomes of C₃/C₄ species pairs

Researcher

Thea Pick

Project leader

Andreas Weber

Project type

Postdoc project

Project start

01.04.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Ilka Axmann

Markus Kollmann

Peter Westhoff

Aim of the project

Unravelling the circadian control of C₄-associated genes through comparative transcriptomics of C₃ and C₄ species pairs in the genus *Flaveria*.

Context within CEPLAS

We will for the first time identify altered temporal gene expression patterns between C₃ and C₄ plants. This will identify candidate C₄ genes that are involved in regulation, development, and establishment of C₄ photosynthesis. These candidates will lead to identification of trait components and the dataset will be used by the Kollmann group to build quantitative models for functional analysis (Phase 2) of the complex trait C₄.

First results

All RNA-seq libraries have been generated and sequenced. Data analysis is ongoing.

Scientific/commercial impact

For the first time temporal regulation of C₄ candidate genes will be assessed. This will contribute to the goal of a synthetic C₄ plant/introducing C₄ photosynthesis into C₃ plants.

Project B3 Experimental evolution towards C₄

Aim of the project

Analysis of anatomical and physiological traits of diverse *Arabidopsis thaliana* ecotypes grown under low (190ppm) and high (700ppm) CO₂ concentrations to identify ecotypic differences for traits related to carbon assimilation efficiency (biomass, photosynthetic rates).

Context within CEPLAS

Artificially high and low CO₂ concentrations during growth reveal genetic plasticity of *Arabidopsis* towards adaptation. Transcriptomes and anatomy of better performing ecotypes in combination with data from other CEPLAS projects working on C₄ and C₃-C₄ intermediate species, will reveal insight into regulatory factors and networks.

First results

Arabidopsis ecotypes displaying diverse phenotypes under altered CO₂ concentrations have been identified. Correlations to their geographic origins are not consistent throughout all ecotypes. Leaf samples have been sampled for RNAseq as well as for anatomical analyses.

Scientific/commercial impact

Discovery of mechanisms underlying adaptation to altered atmospheric CO₂ concentrations.

Researcher

Nadine Höcker

Project leader

Andreas Weber

Project type

Postdoc project

Project start

01.05.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Fabio Fiorani (Jülich Plant Phenotyping Centre, FZJ)

Project B4 Genetic variation for leaf anatomy in *Arabidopsis*

Aim of the project

Identification of genes relevant to C₄-photosynthesis through the analysis of C₃ plants.

Context within CEPLAS

The analysis of different leaf traits (thickness, size, chlorophyll content, and venation) enables understanding which trait has more impact on photosynthesis followed by QTL mapping to identify the responsible genes controlling each trait.

First results

The phenotyping of leaf traits has been conducted. Association mapping is done for all traits. There are several candidate genes as a result of the association mapping that need to be confirmed.

Scientific/commercial impact

Understanding the gene network controlling the leaf traits and their role in photosynthesis in C₃ plants enhances the possibility of identifying the gene network controlling photosynthesis in C₄ plants. In collaboration with group of Siegfried Jahnke, a software for leaf-venation was developed and will be soon freely available.

Researcher

Louai Rishmawi

Project leader

Martin Hülskamp

Project type

Postdoc project

Project start

01.05.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Maarten Koornneef
Ulrich Schurr
Siegfried Jahnke (FZJ)

Project B5 Genetic control of leaf anatomy in *C₃/C₄ Flaveria* and *Arabidopsis*

Researcher

Eva Willée

Project leader

Ute Höcker

Project type

Ph.D. project

Project start

01.05.2013

Cooperations

(other participating
CEPLAS and non-CEPLAS
researchers)

Martin Hülskamp

Maarten Koornneef

Peter Westhoff

Aim of the project

- Effect of light on the establishment of *C₄* traits
- Identify loci controlling leaf anatomy under high-light conditions

Context within CEPLAS

Understanding the genetic control of leaf anatomy in *C₄* photosynthesis is at the centre of Research Area B. This project helps achieve this goal by identifying loci controlling important leaf anatomical traits, such as leaf thickness, vein distance, palisade cell anatomy.

First results

We found that cell type-specific expression of mesophyll- and bundle sheath-specific genes is independent of light in *Flaveria* and *Arabidopsis* seedlings. Differentiation of bundle sheath cell size, in contrast, is strongly light-dependent. We have screened *Arabidopsis thaliana* accessions for variation in leaf anatomy in response to high light. Identified extreme genotypes will be used for QTL mapping of responsible loci. The AMPRIL mapping population is used for QTL mapping of vein density.

Scientific/commercial impact

Provide the grounds for implementing *C₄* photosynthesis into *C₃* plants.

Project B6 Identification of *C₄* genetic dominance by a comparative transcriptome analysis of *C₃* and *C₄ Flaveria spp.* and by an activation tagging approach with *Arabidopsis thaliana*

Researcher

Kumari Billakurthi

Project leader

Peter Westhoff

Project type

Ph.D. project

Project start

01.10.2013

Aim of the project

The project aims to identify genes affecting *C₄* leaf anatomy by comparative transcriptome profiling of leaf development in *F. robusta* (*C₃*) and *F. bidentis* (*C₄*) and by activation tagging with *Arabidopsis*.

Context within CEPLAS

The evolution of *C₄* photosynthesis required the establishment of a photosynthetically active bundle sheath. Understanding the genetic differences in the differentiation of the bundle sheath in *C₃* vs. *C₄* species would be a break-through in *C₄* research.

First results

Transcriptome data sets were sampled from leaves of different developmental stages of both *F. robusta* (*C₃*) and *F. bidentis* (*C₄*) and are presently being analysed for differences in transcript patterns and amounts. *Arabidopsis* plants expressing a plastid-targeted GFP in the bundle sheath cells were transformed with a T DNA construct containing the leaf specific ppcA promoter. T1 plants are being screened for deviations in marker gene expression and leaf anatomy.

Scientific/commercial impact

Knowledge of the genetic basis of bundle-sheath differentiation is imperative for *C₄* engineering of *C₃* crops.

Project B7 Unravelling the mechanisms that control bundle-sheath cell size in leaves of C₄ plants

Aim of the project

Determine factors that control the increased bundle-sheath cell size in leaves of C₄ plants and implement the mechanism into the C₃ plant *Arabidopsis thaliana*.

Context within CEPLAS

Altered bundle-sheath anatomy constitutes a crucial part of C₄ photosynthesis. Therefore creating a genetic module that regulates cell size selectively in this tissue is an initial step required for the implementation of C₄ anatomy into C₃ plants. Recombineering will be implemented for the efficient exchange of promoter structures that can be exploited in the context of other CEPLAS projects.

First results

Genes hypothesised to control cell size were cloned into appropriate vectors for plant transformation. The functionality of the recombineering system that will be used for the implementation of a bundle-sheath specific promoter, was verified.

Scientific/commercial impact

This work will develop a module that controls bundle sheath cell size.

Researcher

Thomas Wrobel

Project leader

Andreas Weber

Project type

Ph.D. project

Project start

01.10.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Kumari Billakurthi

Martin Hülskamp

Project B8 *In silico* exploration of paths towards C₄ metabolism

Aim of the project

We aim to expand our existing model for C₄ evolution by adding explicit dependencies on environmental factors and genotype-phenotype relationships, and to use this model to better understand paths towards C₄.

Context within CEPLAS

Our model will provide the foundation for developing optimal evolutionary strategies towards C₄ that combine artificial evolution and genetic engineering.

First results

The model has been extended to partition light energy optimally in C₃-C₄ intermediates, allowing for the simulation of light limiting environments.

Scientific/commercial impact

We aim for an improved understanding of both natural and artificial C₄ evolution, which will form the basis for experimental evolution efforts that aim at substantial yield increases of major crops.

Researcher

Esther Sundermann

Project leader

Martin Lercher

Project type

Ph.D. project

Project start

01.06.2014

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Andreas Weber

David Heckmann

(iGRAD *plant*)

Project B9 Alterations to the regulation of C₄ enzymes during evolution of C₄ photosynthesis

Researcher

Meike Hüdig

Project leader

Veronica G. Maurino

Project type

Ph.D. project

Project start

01.02.2014

Aim of the project

The project aims to (1) identify and characterise all NAD-MEs of *Tarenaya hassleriana* and *Cleome gynandra* as close relatives to *Arabidopsis thaliana* to analyse and understand changes that occurred during the C₄ evolution. Additionally, (2) the role of NAD-ME in *A. thaliana* will be studied by using *knock-out* lines of NAD-MEs and mMDHs.

Context within CEPLAS

Understanding the role of NAD-ME in C₃ and C₄ species will help to reveal changes that convert a housekeeping enzyme to the major CO₂ feeding enzyme of RubisCO in C₄ photosynthesis.

First results

Cloning of all NAD-MEs of both *Cleomaceae* is completed. Enzymatical characterisation is ongoing. (2) 12 *knock-out* lines of both NAD-MEs and both mMDHs in combinations up to 1 triple *knock-out* line are confirmed, 3 additional were produced. Enzyme activity of total leaf extract has been measured.

Scientific/commercial impact

Revealing the role, differential regulation and evolutionary changes of NAD-ME in C₃ and C₄ species will help to engineer a synthetic C₄ plant.

Project B10 Structural evolution of phosphoenolpyruvate carboxylase kinase (PPCK) in the genus *Flaveria*

Researcher

Johannes Schwabroh

Project leader

Georg Groth

Project type

Ph.D. project

Project start

01.11.2014

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Veronica G. Maurino

Lutz Schmitt

Kai Stühler

Peter Westhoff

Aim of the project

PEP carboxylase is a key enzyme in C₄ photosynthesis catalysing the carboxylation of PEP to oxaloacetate. Although the non-photosynthetic C₃ and the photosynthetic C₄ isoform catalyse the same reaction, they differ in their regulatory and kinetic characteristics. Activity of PEPC is regulated by phosphorylation, which is catalysed by protein kinase PPCK. In the genus *Flaveria* two PPCK isoforms have been identified, named PPCKA and PPCKB. To resolve the molecular basis of the preferential interaction of PPCKA with the photosynthetic C₄-PEPC and of PPCKB with the non-photosynthetic PEPC and to identify key residues in both kinases that determine this interaction we will apply X-ray crystal structure analysis and solve high-resolution structures of C₃- and C₄-typical PPCKA and PPCKB isozymes from the genus *Flaveria*.

Context within CEPLAS

Detailed knowledge of the molecular basis controlling specificity of PPCKA for C₄-PEPC will provide essential information on the regulation of the C₄-module.

First results

n.a. - Project start 01.11.2014

Scientific/commercial impact

Currently, no structural information on PPCK - even a low or medium resolution - is known. Structural information on the different PPCK isoforms will provide a better understanding of the regulation of a key enzyme of C₄ photosynthesis and of its corresponding isoform in C₃ plants.

Project B11 Nitrogen and Sulfur metabolism in C₄ plants

Aim of the project

The aim of the project is to find out the biological relevance of differential distribution of N and S metabolism in C₄ plants and how this distribution affects use efficiency of these nutrients.

Context within CEPLAS

This project connects research areas B and D, contributing the current research programme of RA B. It has a potential to solve long-standing questions of C₄ research, and significantly enhance the outputs of RA B.

First results

The project starts in January 2015. However, the transcriptomics data available in the groups of Peter Westhoff and Andreas Weber have already been used to obtain information about the expression of the key genes of S and N assimilation in *Flaveria* and *Cleome* species with different types of photosynthesis.

Scientific/commercial impact

The project supplements the efforts on engineering C₄ photosynthesis in crops by delivering information on essential metabolic pathways that are differently expressed in C₄ plants.

Researcher

Silke Weckopp

Project leader

Stanislav Kopriva

Project type

Ph.D. project

Project start

01.01.2015

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)
Peter Westhoff

Publications

Weckopp SC & Kopriva S (2014) Are changes in sulfate assimilation pathway needed for evolution of C₄ photosynthesis? *Frontiers in plant science* 5:773.

Project B12 Differentiation of vascular veins and surrounding mesenchyme: the role of leaf meristems within adaptations to C₄ anatomy

Aim of the project

In current CO₂-O₂ conditions, C₄ plants are better adapted than C₃ plants, which relates to Kranz anatomy and the subdivision of CO₂ fixation or photosynthesis to mesophyll and bundle sheath cells, respectively. In the project, we aim to investigate the spatial and temporal interplay between marginal or plate-meristem function in incipient leaf primordia and the establishment of vascular strands. We would like to understand whether the interface between stem-cell niches serving mesenchyme or vasculature relates to evolutionary adaptations from C₃ towards C₄ anatomy.

Context within CEPLAS

Stable and inducible driver lines will be established that allow cell-type-specific expression of transgenes in *Arabidopsis* or rice at earliest leaf developmental stages. Transgene selection will be based on the currently available data, mostly derived from comparative analyses of C₃-C₄ species pursued within CEPLAS.

First results

n.a. - Project start 01.11.2014

Scientific/commercial impact

The analysis of transgenic lines will reveal whether candidate genes identified in CEPLAS contribute to C₃/C₄ adaptations.

