CURRICULUM VITAE

Anna Kasprzewska PhD

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EDUCATION

University	Major	Minor	Date Attending	Degree
Warsaw Agricultural University	Genetics	Mol.Biol.	1995 – 2000	M.Sc.♣
University of Warsaw	Biology	Mol.Biol.	2001 – 2005	Ph.D.

(♣) During master degree studies I attended individual programs of education containing additional courses on:

molecular virology of animals, plant virology, immunology, microbiology, genetic engineering, phytopathologic diagnostics, mechanisms of plant resistance against pathogens

M.Sc. Thesis 2000: "Searching for the gene-specific resistance against the snow mould fungal disease caused by the *Microdochium nivale* - using the chemical mutagenesis approach"; Department of plant genetics, breeding and biotechnology, Warsaw Agricultural University, name of tutor: Prof. Stefan Malepszy

Ph.D.Thesis 2005: "The role of the class IV chitinase in *Arabidopsis thaliana* development"; Department of Plant Experimental Botany, University of Warsaw, name of tutor: Prof. Alina Kacperska

Postdoctoral position 2006-2008: "Small RNAs in Rice and Maize", Department of Biological Sciences, University of South Carolina, USA, Dr Vicki Vance/ Lewis Bowman labs

M. Curie Individual Early Stage Fellowship 2009-2011
"MOLCALM – Molecular and cellular analysis of leaf margin" Sheffield University UK

Visiting Scientist 2011-2012 Sheffield University UK

TEACHING EXPERIENCE

- ✓ Courses on: plant tissue cryopreservation and gene banks, plant stress responses, laboratory course of plant experimental biology techniques, plant tissue cultures, plant physiology for M. Sc students.

- ✓ Presentation of the molecular plant biology techniques during The VI Science Festival, Warsaw 2002. *The goal of this festival is to promote the science in Poland and each year thousands of scholars attend this event.*

RESEARCH WORK EXPERIENCE

Master degree

During master degree studies I worked on a part of a bigger project titled "Searching for the gene-specific resistance against the snow mould fungal disease caused by the *Microdochium nivale* - using the chemical mutagenesis approach". The main aim of this project was to investigate using forward genetics approach molecular bases of rye plant resistance to snow mould and develop new rye cultivars. My duties in this project were to prepare rye plants resistant to pathogenic fungi *M. nivale*. To increase genetic diversity I used chemical mutagenesis (EMS) and after mutagenesis I selected plants resistant to snow mould. Disease resistance of plants selected in growth chamber conditions was confirmed in field experiments. Using this experimental approach I obtained several plants showing increased resistance or increased ability to recover. These plants were further used as objects of studies on molecular bases of snow mould resistance and as components for breeding.

Ph. D. degree

The aim of my Ph.D. thesis was to investigate role of chitinase class IV in plant development. To reach this goal I used reverse genetic approach i.e. *Arabidopsis thaliana* T-DNA mutants (from Salk and Syngenta collections) with insertion in chitinase class IV gene. To confirm that any phenotypic changes observed in these mutants were truly caused by disruption of chitinase function I also used plants with silenced the same gene (construct were made in pHANNIBAL and pART vectors). The disruption of gene function I confirmed by in-gel chitinase activity assay. In both cases (T-DNA insertion and gene silencing) disruption of chitinase class IV function lead to changes in root system architecture due to increased formation of lateral roots. Observation on cleared roots showed increased number of lateral root primordia formation in the mutant plants. No morphological changes within each primordium developmental stage were observed. These phenotypic changes seemed to be dependent on such culture conditions as light intensity and concentration of sucrose in growing medium. These results allowed me to build hypothesis that chitinase class IV could be involved in regulation of *Arabidopsis* lateral root primordia formation under specific growth conditions.

Postdoctoral position

The main aim of "Small RNAs in Rice and Maize" project was to clone and sequence population of small RNAs isolated from different developmental stages, tissues and organs and from plants subjected to different abiotic and biotic stresses. I prepared small RNA libraries from plants subjected to abiotic stresses, virus infections and from mature and meristematic tissues of healthy plants. The libraries complexity I checked by detection of known miRNA conserved between plant species. Moreover, to confirm accuracy of small RNA libraries I checked experimentally a set of potential targets predicted by computational approach for new miRNA sequenced within these libraries. Other libraries from rice mutant plants having disturbed silencing pathways and from maize embryos and floral organs are under construction. All obtained data are currently organized into freely available web interface that will allow to the research community searching for small RNA sequences, their expression profile, predicted miRNA precursors and for predicted miRNA and siRNA targets.
Another project I was involved in is the determination of transcription start sites of miRNA precursor primary transcripts in rice and maize. The determination of miRNA precursor transcription start sites will significantly increase our knowledge about spatial and temporal regulation of miRNA expression and their further function in rice and maize