Researcher

Satish Kumar Eeda

Project leader

Wolfgang Werr

Project type

Ph.D. project

Project start

01.10.2014

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)
Peter Westhoff

3.3 Research Area C

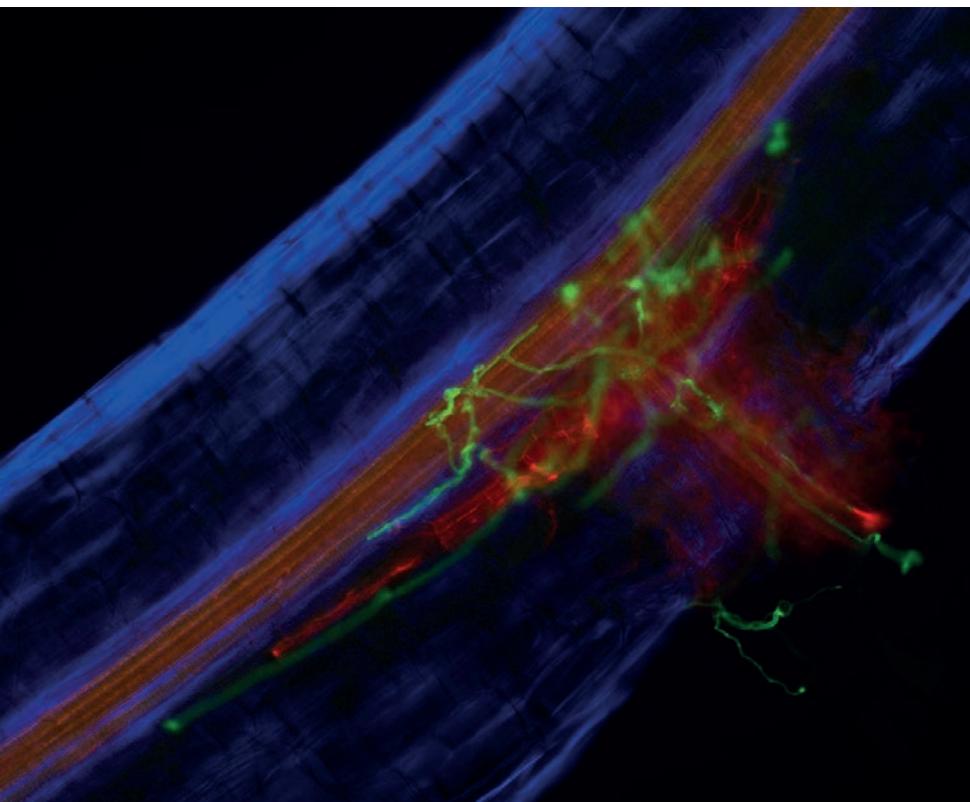
The molecular basis of plant-microflora interaction

Summary

Plants interact with a variety of microbes ranging from those that live outside but in close association with the plant to those that live inside as endophytes. The interaction between plants and their endophytic complement is complex and likely to involve mutual adaptation between the interacting partners. Associations with some endophytic microbial species are beneficial for plants and therefore these interactions can be exploited to enhance plant performance. However, the endophytic microflora remains largely unexplored. Recent technological advances (e.g., next generation sequencing, computational biology, metagenomics and culture-independent microbe detection) are enabling scientists to probe these interactions to a much higher definition than before and begin to understand the fundamental mechanisms shaping endophytic microbial populations and the consequences for the plant.

The aim of Research Area C is to characterise the structure, function and ecology of the plant microbiome within and between species of the Brassicaceae (including *Arabidopsis thaliana* and relatives). Projects in Research Area C are making progress in this exciting frontier of plant biology. Starting from the bacterial communities, the groups of *Paul Schulze-Lefert* (MPIPZ) and *Alice McHardy* (HHU) have teamed up to characterise the bacterial communities associated with relatives of *A. thaliana* (Schlaeppli et al., 2014) and extend these studies to the important crop, barley (Bulgarelli et al., 2015).

To what extent the endophytic bacterial community is affected by co-infection with pathogenic organisms (e.g., the oomycete *Albugo laibachii*) is the focus of a research team involving *Alice McHardy* (HHU) and *Eric Kemen* (MPIPZ). This study takes a metagenomic approach to determine how community structure is shaped by interactions with pathogens. A related project, spearheaded by *Michael Bonkowski* (UoC) and *Laura Rose* (HHU), evaluates which oomycete



▪ Coordinator

Laura Rose

▪ Co-coordinator

Jane Parker

▪ Faculty

Michael Bonkowski
Marcel Bucher
Gunther Döhlemann
Michael Feldbrügge
Martin Hülskamp
Eric Kemen
Karl Köhrer
Maarten Koornneef
Martin Lercher
Alice McHardy
Peter Nürnberg
Richard Reinhardt
Paul Schulze-Lefert
Jürgen Zeier
Alga Zuccaro

and protist species are closely associated with *A. thaliana* and close relatives. These studies have revealed a number of interesting eukaryotic members of the microbiota. Of immediate interest for functional studies is the isolation of members of the protist groups Vampyrellidae and Viridiraptoridae which typically feed on algae, nematodes or fungal spores and may have beneficial effects when cultured with plants. A further potential beneficial effect of predatory protozoa is their influence on complex biofilm formation. This is the focus on a new project by *Eric Kemen* (UoC). Meanwhile, the groups of *Marcel Bucher* (UoC) and *Paul Schulze-Lefert* (MPIPZ) are further characterising the structure of fungal communities associated with roots of *Arabidopsis thaliana* and *A. thaliana*, respectively.

The successful recruitment of *Alga Zuccaro* to UoC/CEPLAS brings along valuable expertise on a model fungal endophyte associated with *A. thaliana*, *Piriformospora indica*. One project she leads will characterise a novel class of effectors secreted by *P. indica*. Most 'effector biology' to date has focussed on well known plant pathogens. A key aim within Research Area C is ultimately to improve plant performance by harnessing microbes and endophyte-derived effectors might be very important components in establishing synthetic symbioses.

On a related front, the group of *Michael Feldbrügge* (HHU) seeks to understand the infection strategy and evolutionary history of a newly described smut, *Thecaphora thlaspeos*, of *Arabidopsis* species. This project addresses goals of Research Area C and also connects to Research Area D. In collaboration with *Eric Kemen* (MPI), the Feldbrügge group will determine how infection of plants by *T. thlaspeos* affects the microbiome community structure. This aims towards understanding how microbe communities associated with plants are assembled, and if and how they depend on few or numerous individual members. The second purpose of this study is a proof-of-concept. With the goal to generate synthetic symbioses for improving plant performance, a handful of existing, successful symbionts must be tested. Since the mode of action of a beneficial endophyte can range from direct effects (e.g., stimulating plant growth or providing micronutrients) to indirect effects (e.g., priming defence responses or modifying microbe community assembly), a prerequisite for developing synthetic symbioses is high infection success and ability to manipulate the symbiont. The Feldbrügge group is working to optimise this fungus for both purposes.

The mechanistic basis of beneficial associations with microbes and plant health is highly dependent on environmental conditions. Nutrient and light availability, temperature and genotype x environment (G x E) interactions likely influence defence trade-offs in the host. Several groups in Research Area C are investigating how abiotic factors affect endophytic associations with *A. thaliana*. *Jane Parker's* group (MPIPZ) is evaluating interactions between temperature, defense homeostasis and bacterial community composition. Her analysis has revealed a high degree of natural genetic variation in temperature effects on accumulation of the defence hormone salicylic acid, without necessarily adversely affecting plant growth and fitness. *Jürgen Zeier's* group (HHU) investigates how defence priming, an important but low-cost resistance mechanism, is affected by light, nitrogen and the C/N balance. *Martin Hülskamp* (UoC) is identifying genetic and environmental factors coordinating phosphate starvation and pathogen defence. In a related project, *Marcel Bucher* (UoC) investigates if and how phosphate starvation affects endophytic fungal community composition. Ultimately, through the collaborative efforts of this diverse group of plant biologists, we hope to harness the potential of endophytic communities to positively influence plant performance.

Project C1 Structural and functional characterisation of the *Arabidopsis thaliana* fungal microbiome

Researcher
Juliana Almaro

Project leader
Marcel Bucher

Project type
Postdoc project

Project start
01.03.2013

Cooperations
(other participating
CEPLAS and non-CEPLAS
researchers)
George Coupland
Alga Zuccaro
Stéphane Hacquard
(MPIPZ)

Aim of the project

We wish to know the core fungal microbiome of perennial *A. alpina*. Ecological genetics of these fungi will be studied and their ability to promote plant growth/fitness.

Context within CEPLAS

Impact of plant genotype on fungal community structure. Novel fungi to study biological and genetic nature of root-microflora interactions. Bioinformatics pipeline for fungal community analysis.

First results

ITS2 sequencing of fungal community validated. Fungal communities in ~100 samples from natural and greenhouse cultures currently being analysed. One isolate has capacity to increase plant biomass, colonise plant roots, transport ³³P to plant.

Scientific/commercial impact

Cologne collection of root-associated fungi (CORFU) and core microbiome enabling genetic studies to identify host genes involved in microbiome structure and function and evolutionary transitions from parasitism to mutualism. Core species to study intra- and cross-kingdom interactions in reconstructed microbiome. Plant growth-promoting fungi for commercial usage. Screening for new antibiotic substances.

Project C2 Bioinformatic analysis of endophytic flora in *A. thaliana* and *A. alpina*

Researcher
Yao Pan

Project leaders
Alice McHardy
Martin Lercher

Project type
Postdoc project

Project start
15.04.2013

Cooperations
(other participating
CEPLAS and non-CEPLAS
researchers)
Michael Feldbrügge
Vera Göhre
Eric Kemen
Zihong Ye (Jiliang University, China)

Aim of the project

Project 1

Genomic analysis of Thecaphora seminis-convolvuli

T. seminis-convolvuli is a fungal pathogen, which destroys seeds of the flowering plant *Calystegia*. Little is known about *T. seminis-convolvuli*. In this project, we will decipher the whole genome sequence of *T. seminis-convolvuli*, and will perform comparative genomic analysis. The aim of the project is to set up a pipeline of *Thecaphora seminis-convolvuli* genome annotation and to examine the evolutionary relationships between *T. seminis-convolvuli* and closely related species.

Project 2

Shotgun metagenomic analysis of epiphytic and endophytic microbial communities in Albugo laibachii infected A. thaliana leaves

Albugo laibachii is an oomycetes pathogen of the model plant *A. thaliana*. In this project, we applied shotgun metagenomic analysis to dissect the epiphytic and endophytic microbial community in *A. laibachii* infected *A. thaliana* leaves. The project aims at studying the core microbial community associated with species of *Albugo laibachii* and on learning how the microbial community interacts with *A. laibachii* during infection of *A. thaliana*.

Project 3

Genomic analysis of *Ustilago esculenta*

U. esculenta is a biotrophic basidiomycete fungus that infects the only host, *Zizania latifolia*. There is no molecular work published so far regarding its life style or pathogenicity. Here, we report the genomic analysis of *U. esculenta*. Our aim is to finish the genome annotation of *U. esculenta* as well as the genomic comparison analysis between *U. esculenta* and other three smut fungi (*U. maydis*, *U. hordei* and *Sporisorium reilianum*)

Context within CEPLAS

Help and assist biologists to analyse and obtain the information from data generating from high throughput sequencing methods.

First results

Project 1

Thecaphora seminis-convolvuli genome 22 Mb contains 5,372 predicted protein coding genes.

Project 2

Epiphytic and endophytic microbial communities on *Albugo laibachii* infected *A. thaliana* leaves showed distinct features on both diversity and functional profiles.

Project 3

U. esculenta genome of 20.2 Mb contains 6,655 predicted protein coding genes with high synteny to *U. maydis*, *S. reilianum* and *U. hordei*. We found RNA interference but no repeat-induced point mutations in the genome of *U. esculenta*, indicating that the fungus uses RNA silencing but not the RIP in defence. Four smut fungi genomic comparison also reveals the differences in repeated DNA, mating loci structures and secreted proteins.

Scientific/commercial impact

Project 1

Thecaphora seminis-convolvuli genome annotation is the first genome information on *Thecaphora* species.

Project 2

Identifying the core community of *Albugo laibachii* infected *A. thaliana* leaves is fundamental in understanding how diversity is generated in such ecosystems.

Project 3

U. esculenta genomic information helps to underlie the mechanism of the gall formation and microbe (*U. esculenta*) - plant (*Zizania latifolia*) interaction.

Project C3 Investigation of the effect of temperature-modulated defence homeostasis on plant-microbe symbioses in *Arabidopsis thaliana*

Researcher

Friederike Brüssow

Project leader

Jane Parker

Project type

Postdoc project

Project start

01.09.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Maarten Koornneef

Paul Schulze-Lefert

Julia Vorholt (ETH Zürich, Switzerland)

Aim of the project

Investigate *Arabidopsis* (*At*) natural variation in salicylic acid (SA) and defence pathway homeostasis in response to temperature (T) and how variation influences leaf-associated microbial communities.

Context within CEPLAS

Explore genetic and environmental factors shaping plant fitness and associated microbial communities

First results

We characterised total SA levels and biomass in 108 genetically diverse *At* accessions grown at 22°C and 16°C for five weeks. We revealed that total SA levels are regulated in a genotype x T-dependent manner. We performed association mapping of this trait on 95 *At* accessions and identified four significant loci underlying variation in this SA x T trait. Interestingly, low biomass was not significantly correlated with high SA levels implying plasticity in *At* for this trade-off. Using microbial culture-dependent and independent approaches, we are measuring differences in total numbers and structure of leaf-associated bacteria in six accessions displaying large SA x T variation, and in SA-response mutant lines. We use bacterial culture information from P. Schulze-Lefert and J. Vorholt.

Scientific/commercial impact

Improve efficiency of microbe use in agriculture.

Project C4 Characterisation of oomycete and protozoan species from tissues of *A. thaliana* and *A. alpina*

Researcher

Sebastian Ploch

Project leader

Laura Rose

Project type

Postdoc project

Project start

01.11.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Eric Kemen

Stéphane Hacquard (MPIPZ)

Aim of the project

The project aims to discover and characterise the impact of oomycetes and protists in the rhizo- and phyllosphere of *Arabidopsis thaliana* and *Arabis alpina*. Cercozoa had been chosen as the first protozoan group to be investigated.

Context within CEPLAS

Eukaryotic organisms are an important, but understudied, group of plant-associated organisms. These organisms may affect plant performance and could represent novel organisms to generate beneficial synthetic communities.

First results

A large number of cercozoan and oomycete species, closely associated with *A. thaliana*, were discovered, including many taxonomically undescribed groups. One cercozoan taxon could be found within plant tissue. This species will be used for further investigation.

Scientific/commercial impact

First insights into community composition of root- and leaf-associated oomycetes and cercozoa.

Project C5 Characterisation of fungal endophytes and basis of fungal-bacterial interactions in plants

Aim of the project

We aim to establish a smut infection system consisting of two genetically tractable partners by characterising the infection process of the endophytic smut fungus *Thecaphora thlaspeos* infecting model Brassicaceae and establishing tools for its molecular engineering.

Context within CEPLAS

We address the establishment of a synthetic symbiosis module, the central aim of Research Area C. In collaboration with E. Kemmen, we will study its influence on host microbiome formation and pathogens, which might lead to generation of a beneficial synthetic microcosm as a proof-of-principle.

In collaboration with Research Area D, we will devise strategies to engineer metabolic pathways for the plant benefit.

This fungus also allows understanding different infection strategies for annual and perennial hosts (Research Area A).

First results

T. thlaspeos grows systemically as an endophyte in *Arabidopsis* spp and in the model plant *A. thaliana*. Infection occurs during co-germination of seeds and spores, a pre-requisite for use in agricultural applications.

Haploid cultures of opposite mating type have been established, which will be used for genetic manipulation. The genome was sequenced and is currently being annotated.

Scientific/commercial impact

The host range of *T. thlaspeos* might extend to crop plants of the Brassicaceae family such as *Brassica napus* (seeds from NPZ, Hohenlieth). Benefit for plant growth or protection hence might be directly translated to agricultural applications.

Researchers

Janpeter Stock
Thorsten Langner
Benedikt Steuten

Project leader

Michael Feldbrügge

Project type

Ph.D. project

Project start

01.11.2013

Cooperations

(other participating
CEPLAS and non-CEPLAS
researchers)

Vera Göhre
Lamprinos Frantzeskakis
Kaitlyn Courville

Project C6 Environmental influences on pipecolic acid biosynthesis, defence priming, and systemic acquired resistance: light, nitrogen supply, and the C/N balance

Researcher

Ziba Ajami-Rashidi

Project leader

Jürgen Zeier

Project type

Ph.D. project

Project start

01.10.2013

Cooperations

(other participating
CEPLAS and non-CEPLAS
researchers)

Shizue Matsubara (FZJ)

Aim of the project

In this project, we investigate how *Arabidopsis thaliana* adapts its immune responses to different environmental conditions such as light (quality and quantity) and nutrient supply.

Context within CEPLAS

A fundamental aim of CEPLAS is to understand how plants adapt to environmental conditions and constraints. This project investigates how plants adapt the systemic acquired resistance (SAR) and immune-related priming responses to defined light and nutrient environments.

First results

Recently, we have identified the Lys catabolite pipecolic acid (Pip) as a novel metabolic regulator of SAR and defence priming associated with SAR (Navarova et al., Plant Cell 2012).

In the current project, we found that Pip biosynthesis and Pip-mediated defence priming responses are positively affected by light availability and the length of a day.

Further, we compared plant responses exposed to continuous light and fluctuating light simulating dynamically changing light environments that occur in nature. Whereas fluctuating light alone slightly stimulates expression of several SAR marker genes, inducible SAR responses such as salicylic acid accumulation is quantitatively lower under fluctuating than under continuous light.

Finally, our results show that microbe-induced Pip accumulation and the magnitude of SAR are compromised in plants suffering from limited nitrogen supply.

Scientific/commercial impact

CEPLAS aims to contribute to the development of strategies ensuring increased plant productivity in the future. A better understanding of the mechanisms and robustness of SAR and defence priming will fuel these strategies because the priming phenomenon activates plant immunity and, in parallel, minimises growth constraints that would be imposed by constitutive defence activation.

Project C7 Role of WRK75 in phosphate, pathogen and temperature responses

Aim of the project

WRKY75 is known to be involved in phosphate starvation and pathogen responses. The project aims to understand its role in these processes and possible coordination.

Context within CEPLAS

Research Area C aims to understand the interaction between the plant and the microbiome and also phosphate responses by identifying the relevant genes. WRKY75 is a bonafide coordinating gene enabling us to perform the more detailed analysis.

First results

RNAseq data are being analysed, constructs are transformed into *Arabidopsis*.

Scientific/commercial impact

Possible impact lies in the identification of the factors possibly coordinating phosphate starvation and pathogen responses.

Researcher

Heike Wolff

Project leader

Martin Hülskamp

Project type

Ph.D. project

Project start

01.11.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Marcel Bucher

Ute Höcker

Kenichi Tsuda (MPIPZ)

Project C8 Shaping of the *Arabis alpina* microbiome by plant host interactions with environmental factors - phosphorous limitation

Aim of the project

Identification of loci and genes affecting root-associated fungal community composition/function. Re-constructed fungal microbiome in *Arabis alpina* x *A. montbretiana* recombinant inbred lines will be studied at different P conditions.

Context within CEPLAS

Understanding the mechanistic basis underlying the network of plants with functionally and phylogenetically diverse fungi is essential for targeted approaches towards designing optimised microbiomes adapted to host plants and changing environments.

First results

To reduce stochastic effects, tools necessary to reliably measure fungal community shifts as response to environmental cues are being established, i.e., a synthetic root-associated fungal community with indexed species, and a cheap and rapid detection method (quantitative ARISA) to screen the composition of re-constructed fungal communities.

Scientific/commercial impact

Develop approaches to optimise plant-microflora interactions in crops exhibiting efficient defence against their enemies and establishing beneficial symbioses to improve plant performance under environmental stress.

Researcher

Michael Thielen

Project leader

Marcel Bucher

Project type

Ph.D. project

Project start

01.05.2014

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

George Coupland

Gunther Döhlemann

Christiane Kiefer

Project C9 Identification of plant growth promoting genes and pathways of bacterial symbionts in the *A. thaliana* rhizosphere

Researcher

Ruben Garrido Oter

Project leaders

Alice McHardy

Paul Schulze-Lefert

Project type

Ph.D. project

Project start

01.10.2013

Cooperations

(other participating
CEPLAS and non-CEPLAS
researchers)

Davide Bulgarelli (Uni-
versity of Dundee, UK)

Richard J. O'Connell
(INRA Versailles,
France)

Publications

Bulgarelli D, Garrido-
Oter R, Münch Phi-
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chom.2015.01.011

Schlaeppli K, Domb-
rowski N, Oter RG et
al. (2014) *Proceedings
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United States of Ameri-
ca* 111(2):585-592.

Aim of the project

The aim of the project is the development of new computational methods for analysis of high-throughput genomic and phenotypic data for the study of root-associated microbial communities in *A. thaliana* and barley.

The project focuses on the analysis of 16S rRNA marker genes, whole genome sequences of isolated exemplars, as well as shotgun metagenome data in order to unravel the complex relationship between these microbial communities and their host.

Context within CEPLAS

The aim of Research Area C is the characterisation of the structure, function and ecology of the plant microbiome. Our on-going studies have the potential to providing us key insights into the mechanisms underlying plant-microbe community associations at the root-soil interface.

First results

We employed a combination of 16S rRNA gene profiling and shotgun metagenome analysis to investigate the structural and functional diversification of the microbiota associated with wild and domesticated accessions of barley.

Our analyses revealed a small but significant genotype effect on the root-associated communities and allowed us to identify us functional categories enriched in the rhizosphere and protein families under positive selection.

Scientific/commercial impact

Our analyses of the barley root-associated microbial communities provide evidence that the combined activities of microbe-microbe and host-microbe interactions ultimately shape the plant root microbiota.

We believe our findings will also have implications for other research groups investigating the relationship between an eukaryotic host and its associated microbiota.

Project C10 Role of fungal WSC lectin-like proteins in the interaction of endophytic fungi with plant roots

Aim of the project

We are aiming to understand the molecular mechanisms underlying the beneficial effects of the root colonising fungal endophyte *Piriformospora indica*. The project focuses on the role of lectin-like proteins, which are expanded in the genome of *P. indica*.

Context within CEPLAS

In Research Area C we are investigating the influence of microorganisms on plants' metabolism and fitness. *P. indica* displays an ideal model system because of its beneficial effects and its broad host range.

First results

We identified a plant responsive WSC-lectin, which binds to β -1,3-glucan and is able to

- i) inhibit the sporulation of pathogenic fungi and
- ii) has an impact on the plant innate immunity.

Scientific/commercial impact

This work will contribute to our knowledge on plant-fungus interactions and in plant defence mechanisms and it might lead to novel strategies for the control of pathogenic fungi and the production of resistant plants.

Researcher

Philip Fesel

Project leader

Alga Zuccaro

Project type

Ph.D. project

Project start

01.09.2014

Project C11 Mutualistic interactions of pathogenic and non-pathogenic protists in foliar biofilms

Aim of the project

Sharing burdens during host colonisation through biofilm formation likely enhances pathogen fitness. Aims of this project are therefore to investigate collaborative advantages of foliar protozoan biofilms for host colonisation, their interactions and signalling networks within biofilms, as well as their impact on the bacterial leaf microbiota.

Context within CEPLAS

Plant associated microbial communities are complex networks that can significantly influence plant health. This project aims on combining diversity studies of predatory protozoa and their impact on complex biofilm formation. Mechanistic insights into microbe-microbe interactions will help to resolve functional aspects of complex traits that help to define leaf microbiota, capable of rendering plants more resistant to pathogen attack under field condition.

First results

We have started to isolate protozoa from leaves.

Scientific/commercial impact

These findings will significantly contribute to higher plant performance and therefore to develop methods for sustainable crop productions in future.

Researcher

Alfredo Mari

Project leader

Eric Kemen

Project type

Ph.D. project

Project start

01.11.2014

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Michael Bonkowski

Paul Schulze-Lefert

3.4 Research Area D

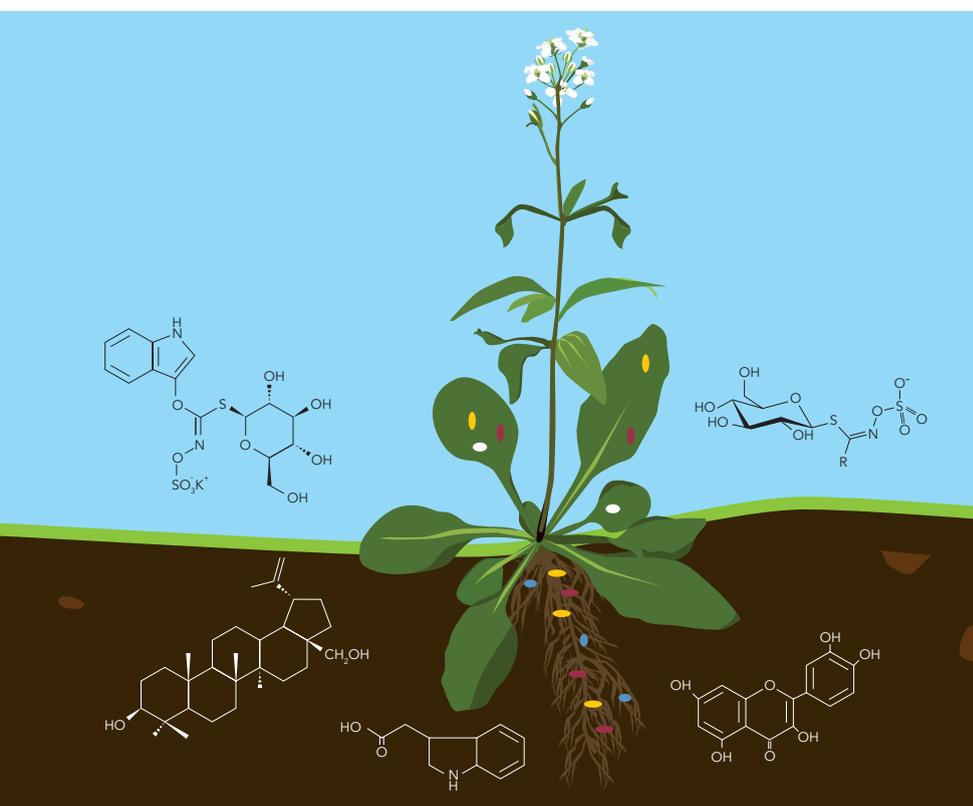
Plant metabolism: from biotic challenges to synthetic biology

Summary

Plants produce a great variety of biochemically unexplored secondary metabolites, which derive from only a few primary metabolites. Environmental stressors shape the chemical diversity of plant secondary metabolites that could have both beneficial and/or detrimental effects on plants.

We aim at the identification and functional analysis of plant secondary metabolites which are determinative in the interaction of plants with the root endophyte community (co-operation with Research Area C). So far, we have identified the role of MYB and bHLH transcription factors in the biosynthesis of various secondary compounds. The multiple *myb* and *bhlh* mutants have been further used to characterise the role of flavonoids, indolic phytoalexins, and aliphatic and indolic glucosinolates in plant-microbe interactions. The development of predictive mathematical models simulating secondary metabolite biosynthetic pathways is in progress.

In addition, a next generation sequencing approach has been applied to characterise the root microbial community of *Arabidopsis thaliana* mutants. First results revealed the importance of indolic phytoalexins in shaping the root-microbiome and in deterring the growth of cellulolytic bacteria. These indolic metabolites were also identified in a metabolic survey for microbe inducible plant secondary metabolites. We determined regulatory principles of their induced biosynthesis, and investigated their role in the establishment of systemic acquired resistance (SAR).



▪ Coordinator

Ulf-Ingo Flügge

▪ Co-coordinator

Karl-Erich Jaeger

▪ Faculty

Ilka Axmann

Michael Bonkowski

Oliver Ebenhöf

Thomas Drepper

Tamara Gigolashvili

Stanislav Kopriva

Martin Lercher

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Lutz Schmitt

Vlada Urlacher

Andreas P. M. Weber

Jürgen Zeier

We additionally aim to identify and characterise further components of regulatory modules controlling the biosynthesis and transport of secondary metabolites in roots and in the rhizosphere. This approach intends to understand reciprocal metabolic signalling in plant-microbe interactions. Plant growth promoting rhizobacteria (PGPR) (identified by Research Area C) use so far largely uncovered metabolic pathways to stimulate growth in Arabidopsis. We therefore developed a method for metabolite profiling of roots and root exudates using UPLC-qTOF MS. Studies of bacterial signal molecules revealed that adipic acid, which was identified by pyrolysis-field ionization mass spectrometry, most probably functions as an exudate compound in root-bacteria communication.

Exuded secondary metabolites serve as important communication signals between plant roots and their surrounding rhizosphere. Specifically in response to nutrition shortage, e.g., sulfur deficiency, the microbiome can be modified by root exudates. We observed strong differences of bacterial sulfatase activity in the rhizosphere of several Arabidopsis ecotypes which will be used in a genome-wide association study (GWAS) to identify new regulatory compounds and modules.

The production of specific secondary metabolites is a prerequisite for exudation, but the engineering of complete pathways requires transport of the secondary metabolites from the cell and finally into the rhizosphere. We expressed several root-specific ABC-transporters in *Pichia pastoris* which will now be used together with collected root exudates for transportomics to tag the transporter-specific substrates.

Our ultimate goal is to transfer identified biosynthetic pathways having positive effects on plant growth first into microbial hosts and further into model plants and/or endophytes for the production of phytochemicals and to investigate molecular interactions and underlying mechanisms. Recently, we succeeded in the production of 2,3-oxidosqualene in the phototrophic purple bacterium *Rhodobacter capsulatus*. This compound serves as the precursor for a great variety of triterpenes which can subsequently be analysed for bioactivity including putative PGPR activity.

The identification and the molecular assembly of compatible genetic elements in “synthetic microbes” and “synthetic plants” will have a significant impact on developing next generation agricultural products and may also lead to biotechnological and pharmaceutical innovations.

Project D1 Evolutionary pattern of secondary metabolites in roots and root exudates of *Arabidopsis thaliana* and its relatives grown under defined conditions

Researcher

Magdalene Kutyniok

Project leader

Ulf-Ingo Flügge

Project type

Postdoc project

Project start

01.11.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Michael Bonkowski

Stanislav Kopriva

Paul Schulze-Lefert

Katharina Sklorz

Aim of the project

Plants use specialised metabolites to interact with the environment such as root-associated bacteria. Plant growth promoting rhizobacteria (PGPR) can assist plants in dealing with unfavourable biotic and abiotic conditions. Plant-derived metabolites are important drivers of these interactions. Our aim is to understand metabolic interdependencies existing in plant-bacteria interactions and to identify crucial (secondary) compounds of this interaction that are synthesised in response to specific bacteria or are used by bacteria as a cue for the association with a plant.

Context within CEPLAS

Knowledge about the involvement of plant biosynthetic pathways and specific metabolites in the communication between plants and PGPR are the prerequisite for the mechanistic understanding of plant-PGPR interactions and for the development of strategies for improved plant-bacteria interactions and better plant performance.

First results

Several PGPR from the *Arabidopsis thaliana* root microbiome provided by Research Area C were tested for their growth promotion capacity under different conditions e.g., in hydroponic systems (co-operation with Michael Bonkowski). A method for metabolite profiling of roots and root exudates with UPLC-qTOF MS was developed in co-operation with the MS platform at the Cologne Biocenter. Root exudates were collected from plants grown under different nutritional conditions with/without the presence of PGPR and metabolite profiling will be performed as a next step to pinpoint important (secondary) metabolites for the plant-PGPR interactions.

Project D2 Plant secondary metabolites in plant-bacteria-interaction and improved plant nutrition

Researcher

Linda Reinhardt

Project leaders

Ulf-Ingo Flügge

Stanislav Kopriva

Project type

Ph.D. project

Project start

01.10.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Henning Frerigmann

Tamara Gigolashvili

Christina Lucas

Sabine Metzger

Aim of the project

Plants are themselves not able to access soil organic-sulfur sources but rely on soil microbes to mineralise sulfur, an essential macronutrient for plants. We analyse how plants communicate with beneficial soil bacteria under sulfur starvation to improve their nutritional status. Further, we study the impact of glucosinolates, sulfur-containing secondary metabolites, on specific microbial functions.

Context within CEPLAS

The identification of specific plant metabolites as signalling components in plant-bacteria interactions is essential for improved plant nutrition.

First results

Bacterial strains capable of mineralising organo-sulfur sources and inducing root growth in *A. thaliana* have been isolated. Root extracts of inoculated plants grown under sulfur starvation are collected for untargeted metabolite profiling. A precise glucosinolate pattern in shoots and roots of *A. thaliana* accessions has been determined and its impact on soil bacterial activity is now being analysed.

Project D3 Natural variation in the interaction of plants and bacteria in the rhizosphere

Aim of the project

Natural variation in *Arabidopsis* accessions will be used to identify genes that affect the interaction of plants and bacteria in the rhizosphere. We determine the enzymatic reaction of sulfatase in sulfur-limiting rhizosphere as a measure of how the different ecotypes shape the bacterial community. The data will be used for genome-wide association study to identify genes underlying this variation. These genes will be used in synthetic approaches to create plants better capable to shape their microbiome and to underpin other projects of RA D. In addition, these experiments address the contribution of bacteria to plant sulfur nutrition.

Context within CEPLAS

Genes involved in synthesis of secondary plant metabolites will be prioritised for analysis in order to help selection of suitable compounds, enzymes, and transporters analysed in other projects of Research Area D.

First results

Using the MPI Cologne soil as inoculum we observed at least fivefold difference in the sulfatase activity in the rhizosphere of individual accession. The data from ca. 120 accessions show normal distribution and suitability for GWAS study, when the whole set of 360 accessions is analysed.

Researcher

Dr. Anna Koprivova

Project leader

Stanislav Kopriva

Project type

Core project
Kopriva group

Project start

01.07.2014

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)
Irene Klinkhammer
Dr. Achim Schmalenberger (University of Limerick)

Project D4 The impact of secondary metabolites on the composition of the endophytic microflora in *Arabidopsis thaliana*

Aim of the project

Secondary metabolites are important features of plants to interact with their biotic environment. The biosynthesis of glucosinolates is well studied in the model system *A. thaliana* and can therefore serve as a tool to elucidate the complex biosynthetic network controlling production of secondary metabolites. This project thus contributes to a better understanding of the regulatory network of secondary metabolites in plant-microbe interactions.

Context within CEPLAS

Knowledge on the structure and function of regulatory networks controlling the biosynthesis of secondary metabolites will serve as a promising target to generate synthetic plants with improved abilities in plant-microbe interactions.

First results

The role of different MYB transcription factors could be characterised as important regulators of glucosinolate biosynthesis upon biotic and abiotic challenges (Frerigmann & Gigolashvili, 2014 a,b). We also identified several interaction partners of MYBs belonging to the group of bHLH transcription factors. bHLH proteins are crucial to control specific secondary metabolites upon jasmonate signalling (Frerigmann et al., 2014).

Publikations

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Researcher

Henning Frerigmann

Project leader

Ulf-Ingo Flügge

Project type

Postdoc project

Project start

01.01.2013

Cooperations (other participating CEPLAS and non-CEPLAS researchers)
Veronica G. Maurino
Paul Schulze-Lefert
Elia Stahl
Miltos Tsiantis
Alga Zuccaro
Pawel Bednarek (Polish Academy of Sciences)
Erich Glawischnig (TU München)

Project D5 Interactions of microbial endophytes with the glucosinolate metabolism in plants

Researcher

Katharina Sklorz

Project leader

Michael Bonkowski

Project type

Ph.D. project

Project start

01.03.2013

Cooperations

(other participating
CEPLAS and non-CEPLAS
researchers)

Ulf-Ingo Flügge

Henning Frerigmann

Tamara Gigolashvili

Christina Lucas

Sabine Metzger

Aim of the project

The project investigates the interdependency of *A. thaliana* with root-associated microorganisms (e.g., bacteria, protozoa) at the metabolic level. The aim is to identify how microbial signal molecules affect root growth and secondary metabolism of Brassicaceae, and how the ability of plants to synthesise secondary metabolites can influence microbial endophytes.

Context within CEPLAS

Understanding the role of microbial signals on plant performance and fitness.

First results

Bacterial signal molecules (C₄-AHLs) had profound effects on root architecture of *A. thaliana*. It also influenced exudate profiles. By pyrolysis-field ionization mass spectrometry, we identified an exudate compound with a m/z ratio of 172, indicating a role of adipic acid in root-bacteria communication.

Scientific/commercial impact

Manipulation of root growth by microbes is central in plant-microbe interactions, since it regulates nutrient and water uptake capacity of plant hosts. Plant metabolites play important roles in these interactions as both, signals and defense compounds.

Project D6 Mathematical modelling of glucosinolate metabolism in plants

Researcher

Suraj Sharma

Project leader

Oliver Ebenhöh

Project type

Ph.D. project

Project start

01.10.2014

Cooperations

(other participating
CEPLAS and non-CEPLAS
researchers)

Tamara Gigolashvili

Stanislav Kopriva

Aim of the project

Develop mathematical models of the glucosinolate biosynthesis pathway and employ the model to understand what controls the composition of glucosinolate in terms of chain length, side chain modifications, etc. The model will be used to understand which mechanisms are of key importance for these regulations.

Context within CEPLAS

Glucosinolates play an important role in defense against pathogens. It is therefore of paramount importance to understand (i) how a plant regulates the synthesis of particular glucosinolates to alter the specific composition and (ii) how the glucosinolate composition affects the pathogens.

First results

We have constructed a simplified prototype model, which takes into account the fact that many enzymes are not highly substrate-specific but actually accept as substrates chain-elongated amino acids where the exact number of introduced carbon atoms is not specified. Preliminary results indicate that the chain-length distribution is highly non-trivial.

Scientific/commercial impact

It is envisaged that the basic understanding we produce will have impacts on the field of secondary metabolism in general. Further, understanding how pathogens trigger glucosinolate production and how this affects pathogen defense is obviously of fundamental interest for plant breeders.

Project D7 Synthetic microbes for the production of plant secondary metabolites

Aim of the project

This project aims to develop synthetic modules for the production of plant triterpenes (TTP) in microbes. TTPs have important roles for various plant-microbe interactions, however, the underlying mechanisms are largely unknown. Hence, we focus on synthesis, modification and functional screening of TTP aiming to analyse physiological effects together with partners from Research Areas C and D.

Context within CEPLAS

- (1) Detailed characterisation of TTP pathways;
- (2) establishing TTP production in microbes for a broad screen of functions (e.g., plant growth promotion, Research Area D);
- (3) transfer of modules into plant-associated bacteria (Research Area C) to establish improved plant-microbe interactions.

First results

A TTP precursor module has been constructed and analysed in different microbes. We succeeded for the first time to establish this synthetic module in the carotinogenic bacterium *Rhodobacter capsulatus* allowing the efficient two-step catalytic conversion of the carotenoid precursor FPP into 2,3-oxidosqualene from which all terpenes derive and which serves as substrate for oxidosqualene cyclases (OSCs) producing a huge variety of cyclisation products, including marneral, thalianol and β -amyrin. Currently, we construct and evaluate a TTP synthesis module encompassing all known TTP cyclases from *A. thaliana*. In addition, a promiscuous OSC will be introduced that simultaneously produce >20 different triterpenes, potentially in combination with a promiscuous P450. The resulting huge variety of triterpenes with potentially novel biochemical structures and activities will be analysed for their bioactivity (i) in human or murine cell lines containing bioactivity-responsive cis-elements fused to reporter constructs and (ii) in growth promotion assays. Compounds with highest biological activities will be studied in more detail in co-cultivation assays of plants and triterpene-producing bacteria.

Scientific/commercial impact

The characterisation of TTP functions can provide a basis to develop novel strategies to improve plant growth as well as resistance against biotic and abiotic stresses.

Researcher

Anita Loeschcke

Project leaders

Karl-Erich Jaeger

Thomas Drepper

Project type

Postdoc project

Project start

01.01.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Ilka Axmann

Marcel Bucher

Ulf-Ingo Flügge

Tamara Gigolashvili

Stanislav Kopriva

Lutz Schmitt

Vlada Urlacher

Stéphane Hacquard

(MPIPZ)

Publications

Kaschner M, Loeschcke A, Krause J et al. (2014) *Molecular microbiology* 93(5): 1066-1078.

Liebl W, Angelov A, Juergensen J et al. (2014) *Applied microbiology and biotechnology* 98: 8099-8109.

Project D8 Programming triterpene biosynthesis pathways in cyanobacteria using synthetic RNA-based devices

Aim of the project

This project aims at establishing extended sRNA-based comparator devices for self-adjustment and balancing of engineered triterpene biosynthetic pathways in *Synechocystis* 6803.

Context within CEPLAS

- (i) Establishing engineered biosynthesis of plant triterpenes in cyanobacteria finally targeting plant-growth promotion;
- (ii) Development of novel synthetic regulatory (RNA-based) modules for optimised microbial triterpene biosynthesis, transferable to other microbes (including plant-associated bacteria).

Researcher

Dennis Dienst

Project leader

Ilka Maria Axmann

Project type

Postdoc project

Project start

01.08.2014

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Thomas Drepper

Vlada Urlacher

Pia Lindberg (Uppsala University, Sweden)

Sven Findeiß (University of Vienna, Austria)

André Estévez-Torres (CNRS Marcoussis, France)

First results

Conjugable expression plasmids that allow for trace metal-inducible production of marneral, thalianol and β -amyrin were successfully constructed to be transferred to a squalene accumulating mutant strain of *Synechocystis* 6803, kindly provided by P. Lindberg. A robust fluorescence-based reporter assay for functional analysis of synthetic RNA-based devices has been established: a self-replicative, conjugable plasmid harbouring cassettes for alternating Co^{2+} - and Ni^{2+} -inducible expression of mVenus (YFP) and mCerulean (CFP), respectively.

Scientific/commercial impact

(I) Enhancing the host spectrum with an oxygenic phototroph should help to elucidate potential bottlenecks for microbial triterpene production.

(II) Implementation of synthetic RNA devices in cyanobacteria will expand the field of synthetic biology and improve the toolbox for valuable metabolic engineering strategies.

Project D9 Functional expression and biochemical characterisation of plant P450s

Researcher

Sarah Kranz

Project leader

Vlada Urlacher

Project type

Ph.D. project

Project start

01.04.2013

Cooperations

(other participating CEPLAS and non-CEPLAS researchers)

Ilka Axmann

Marcel Bucher

Thomas Drepper

Ulf-Ingo Flügge

Tamara Gigolashvili

Karl-Erich Jaeger

Stanislav Kopriva

Lutz Schmitt

Aim of the project

The overall aim of the project is to develop synthetic modules for the production of plant triterpenes and their oxygenated derivatives in microbes. To achieve this goal selected plant cytochrome P450 monooxygenases will be produced and characterised regarding their substrate specificities. Furthermore, P450 enzymes will be combined in e.g., *E. coli* with various plant terpenoid synthases in order to increase the diversity of triterpenoids with useful properties.

Context within CEPLAS

- (i) Understanding the biosynthesis and the regulation of triterpenoids,
- (ii) Improved performance of plants modified in triterpene biosynthesis and signalling. The long-term objective is to transfer synthetic modules into plant-associated bacteria (Research Area C) for improved plant-microbe interactions.

First results

Heterologous expression of CYP71A16 and CYP705A12 from marneral pathway, thalianol hydroxylating CYP708A2, cytochrome P450 reductase and marneral and thalianol synthases from *A. thaliana* in *E. coli* was completed. Various constructs allowing functional coexpression of P450s with synthases and/or reductase were established. A P450 mutant library for β -amyrin oxidation was expressed.

Scientific/commercial impact

Synthetic microorganisms will be constructed producing higher amounts of plant secondary metabolites with properties beneficial for plant development and growth and/or of pharmaceutical relevance.

Project D10 *In vitro* analysis of selected ABC transporters from plants

Aim of the project

Identification of the substrate spectrum of ABC transporters.

Context within CEPLAS

ABC transporters play a key role in the sequestration of secondary metabolites and exudation of these metabolites into the plant rhizosphere. However, their role in plant-microbe interaction and knowledge on the transport substrate(s) is rather limited.

First results

After overcoming unexpected problems in cloning of the cDNA of plant ABC transporters of the PDR family, we have cloned and expressed the root-specific transporters PDR2, PDR8 and ABCG2 in *Pichia pastoris*. Solubilisation and purification trials were promising and detergents for solubilisation have been identified. The purification protocols will be optimised and an *in vitro* characterisation will lead to the identification of their substrate(s).

Scientific/commercial impact

The *in vitro* set-up will allow the identification of the transport substrate(s) and insights into the mechanism of transport.

Researchers

Katharina Gräfe
Kalpana Shanmugarajah

Project leader

Lutz Schmitt

Project type

Ph.D. project

Project start

01.06.2013

Project D11 Regulatory and evolutionary aspects of stress-inducible plant metabolic pathways

Aim of the project

This project searches for novel metabolites synthesised in plants upon interaction with pathogenic and non-pathogenic microbes, investigates their biological function, and dissects the regulatory networks of inducible metabolite biosynthesis.

Context within CEPLAS

Research Area D aims at investigating the regulatory principles of plant secondary metabolism and the role of small metabolites in plant-microbe interactions. This project centrally contributes to these aims by focussing on the regulation and function of stress-inducible metabolic pathways in *Arabidopsis*.

First results

We have identified several indolic metabolites that accumulate upon activation of systemic acquired resistance in distal leaves of plants locally inoculated with bacterial pathogens, determined regulatory principles of their biosynthesis, and investigated their role in SAR establishment. Further, we have detected stress-inducible production of the three lipid-soluble antioxidants beta, gamma and delta-tocopherol and have determined their regulation patterns of accumulation. Our data reveal a novel function for tocopherols in *Arabidopsis* basal resistance to bacterial pathogens and highlight their capacity to prevent lipid peroxidation in plant cells upon bacterial infection.

Scientific/commercial impact

The identification and functional characterisation of immune-regulatory small metabolites holds great potential for the development of novel plant protection strategies.

Researcher

Elia Stahl

Project leader

Jürgen Zeier

Project type

Ph.D. project

Project start

01.04.2013

Cooperations (other participating CEPLAS and non-CEPLAS researchers)
Ulf-Ingo Flügge
Henning Frerigmann

3.5 Publications

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4.

Plant
Metabolism
and
Metabolomics
Laboratories



Core facilities

The chemical nature of metabolites, small organic molecules, is very diverse and more than 200.000 small molecules are estimated to play a biological role in the organisms of the plant kingdom (Pichersky & Lewinsohn, 2011). The CEPLAS Plant Metabolism and Metabolomics Laboratories provide expertise and instrumentation for the qualitative and quantitative analysis of plant metabolism and for the integration of metabolomics data with other data domains, such as transcriptomics and proteomics data. Metabolite analysis provides quantitative and qualitative phenotypic information at the level of metabolism in analyses of mutant lines or experiments under perturbed environmental conditions.

Our core facilities for metabolic analyses are open to all CEPLAS members as well as external collaborators. We offer a wide range of services for the identification and quantification of metabolites from diverse biological material.

The aim of the laboratories is to

- do routine measurements of key metabolites of the primary (e.g., organic acids, amino acids, sugars) and secondary (e.g., glucosinolates, flavonoids) plant metabolism.
- establish new methods for the extraction and subsequent analysis of primary and secondary metabolites together with our collaborators (mainly via liquid and gas chromatographic separation methods that are hyphenated with mass spectrometry detection)

One of the most crucial aspects of metabolic analysis is appropriate experimental design and sampling technique. Thus, the core facilities interact with CEPLAS groups already at the level of experimental design and sample preparation in order to obtain a high level of analytical correctness. Methods of measurements are optimised and validated together with the researchers. For data processing and interpretation, we provide adequate software tools, or, if demanded, we process the data directly and pass them on to our collaborators. For large-scale experiments, we are therefore able to offer the possibility to analyse samples in automated, high-throughput processes. Moreover, a more detailed analysis involving identification and structure analysis of unknown components via high-resolution MS and MS/MS is also possible with our instrumentation.

Thanks to our specialised laboratories, the optimal analytical instrument can be chosen for each scientific question, whereas a single lab normally cannot provide this large choice of instruments. Additionally, our personell has profound experience and gets regular external and internal training to ensure cutting-edge work-flows.

The main instruments of our facility include six spectrometres that are used depending on the respective experiment:

At the HHU

- Our GC-MS device is a 7200 accurate mass GC-QTOF from Agilent Technologies. We routinely quantify 50 metabolites, mainly of the central carbon metabolism in various species ranging from photosynthetic organisms such as *Arabidopsis*, *Flaveria*, *Cleome*, *Zea mays*, different water plants, red algae and cyanobacteria to yeast, as well as the sea slug *Elysia chlorotica*.
- With the 1290 UHPLC system coupled to the triple-quadrupole 6490 (UHPLC-MS/MS), both from Agilent Technologies, we aim to quantify metabolites of central metabolism, which are not detectable or distinguishable by GC-MS such as phosphorylated sugars (e.g., intermediates of the Calvin-Benson cycle) and certain isomers (e.g., citrate and isocitrate).
- Elemental analysis is performed by gas source stable isotope ratio mass spectrometry (EA-IRMS). With our Vario ISOTOPE cube coupled to the IsoPrime100 mass spectrometer, we analyse C and N content, C to N ratio and carbon isotope ratio ($\delta^{13}\text{C}$). $\delta^{13}\text{C}$ of plant material is an important parameter to distinguish different pathways of carbon fixation such as C_3 , and C_4 photosynthesis.

- Moreover, we analyse a bigger set of amino acids than quantifiable by GC-MS by ultra-high performance liquid chromatography coupled to DAD detection (UHPLC-DAD) with the 1290 UHPLC system (Agilent Technologies).

At UoC

- Quantification of metabolites is performed with the AB Sciex QTRAP 5500 coupled to an Agilent 1260 HPLC (LC-QTRAP). Using MRM, we routinely quantify more than 20 flavonoid aglycones in parallel in different genera such as *Arabidopsis* and *Lotus*.
- The high resolution QTOF mass spectrometer maxis4G (Bruker) we mainly use for the identification of unknown compounds. This MS can be equipped with different ion sources to analyse polar (API) and apolar (APCI, GC) compounds and to analyse compounds in very low concentration (nano ESI offline).
- Multi-element analysis is performed with an Agilent ICP-MS 7700.

Examples for measurements within the last year

In a project with *Juliette de Meaux* (UoC), we compared the metabolite levels in *Arabidopsis thaliana*, *A. lyrata* and *A. halleri* after desiccation to elucidate their different susceptibility to drought. Our GC-MS metabolite dataset is currently integrated with RNAseq data.

Submerged aquatic plant species can cope with CO₂ limitation by using bicarbonate as additional carbon source (carbon concentrating mechanisms, CCM). However, the underlying mechanisms are largely unknown. In a project of *Dr. Andreas Hussner* (HHU), we analysed metabolites, C and N-ratios in ten submerged aquatic plant species with different capacity to use HCO₃⁻ under low and high HCO₃⁻ conditions. We found interesting differences helping to increase our understanding of the different CCM mechanisms in these ecologically important species.

Ute Höcker's group (UoC) is interested in understanding the cross talk between environmental signals in the biosynthesis of anthocyanins. As a source of natural genetic variation, her lab uses *Arabidopsis* accessions and available recombinant inbred populations. Our mass spectrometry laboratory established a protocol to identify various anthocyanins, anthocyanidins and anthocyanidin precursors in small amounts of tissues (collaboration of the groups *Bucher*, *Hülkamp* and *Höcker*). Currently, we are employing this method to analyse the natural genetic variation in *Arabidopsis* with respect to light- and sucrose-induced anthocyanin biosynthesis.

In a project with the group of *Ulf-Ingo Flügge* (UoC), we analysed the glucosinolate pattern and compare different root and shoot tissue using different accessions and mutant lines of *Arabidopsis thaliana*. In a second project, we currently establish an untargeted LC-MS method for the analysis of plant secondary metabolites in roots and root exudates which are important for plant-microbe interaction under sulfur-deficient conditions.

▪ Heads

Dr. Tabea Mettler-Altman (HHU)
Dr. Sabine Metzger (UoC)

▪ Technical/scientific support

Maria Graf (HHU)
Elisabeth Klemp (HHU)
Katrin Weber (HHU)
Christina Lucas (UoC)

▪ Instruments

GC-MS (Agilent Technologies)
UHPLC-MS/MS (Agilent Technologies)
EA-IRMS (Isoprime)
UHPLC-DAD (Agilent Technologies)
LC-QTRAP (AB Sciex)
QTOF (Bruker Daltonics)
ICP-MS (Agilent Technologies)

5.

Bachelor Programme in Quantitative Biology

Summary

Apart from cutting-edge science, the aim of CEPLAS is to develop new programmes for early career researchers, from the undergraduate to the postdoctoral level. In recent years, technological progress and the resulting introduction of many new, high-throughput methods have strongly changed the requirements that biologists have to meet. Therefore, CEPLAS has initiated the establishment of a novel undergraduate study programme that responds to this challenge.

Structure and content of the study programme

The foundation is laid during the first four semesters that are taught jointly with the regular study programme for biologists. This initial phase mediates mainly knowledge and skills in biology and other natural sciences. The second phase represents the core of this programme. Here, specifically composed courses in computational biology, biostatistics and mathematical modelling will be taught, together with physical biology of the cell and systems biology. A final module during the 7th semester is dedicated to synthetic biology. 40% of the courses will be theoretical and as much as 60% practical to efficiently combine the acquirement of quantitative-analytical skills with a biological background. The third phase is called research phase and comprises different electives, where the students may train the acquired skills according to their specific needs. In this phase there is also time for internships abroad.

We keep up our long-term goal to create a full four-year Bachelor Programme in Quantitative Biology. This programme will be implemented once currently open professorships, e.g., in Quantitative Genetics and Synthetic Biology, will have been filled within the next years.

Admission and recruitment

Students will be able to apply for admission to the programme of Quantitative Biology after completing all modules of the third semester of regular Bachelor programmes in Biology or Biochemistry. Admission to the programme will be based on their current academic achievements, especially in mathematics. We will compare these achievements in form of ECTS grades to ensure equal opportunities for applicants from other universities within and outside of Germany.

According to a student survey in summer semester 2014, we estimate to receive enough applications from students of both universities to fill 10 to 15 places already in 2015.



- **Responsible PI at HHU**

Peter Westhoff

- **Responsible PI at UoC**

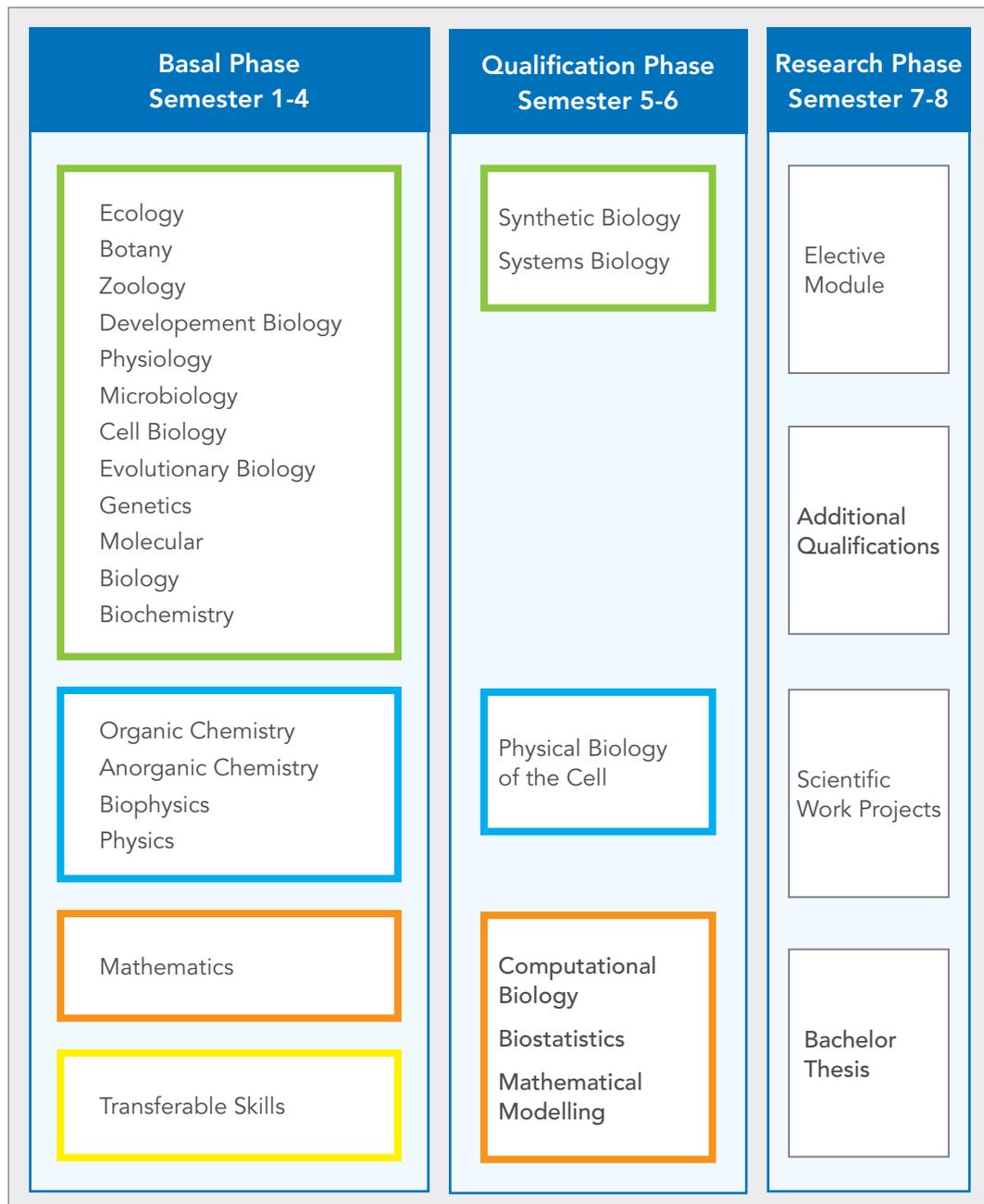
Stanislav Kopriva

- **Coordinator**

Veiko Krauß

Time schedule

Faculty and Rectorate of the Heinrich Heine University institutionally support the programme in Quantitative Biology as a novel variant of the Bachelor of Science Biology. Concurrently, an agreement of cooperation between the two universities is in preparation. Documents for accreditation of the programme were submitted in February 2015. At the beginning of the summer semester 2015, promotional events will be held to solicit applications for the scheduled start of the programme in winter semester 2015/2016. Web pages providing German and English information about the programme including application forms are already online. Advertising flyers and posters are in preparation.



We originally planned to start the programme with the winter semester 2014/15. However, consulting all the various decision bodies at the two Universities and setting up the legal framework took much more time than anticipated. The curriculum will be reviewed for accreditation in June 2015, and the programme will start with the winter semester 2015/16.

6.

Young researchers



6.1 CEPLAS Graduate School

Brief summary

The CEPLAS Graduate School is dedicated to provide Ph.D. candidates with excellent training for a future career in the field of modern plant sciences in academia as well as in industry. To this end, the structured three-year Ph.D. programme is jointly organised by all four participating institutions and includes three major areas: the doctoral research project, the scientific, as well as the career training programme. The **research project** is the foundation of the Ph.D. study and aims to qualify CEPLAS doctoral researchers for independent and professional scientific work. Because CEPLAS research projects are highly collaborative, Ph.D. students benefit from the combined expertise of molecular biologists, biochemists and theoretical biologists.

The **scientific training programme** includes scientific courses as well as symposia and seminars that allow doctoral researchers to broaden their scientific knowledge.

Career-relevant transferable skills to further train doctoral researchers for their future career are part of the **career training programme**. This includes workshops and seminars, excursions to industry and the CEPLAS Career Day.

Development of the Graduate School

Currently the CEPLAS Graduate School has 28 members including a first associated doctoral researcher who joined the programme in September 2014.

The most recent 8 Ph.D. projects started in October 2014 and February 2015. All positions were internationally advertised and 12 international and national candidates were invited for a selection workshop in June 2014, where applicants were selected based on short presentations about their previous research and personal interviews by three CEPLAS PIs each.



- **Speaker Graduate School**
Ute Höcker
- **Coordinator Graduate School**
Esther Jawurek
(*Petra Fackendahl* during maternity leave)
- **Doctoral Representative**
Thomas Wrobel
Katharina Sklorz (deputy)
- **Current number of doctoral researchers**
28
- **Number of males/females**
64% Female
- **Internationality**
36% international doctoral researchers from six countries: Italy, China, Poland, Iran, India and Spain

Training measures

Scientific training programme

Apart from the compulsory course *Good Scientific Practice*, the Graduate School curriculum includes a minimum of three additional scientific courses. In 2014 the Graduate School organised four courses (*Advanced Methods for Fluorescence Imaging*, *Natural Variation*, *RNA Sequencing for Beginners* and *Mass Spectrometry in Protein Analysis*) that were attended by more than 20 of our Ph.D. students.

Since the training programme is organised in collaboration with other local graduate schools in plant science (International Max Planck Research School, IMPRS and the International Graduate School for Plant Science, iGRAD *plant*) and the umbrella graduate schools of the participating universities (Graduate School for Biological Sciences, GSfBS and Interdisciplinary Graduate and Research Academy, iGRAD) free spots in CEPLAS courses were offered to iGRAD *plant* and IMPRS students. In turn, five CEPLAS Graduate School members attended a course in *Bioinformatics* via the GSfBS in Cologne. Individual students also took courses offered by other graduate programmes, external workshops or summer schools, including the *Marie Curie Initial Training Network AccliPhot (Environmental Acclimation of Photosynthesis)* or a *Summer School at the Isaac Newton Institute for Mathematical Sciences*. Funding for external courses is generally available via the CEPLAS Graduate School and gladly supported if it tailors the student's individual needs. For 2015, courses in *Biostatistics*, *Applied Plant Genetics* and in *Mass Spectrometry in Metabolite Analysis* are scheduled.

Career training programme

The career training programme includes four mandatory courses in career-relevant transferable skills. The courses *Scientific Presentation* and *Scientific Writing* (in collaboration with iGRAD) are compulsory, whereas the remaining topics can be chosen from the iGRAD and GSfBS course programmes. In 2014 several students joined the following iGRAD courses: *Time Management*, *Fundamentals of Project Management*, *Stimme und Körpersprache [Language and body language]* and *Negotiation Training*.

Apart from the annual *Women's Career Day* (see promotion of gender equality), the CEPLAS Graduate School uses the close cooperation with other graduate schools to offer as many career talks to Ph.D. students as possible. For instance, *Stephen M. Schrader* from DuPont Pioneer, USA was invited in October 2014 (in collaboration with iGRAD *plant*, Düsseldorf) to talk about his 10-year career in plant biotechnology industry.

Internal Evaluation

To be able to adapt the programme of our Graduate School even better to the needs and requirements of our students, our programme is internally evaluated on a yearly basis.

90% of our doctoral researchers participated in the survey and stated to be overall very satisfied with the CEPLAS Graduate School and the scientific supervision of their research projects. Also the scientific and career training programmes were generally rated very positively. Furthermore, most Graduate School members highly value the possibility of networking and collaborating with other researchers with diverse focuses and expertise within the cluster.

One aspect that should be improved, however, was that doctoral researchers would like to intensify their contact to industry. With the help of *Günter Strittmatter* (formerly Head of Research and Development at KWS SAAT AG) who recently joined CEPLAS as a consultant the CEPLAS Graduate School will therefore concentrate on organising more career events next year, such as excursions to industry and career talks / discussions. The next excursion to industry will be to Qiagen, Hilden in early 2015.

6.2 CEPLAS Postdoc Programme

Brief summary

The CEPLAS Postdoc Programme aims at providing our postdocs with the best possible skills, expertise and network to allow them to achieve their personal career goals. The components of an Individual Development Plan (IDP) shall help our postdocs to optimally prepare for their future career in or outside academia. Supervision and mentoring are core components of the IDP, which are complemented by offering a wide range of training courses and opportunities to get in contact with industry. Active participation at CEPLAS events, like the annual CEPLAS Symposium and Young Researchers Retreat and the monthly CEPLAS Friday and Young Researchers Seminar as well as the Research Area meetings integrate the postdocs into the CEPLAS network, broaden their scientific focus and train presentation skills.

Development of the programme

Currently 18 postdocs are members of the CEPLAS Postdoc Programme. In addition to the postdoc positions that started in 2013, four additional postdocs were associated to the postdoc programme in 2014. Associated postdocs are not directly funded by CEPLAS but work on CEPLAS related topics and participate in the programme. One postdoc left the programme already after 1 year as she got an attractive job offer from a company.

A new cohort of postdocs will begin in 2015. The research areas agreed on a total of 14 new postdoc projects that are financed by the cluster. The positions were internationally advertised and candidates will be invited for a selection workshop in spring 2015.

Training measures

The participants of the Postdoc Programme are offered a wide range of courses to further develop their skills and competencies. Participation is on a voluntary basis and postdocs can choose the courses that fit to their needs. Basically workshops offered at all four institutions are open for CEPLAS postdocs, e.g., *Leadership Skills*, *Negotiation Training*, *Optimising Writing Strategies*, *Safety in Gene Technology* (according to §15 GenTSV). The CEPLAS homepage provides an overview about a selection of current course offers. In addition, depending on the postdocs' requirements, courses are also organised exclusively for CEPLAS postdocs and funding for external courses is generally available upon request.

According to the wishes of the postdocs, the following courses were organised 2014 exclusively for CEPLAS postdocs:

- **Scientific training** *Statistics Short Courses*, Trainers: Prof. Dr. Holger Schwender, Martin Schäfer, Julia Schiffer (CBiBs Center for Bioinformatics and Biostatistics at HHU), part 1: March 5-7 2014, part 2: April 3, 4 and 11 2014
- **Career training** *Find your perfect job – Career development for postdocs*, Trainers: Dr. Christian Dumpitak and Dr. Debbie Radtke (iGRAD), Marcus Holzheimer (MH³ Beratung), August 25 2014. The majority of the participants took the opportunity of individual consultations that is offered as follow up.

In total 17 postdocs participated in these workshops. In 2015 a workshop in Grant Writing and one on Social Networking are in planning. Furthermore, we would like to offer more scientific courses and therefore the postdocs were encouraged to propose potential topics.

Mentoring

All postdocs participating in the programme chose a mentor with whom they meet about twice a year during their time in CEPLAS. As the feedback shows, most postdocs really benefit from this compulsory component of the programme.



- "It broadened my perspective on scientific careers ..."*
- "good career advice, honest comments on career development"*
- "help to plan next steps"*
- "be more focused"*
- "Wrote a grant application, my mentor suggested it and encouraged me to go for it"*
- "I got and keep contact to another not-CEPLAS-involved institution"*
- "It gave me an external point of view"*
- "We had good talks about career perspectives and alternative careers"*
- "My mentor and me found institutes I could apply at and grants I could apply for"*

Networking with industry

At this year's excursion to industry, CEPLAS postdocs visited the Department of Plant Systems Biology at the VIB (Vlaams Instituut voor Biotechnologie) and the companies BASF CropDesign and Syngenta at the Technology Park in Ghent, Belgium. The group of postdocs that was accompanied by Rüdiger Simon and Günter Strittmatter, received information about the research organisations and activities and got an insight into their facilities. The postdocs also presented the central research topics of CEPLAS, and open discussions have shown mutual interests for future collaborations. The overall feedback of the participants was very positive, 43% rated the event as "excellent", 50% as "good". Apart from getting in contact with industry at the excursions, postdocs are offered assistance in finding cooperation partners for joint research projects, assistance in organising internships at companies, and assistance with finding a mentor from industry.

Evaluation of the Postdoc Programme

Same as for the Graduate School our Postdoc Programme is annually evaluated to adapt the programme even better to the needs and requirements of our postdocs. The response rate was 78%. The members of the programme were satisfied with the organisation of the programme and the training courses offered. Most of them are also satisfied with the opportunities for networking and getting in contact with industry. However, the latter leaves room for improvement. The overall satisfaction with how the programme prepares for the future career was reasonably good (21% very satisfied, 50% rather satisfied). In the future, excursions to industry will be jointly organised by the Graduate School and Postdoc Programme for being able to offer a broader choice of interesting companies. Furthermore, postdocs are offered assistance in finding a cooperation partner for joint research projects and organising internships at companies. A point of critique was that many Postdocs are rather dissatisfied with the scientific supervision.

▪ Speaker Postdoc Programme	Rüdiger Simon
▪ Coordinator Postdoc Programme	Juliane Schmid
▪ Postdoc Representative	Wilma van Esse, Nadine Höcker (deputy)
▪ Current number of participating postdocs	18
▪ Number of males/females	67% Female
▪ Internationality	44% international postdocs from seven countries: China, Columbia, France, Netherlands, Palestine, Switzerland, USA

6.3 Young researchers activities

Participants of the CEPLAS Graduate School and the CEPLAS Postdoc Programme continued joint activities that were initiated in 2013 and started new activities in 2014.

Young Researchers Seminar

Within the Young Researchers Seminars, doctoral and postdoctoral researchers meet among themselves and present their results in an informal manner. The seminar is a forum for discussions and scientific exchange.

Besides presentations, young researchers have introduced flip-chart sessions as a new format: Three to four persons explain their project, problems and pitfalls or new experiments they want to set up by using a flip-chart only. Experience so far is that this format supports feedback in an interactive way and leads to fruitful discussions. Young researchers profit from the interactive feedback and the challenge to explain core points in a few sentences. Moreover, they get to know associated topics and challenges as well as their peers, which stimulates collaborations between them.

The Young Researchers Seminar is held on a monthly basis before CEPLAS Fridays at which all CEPLAS members meet. Young researchers have lunch together before joining the CEPLAS Friday. The seminar is jointly organised by the postdoc and doctoral researcher representatives.

Young Researchers Retreat

The yearly retreat of the CEPLAS Graduate School and Postdoc Programme took place at the end of September (29th-30th 2014) at the Physikzentrum in Bad Honnef. 43 young researchers and 28 faculty members participated at the retreat.

Besides progress reports that were given by the young researchers in the form of presentations or posters, the retreat included an interactive session with the young researchers only. In groups, young researchers worked out the objectives and benefits of the research areas and the cluster in general. The goal of this task was to foster young researchers to extend their knowledge about other research areas, face up to the overall objectives of the cluster and to think about the role of their projects in achieving the cluster's goals.

In a second activity young researchers prepared non-scientific project descriptions for lay audiences in order to train communication of their own research project in simple words to the general public.



The young researchers' feedback regarding this session was very mixed (from excellent to fail), however the comments showed that some profited from the session:

"I gained a holistic view of CEPLAS. Active engagement was very useful to help retain info."

"I got to know some of the people outside my research area better, that was a definite plus."

"I really liked the interactive morning session on day 1. That was a very good idea. I think that was especially helpful for new CEPLAS members and increases the group/network dynamics."



Young Researchers Retreat 2014, Bad Honnef, Germany



Group work at the Young Researchers Retreat 2014



Career Development Workshop, August 2014

Planter's Punch

In July 2014 young researchers started a monthly feature named 'Planter's Punch', which is presented on the front page of the CEPLAS web site. Each month, a young researcher presents his/her research topic in a brief statement directed towards the public.

Outlook

In 2015 we plan to offer two excursions to industry for the CEPLAS young researchers. Joint organisation shall provide our young researchers with a wider choice of interesting companies. In addition, young researchers are offered assistance in organising internships at companies and in finding an industrial cooperation partner for joint research projects.

A joint course on social networking platforms is planned for 2015. Furthermore, postdocs are highly encouraged to contribute to the training of the CEPLAS Graduate School.



7.

Promotion of gender equality

Brief summary

Though the proportion of excellent female scientists is relatively high, women are still under-represented especially in leading positions. Whereas more than 50% of the doctoral degrees are awarded to female scientists, only approximately 20% of the faculty positions are filled with women (German Research Foundation, DFG, final report equal opportunity standards, 2013). The two main reasons for the severe dropout rate after the Ph.D. phase are the lack of female role models in a male-dominated and competitive environment and the difficulties of balancing family and a research career.

Therefore, the CEPLAS equal opportunity programme pursues two major approaches to increase the proportion of women in leading positions:

- the career support of female scientists during their Ph.D., postdoctoral and group leader phases and
- the improvement of the work-family balance for CEPLAS scientists with children.

Increasing the proportion of female scientists

CEPLAS is committed to actively recruit female scientists during all career stages. Currently 64% of the doctoral researchers and 64% of the postdoctoral researchers are female. Out of eight new faculty positions four were filled with women. Altogether 29% of all PIs and AIs are female.

Career and Family Support

Within the CEPLAS career and family support various measures have been implemented to promote women in science and to make work more family-friendly for female and male CEPLAS scientists.

Career Workshops

CEPLAS female researchers were encouraged to attend career workshops offered in close cooperation with the Equal Opportunity Offices of HHU and UoC. CEPLAS financially supported participation of doctoral and postdoctoral researchers as well as group leaders in several courses at the Female Career Center of UoC.



Invited speakers at the 2nd Women's Career Day

- *Dr. Milena Ouzunova*, Head of Biotechnology Maize and Oilseed Crops, KWS Saat AG Einbeck, Germany
- *Jun.-Prof. Dr. Maria von Korff Schmising*, Institute for Plant Genetics, HHU
- *Dr. Lydumyla Malysheva-Otto*; CEO IT-Breeding GmbH, Gatersleben, Germany
- *Dr. Susanne Brink*, Trends Group Manager and Editor at Trends in Plant Science, Elsevier Ltd., London, UK

Women's Career Day 2014

For the second time the CEPLAS Women's Career Day took place at the Cologne Biocenter on June 16th, 2014. Similar to last year's event the morning session included talks from four female guest speakers from academia, industry and publishing. During the afternoon session 22 female doctoral and postdoctoral researchers had a chance to personally discuss scientific career issues and the compatibility of work and family with the four speakers in smaller groups. A highly positive evaluation by the participants confirmed that the CEPLAS Women's Career Day has been established as a valuable event attracting researchers from CEPLAS and non-CEPLAS laboratories. Especially the round-table coffee break with personal discussions was very much appreciated.

Helping hands programme

For pregnant women, parents on maternity/parental leave and CEPLAS fellows (female and male) with young children we provide funding for student helpers in order to relieve them from routine tasks at work and/or ensure continued operation of their research projects during leave. In 2014 six student helpers were employed via the helping hands programme for three to six months. Funding will continue in 2015. In addition one Postdoc position was funded for six months to support a female Junior Professor during her time of parental leave. This ensured continued operation of research and supervision of doctoral researchers and undergraduate students in the research group. This funding will be continued for as long as the junior professor works part-time (until mid 2015).

Child Care during CEPLAS events

First initiated in 2013, child care service was very positively perceived by the parents and has become a regular feature at CEPLAS events depending on the parents' demand. In collaboration with the University's Family Support Services on-site child care service is offered for CEPLAS Symposia, CEPLAS Fridays, General Assemblies and other events.

Parent-child room

At the University of Cologne, CEPLAS supported an already existing parent-child room, located at the Institute for Biochemistry. In 2013 this room was equipped with an additional desk including a desktop computer. In Düsseldorf we successfully established a nursing room near the biology department in collaboration with the Faculty of Mathematics and Natural Sciences at the end of 2014. Both rooms are equipped with child care needs (changing table, baby bed, armchair, small play area with toys) and a work area (desk and chair, internet connection; a printer and desktop computer are available in Cologne) allowing parents to work close to their institutes and look after their children at the same time.

Travel grants for female scientists

To enable young scientists to communicate their research to an international community, CEPLAS appeared as a sponsor for the 2014 EMBO-ESF Conference "Biology of Plastids - Towards a Blueprint for Synthetic Organelles" (Poland, 21-26 June, 2014). On that account CEPLAS provided travel grants for two international female scientists from Australia and Portugal.

8.

Staff



Principal investigators

1	<i>Jun.-Prof. Dr. Maria Albani</i>	Cologne Biocenter, UoC at MPIPZ
2	<i>Jun.-Prof. Dr. Ilka Axmann</i>	Institute of Synthetic Microbiology, HHU
3	<i>Prof. Dr. Marcel Bucher</i>	Cologne Biocenter, UoC
4	<i>Prof. Dr. George Coupland</i>	MPIPZ
5	<i>Prof. Dr. Gunther Döhlemann (since 05/2014)</i>	Cologne Biocenter, UoC
6	<i>Jun.-Prof. Dr. Oliver Ebenhöf</i>	Institute of Quantitative and Theoretical Biology, HHU
7	<i>Prof. Dr. Michael Feldbrügge</i>	Institute of Microbiology, HHU
8	<i>Prof. Dr. Ulf-Ingo Flügge</i>	Cologne Biocenter, UoC
9	<i>Prof. Dr. Ute Höcker</i>	Cologne Biocenter, UoC
10	<i>Prof. Dr. Martin Hülskamp</i>	Cologne Biocenter, UoC
11	<i>Prof. Dr. Karl-Erich Jaeger</i>	Institute of Molecular Enzyme Technology, HHU
12	<i>Prof. Dr. Markus Kollmann</i>	Institute of Mathematical Modelling of Biological Systems, HHU
13	<i>Prof. Dr. Maarten Koornneef</i>	MPIPZ
14	<i>Prof. Dr. Stanislav Kopriva</i>	Cologne Biocenter, UoC
15	<i>Prof. Dr. Martin Lercher</i>	Institute of Informatics, HHU
16	<i>Prof. Dr. Alice McHardy</i>	Institute of Informatics, HHU
17	<i>Prof. Dr. Jane Parker</i>	MPIPZ
18	<i>Prof. Dr. Laura Rose</i>	Institute of Population Genetics, HHU
19	<i>Prof. Dr. Lutz Schmitt</i>	Institute of Biochemistry, HHU
20	<i>Prof. Dr. Paul Schulze-Lefert</i>	MPIPZ
21	<i>Prof. Dr. Ulrich Schurr</i>	Institute of Bio and Geosciences-2 (IBG-2), FZJ
22	<i>Prof. Dr. Rüdiger Simon</i>	Institute of Developmental Genetics, HHU
23	<i>Prof. Dr. Miltos Tsiantis</i>	MPIPZ
24	<i>Jun.-Prof. Dr. Maria von Korff Schmising</i>	Institute for Plant Genetics, HHU at MPIPZ
25	<i>Prof. Dr. Andreas P.M. Weber</i>	Institute of Plant Biochemistry, HHU
26	<i>Prof. Dr. Peter Westhoff</i>	Institute of Plant Molecular and Developmental Biology, HHU
27	<i>Prof. Dr. Jürgen Zeier</i>	Institute of Plant Molecular Ecophysiology, HHU
28	<i>Prof. Dr. Alga Zuccaro (since 02/2014)</i>	Cologne Biocenter, UoC

Participating researchers

1	<i>Prof. Dr. Petra Bauer</i>	Institute of Botany, HHU
2	<i>Prof. Dr. Michael Bonkowski</i>	Cologne Biocenter, UoC
3	<i>Prof. Dr. Juliette de Meaux (since 09/2014)</i>	Cologne Biocenter, UoC
4	<i>Dr. Thomas Drepper (since 01/2014)</i>	Institute of Molecular Enzyme Technology, HHU
5	<i>Dr. Tamara Gigolashvili</i>	Cologne Biocenter, UoC
6	<i>Prof. Dr. Georg Groth</i>	Institute of Biochemical Plant Physiology, HHU
7	<i>Dr. Eric Kemen</i>	MPIPZ
8	<i>Prof. Dr. Karl Köhrer</i>	Centre for Biological and Medical Research (BMFZ), HHU
9	<i>PD Dr. Veronica Graciela Maurino</i>	Institute of Plant Molecular and Developmental Biology, HHU
10	<i>Prof. Dr. Peter Nürnberg</i>	Cologne Center for Genomics (CCG), UoC
11	<i>Prof. Dr. Uwe Rascher</i>	IBG-2, Plant Sciences, FZJ
12	<i>Dr. Richard Reinhardt</i>	Max Planck Genome Centre, MPIPZ
13	<i>Prof. Dr. Kai Stühler</i>	Centre for Biological and Medical Research (BMFZ), HHU
14	<i>Prof. Dr. Klaus Theres</i>	MPIPZ
15	<i>Prof. Dr. Vlada B. Urlacher</i>	Institute of Biochemistry, HHU
16	<i>Prof. Dr. Wolfgang Werr</i>	Cologne Biocenter, UoC

Overview

Total overview of personnel that was financed in 2014 by the cluster or by the universities (as additional support for the Graduate School).

Category	male	female	total
W3	2	-	2
W1 and W2	1	4	5
Postdocs	10	18	28*
Platform Coordination	-	2	2
Ph.D. positions (cluster- and university-funded)	17	18	35**
Technical/administrative staff	3	13	16
Student and research assistance	23	24	47
Guest scientists	2	3	5
Cluster administration	1	5	6
Total	59	87	146

* 18 active members of the Postdoc Programme

** 28 active members of the Graduate School

New faculty

In 2013 we announced nine new faculty positions (4 W4 and 5 W1/W2) at the universities Cologne and Düsseldorf. Till the end of that year we succeeded to fill five of the announced positions.

Within 2014 we managed to fill another two positions with *Gunther Döhlemann* and *Alga Zucaro* at the University of Cologne.

In October 2014 *Markus Pauly* (UC Berkeley) accepted the call for the W3 position in Plant Cell Biology and Biotechnology at HHU. Planned starting date is July 2015. For the remaining open W3 position in Plant Quantitative Genetics and Genomics negotiations are ongoing.



Gunther Döhlemann

Position W3 Terrestrial Microbiology

Start May 2014

Our research aims to identify and understand molecular mechanisms of microbe-plant interactions. Focus is on secreted virulence factors (effectors) of biotrophic microbes, which suppress host immunity and plant metabolism. As model organisms, we are using the basidiomycete fungi *Ustilago maydis* and *Ustilago hordei*, which parasitise their respective host plants maize and barley to cause smut disease.

Specific research projects in the lab address:

- i) Suppression of apoplastic immunity by *Ustilago* effectors,
- ii) Organ specificity of the *Ustilago maydis* – maize interaction, and
- iii) Establishing of the *Ustilago hordei* – barley system to study biotrophy-factors of obligate biotrophs.

Within CEPLAS, we are particularly interested to study the role of cysteine proteases in plant immunity and how they are modulated by microbial effectors, particularly in root-colonising endophytes.

Short CV

Since 05/2014	Professor (W3) for Terrestrial Microbiology, CEPLAS, University of Cologne
05/2012	Habilitation in Genetics at the Biology Department, Philipps University Marburg on the topic „Etablierung der kompatiblen Interaktion des Brandpilzes <i>Ustilago maydis</i> mit seiner Wirtspflanze Mais“
Since 06/2011	Research Group Leader (independent PI) at the Max Planck Institute for Terrestrial Microbiology, Marburg
03/2009-06/2009	Research stay at Stanford University (Walbot Lab, Dept. of Biology) supported by an EMBO short term fellowship
01/2009-05/2014	Group Leader at the MPI for Terrestrial Microbiology, Marburg



Alga Zuccaro

Position W2 Microbial Ecological Genetics

Start February 2014

Currently my research is focused on the mechanisms that enable symbiotic fungi to colonise plants successfully and on the processes accounting for variations in host preferences and fungal lifestyles, especially in mutualistic root endophytes. With respect to insights into how symbiotic fungi establish themselves in metabolically active root cells and how the plants are reprogrammed for enhanced performance, in my group we routinely use integrated approaches that rely on the combination of reverse genetics, transcriptomics, cell biology, biochemistry, and comparative genomics. We propose to analyse the genetics and cell biology of the root endophyte *Piriformospora indica*, and the closely related orchid mycorrhizal fungus *Sebacina vermifera* in their symbioses with the model plants barley (monocot) and Arabidopsis (dicot) and to understand the evolutionary mechanisms involved in the establishment of biotrophy.

Primary interest is to find answers to the following questions:

1. Which mechanisms allow root endophytes to suppress host defense in a wide range of unrelated plants? Does *P. indica* and *S. vermifera* use effector-like molecules in the form of small secreted proteins in order to manipulate different host plants and/or are there other mechanisms like shielding itself in place?
2. Which signals mediate the switch from the biotrophic to the cell death associated phase? And is this latter phase actively triggered by the fungus?
3. What are the basic events involved in the transition from saprotrophic to symbiotic lifestyles?

Short CV

Since 10/2014	Professor at the University of Cologne (tenured W2), CEPLAS, Department of Genetics
02/2014-09/2014	Professor at the University of Cologne (W1), CEPLAS, Department of Genetics
Since 04/2010	Call for a W2/W3 professorship at the LMU Munich Research Group Leader at the Department of Organismic Interactions, Max Planck Institute for Terrestrial Microbiology, Marburg
07/2007-03/2010	Group Leader at the Institute of Phytopathology and Applied Zoology, Justus Liebig University of Gießen
05/2005-09/2005	Postdoctoral research project (AFTOL assembling the fungal tree of life) at the Department of Botany and Plant Pathology, Oregon State University (OSU), USA
06/2004-06/2007	Postdoc and staff member at the Institute of Microbiology, Technical University of Braunschweig
08/2001-05/2004	Doctorate at the Institute of Microbiology, Technical University of Braunschweig
10/1994-03/2000	Study of marine biology at the University of Ancona, Italy



9.

Finances

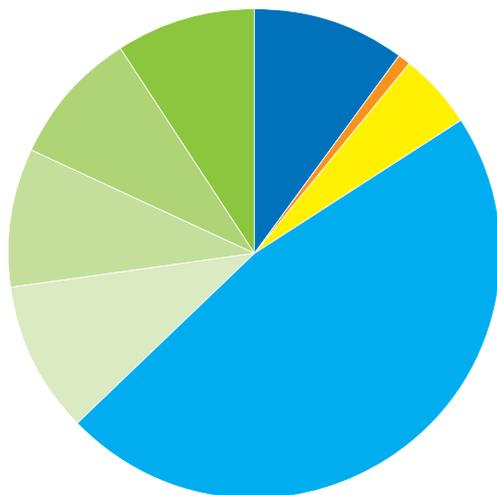
Granted funds and total amount spent

For the year 2014 CEPLAS was granted a total funding amount of € 7.4 million. Due to the delayed hiring of the two W3 faculty positions, we decided to shift a part of our budget to 2015 to end up with a final funding amount of € 5.255.700 for 2014.

Total amount of funds spent in 2014 (k€)

New total allowance 2014	Actual amount spent (provisional calculations 02/2015)
5,256	5,186

Overview total funds spent in 2014 (% of total budget)



New faculty	47
Research Area A	10
Research Area B	9
Research Area C	9
Research Area D	9
Central funds	10
Gender measures	1
Core facilities	5

Overview research areas (k€)

	Amount spent (02/2015)	% of total budget	Additional funds by universities
Research Area A	511	10	2 Ph.D. positions
Research Area B	489	9	4 Ph.D. positions
Research Area C	463	9	4 Ph.D. positions
Research Area D	469	9	–
Total	1,933	37	10 Ph.D. positions

Overview central funds (k€)

	Amount spent (02/2015)	% of total budget
New faculty	2,413	47
Administrative staff	299	6
Guest scientists	6	<1
Workshops Postdocs/ Doctoral researchers	4	<1
Symposia/retreats	47	1
Publications	19	<1
Travel costs	4	<1
Mobility fund	27	1
Recruiting	43	1
Gender	76	1
Public outreach	64	1
Mass spectrometry platform	250	5
Total	3,252	63

Third-party funding

Additional third-party funding raised in 2014 (k€)

Funding body	Number	Funding amount
German Research Foundation (DFG)		
Research grants	4	733
International Research Training Groups (subprojects)	1	156
Collaborative Research Centres (subprojects)	2	579
Priority Programmes (subprojects)	6	1,437
Others	1	4
Federal Ministry of Education and Research	1	212
Ministry of Innovation, Science, Research and Technology of North Rhine Westphalia	8	1,087
EU	1	312
Others	2	380
Total	26	4,900

10.

Technology transfer and cooperation management



Objective

A major goal of the CEPLAS programme is to provide a basis for the transfer of research results into industrial application and to establish mutually beneficial cooperations with external partners from academia as well as from industry. This is deduced from the conviction that research results should also generate economic value, that cooperative projects can create synergistic effects in using the expertise of the partners, and that young scientists can widen their perspectives and chances in career planning in such cooperations.

Therefore, CEPLAS has initiated such collaborations with partners from academia and industry, already from its beginning. To intensify and coordinate its activities in this area, CEPLAS has acquired the support from *Dr. Günter Strittmatter*, as a Consultant for "Technology Transfer and Cooperation Management", beginning of July 2014.

Perspectives for application

With respect to the transfer of research results into application, CEPLAS provides a variety of perspectives:

- Molecular engineering of perennial plants from annuals by integrating the genetic determinants of perennialism
- Transfer of favourable agronomic traits from perennials into annuals: abiotic stress tolerance, nutrient-use efficiency
- Design of synthetic C_4 plants by transferring the genetic/molecular determinants of C_4 photosynthesis into C_3 plants
- Molecular engineering of optimised C_3 and C_4 photosynthesis by targeting the rate limiting steps in the conversion of light energy to chemical energy
- Development of "biologicals" for plant protection and plant nutrition, on the basis of understanding the composition and function of the plant microbiome
- Design of novel chemical components for plant protection and plant nutrition, on the basis of understanding the biochemical processes of "communication" between plants and their microbiome
- Molecular breeding and engineering of plants with improved disease resistance using the molecular components that determine the type and outcome of plant-microbe interactions.
- Modelling of approaches for molecular engineering of improved plant traits
- Development of tool boxes for molecular engineering of improved plant traits



Opportunities for cooperation

On the basis of internal discussions, CEPLAS has decided to offer the following opportunities for cooperations to interested partners from the private sector:

- Joint research projects in fields of common interest
- Out-licensing of IPR-protected research results for economic exploitation
- Internships for scientists at various levels:
 - CEPLAS scientists at companies
 - Company scientists at CEPLAS
- Establishment and co-funding of Graduate Schools in research areas of common interest
- Funding of Postdoctoral Fellows within the CEPLAS programme
- Sponsoring of Junior Professorships

Activities and achievements in 2014

As a first step for an effective and efficient contribution by the Consultant for “Technology Transfer and Cooperation Management”, an intensive discussion round with each individual Principal Investigator and Associated Investigator has been completed, to get an overview on research projects within CEPLAS with potential for cooperation, especially with the private sector. Various such research projects have been identified. Further discussions are required to specify the possibilities and to establish contacts with potentially interested external partners. Beyond these internal discussions, various steps were undertaken to address and attract external partners from industry:

- Development of a “Flyer” describing the application potential of CEPLAS results and the opportunities for cooperation; widespread distribution of this “Flyer” to potential partners
- CEPLAS Days with two German plant breeding companies: personal presentation of the CEPLAS programme by CEPLAS representatives; discussion on cooperation interests and opportunities; with both companies, negotiations on cooperative activities in research and education are under way
- Visits at Syngenta, Gent, and BASF Plant Science, Gent with a group of Postdocs: Presentation of the CEPLAS programme; discussion on cooperation interests and opportunities; negotiations on cooperative activities in research and education ongoing

As a result from these undertakings, a joint project with a large German Life Science company in the field of mathematical modelling of biological processes has been initiated, beginning of 2015.

Outlook

In January we will organise the event “CEPLAS Meets Industry”, in Düsseldorf where we present an overview on the CEPLAS programme and presentation of highlights and perspectives of Research Area B and Research Area C to invited representatives from industry in Germany and neighbouring countries. In addition, a seminar for CEPLAS Graduate Students and Postdocs on career opportunities in plant breeding companies will be held (March 2015, Cologne), in order to assist these young scientists in their professional career planning. Finally, a presentation on “Requirements for Establishing Successful Cooperations with Industry” will be given by G. Strittmatter in the course of the regular CEPLAS Friday seminar series.

11.

Public outreach

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Public relations work and press

Internet and social media presence

At the beginning of 2014, the CEPLAS website was reconstructed and relaunched. It includes now a members login function as well as a German part for the interested public. In addition, the website was equipped with an analytics tool and since its installation in June our homepage was visited almost 20.000 times with over 100.000 page views.

At the same time CEPLAS started its own facebook page (www.facebook.com/plantsoftomorrow) where we share events, publications, interesting articles and open positions.

Public lecture series:

Vom Urweizen der Steinzeit zu den Genpflanzen der Zukunft

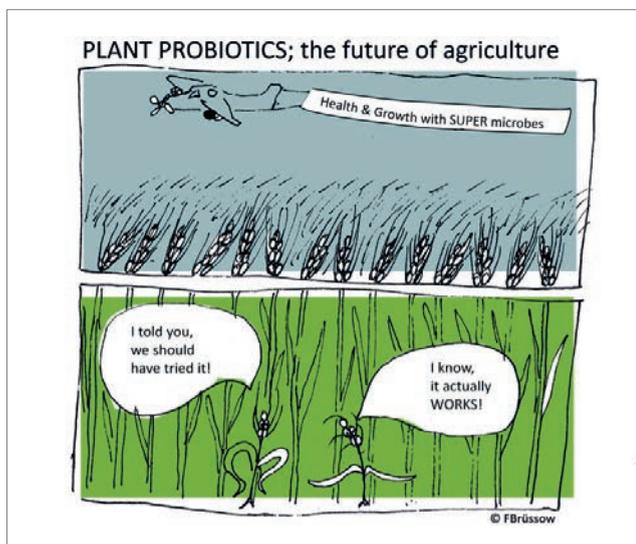
During the summer term 2014, we organised a public lecture series on the domestication of crop plants and their future development that was held by *Peter Westhoff*.

Over 60 people followed his comprehensive overview from the beginning of agriculture and the methods of plant breeding to the challenges of the future. Following the lecture, the mixed-aged audience had the opportunity to discuss further questions and issues in a comfortable atmosphere.

Planter's Punch

In order to communicate more of the science that is done in the CEPLAS labs to the public, a new feature was started in July 2014. Under the heading "Planter's Punch" CEPLAS young researchers present each month one special aspect of the CEPLAS research programme.

All contributions are published in English and German via our homepage and facebook page.



CEPLAS Women's Career Day

Our second Women's Career Day took place on June 16th 2014 in Cologne. Four female guest speakers gave insight into their work in industry, academia and publishing, as well as into their personal career development. Following the talks there was an informal meeting with the speakers for further discussion on career and family issues.

CEPLAS initiative "Research & Education"

This initiative is focused on promoting the interest of students and teachers in plant science research at advanced secondary-schools ("Oberstufe") aiming to increase the awareness of complex relationships of plants and their environment. It will be implemented in cooperation with secondary-schools within the Cologne-Düsseldorf area together with the "Zentrum für LehrerInnenbildung" (ZfL) and the "Fachdidaktik Biologie" at the University of Cologne (UoC). In cooperation with the ZfL and the Fachdidaktik Biologie „teaching packages“ for selected CEPLAS research topics will be established to support teachers with an educational design.

In addition, lectures and workshops related to individual CEPLAS topics will further attract interested secondary-school students and teachers for these research areas.

Eventually, outstanding and highly motivated secondary-school students will have the possibility to do their mandatory school-practical ("Facharbeit") for four weeks in a CEPLAS laboratory.

Sponsoring activities

In 2014 CEPLAS supported two international conferences. For the ESF (European Science Foundation) conference "Biology of plastids - towards a blueprint for synthetic organelles in Poland we offered two travel grants for female scientist.

In addition we supported the 2nd European Workshop on Peptide Signalling in Plants.

Outlook

Due to the positive feedback, the public lecture series mentioned above will be repeated in 2015 in Cologne.

Moreover CEPLAS participates in the reconstruction of the crop plants section of the Botanical Garden of the HHU that is currently ongoing. CEPLAS will design a small section about current and future (energy-)plants. Opening of the whole section is planned for June 2015.

For students, CEPLAS plans to offer a course on plant sciences within the school holidays. The goal is to give interested students an insight into plant science, which might help them to decide about their future subject of studies.

Media presence 2014

Articles and press releases

- **March 31**
„Vom Urweizen der Steinzeit zu den Genpflanzen der Zukunft“ – Öffentliche Vortragsreihe im Rahmen des Exzellenzclusters CEPLAS der HHU. *Press release of the Heinrich Heine University Düsseldorf.*
- **April**
Freundliche Pilze: Wissenschaftler erforschen das Mikrobiom von Pflanzen – Friendly Fungi: Scientists have been researching the Microbiomes of Plants“ *forschung 365, Das Wissenschaftsmagazin der Universität zu Köln/The University of Cologne's Journal of Science*, Ausgabe 01/14, pp. 72-73.
- **April 29**
Prof. Westhoff: Vom Jäger und Sammler zum Ackerbauern und Viehzüchter. *Press release of the Heinrich Heine University Düsseldorf.*
- **May 13**
Gene und Genome – was macht ein Pflanzenzüchter? *Press release of the Heinrich Heine University Düsseldorf.*
- **May 21**
„Genpflanzen“ – Chance für die Landwirtschaft oder Gefahr für den Verbraucher? *Press release of the Heinrich Heine University Düsseldorf.*
- **May 27**
Genpflanzen – Chance für die Landwirtschaft oder Gefahr für den Verbraucher? *Rp-online.*
- **May 30**
Neue Gesichter bei CEPLAS. *Press release of the Heinrich Heine University Düsseldorf.*
- **May 31**
„Genpflanzen“ – Chance für die Landwirtschaft oder Gefahr für den Verbraucher? Vortragsreihe Prof. Westhoff/CEPLAS. *Press release of the Heinrich Heine University Düsseldorf.*
- **June 4**
“Questions for Prof. Dr. Bucher“. Contribution in the brochure “meeting point cologne 2014/2015“, Cologne Convention Bureau, pp. 8-9.
- **June 11/12**
Jungen Wissenschaftlerinnen ein Vorbild geben – Zweiter Women's Career Day des Exzellenzclusters CEPLAS an der Universität zu Köln. *Press release of the University of Cologne and Heinrich Heine University Düsseldorf.*
- **June 12**
Pflanzen und Mikroben – Freunde oder Feinde? *Press release of the Heinrich Heine University Düsseldorf.*
- **June 13**
Kölner CEPLAS Professuren komplett. *Press release of the University of Cologne.*
- **June 26**
Die Herausforderungen der Zukunft. *Press release of the Heinrich Heine University Düsseldorf.*
- **August 23/24**
Gentechnik auf der Gabel. *Kölner Stadtanzeiger – Magazin am Wochenende: Rettung oder Risiko? Ob Grüne Gentechnik den weltweiten Hunger besiegt oder ob sie uns krank macht – darüber streiten selbst Experten. Ausgabe Nr.195.*

- December
CEPLAS-Cluster of Excellence on Plant Sciences, members feature, *EPSO* (European Plant Science Organisation) Newsletter December 2014.
- December 29
Die Universität im Aufbruch. *MAGAZIN DER HEINRICH-HEINE-UNIVERSITÄT DÜSSELDORF*, Ausgabe 03/2014, pp. 6-8.

Radio features

- February 5
WDR5, Westblick, Prof. Dr. Peter Westhoff, Title: „Genmais auch in NRW?“
- April 6
WDR2, Stichtag, Prof. Dr. Paul Schulze-Lefert, Title: „Erster Versuch mit Gen-Weizen. Brotweizen, resistent gegen Pilzbefall.“

Print products 2014

Flyers

- CEPLAS – english and german
- Excellent science for tomorrow's plants: Opportunities for Public-Private Partnerships
- CEPLAS Meets Industry
- Vom Urweizen der Steinzeit zu den Genpflanzen der Zukunft

Posters

- Displays of Research Area Topics
- Posters for several CEPLAS events

Others

- Notepads
- CEPLAS pen
- Christmas cards



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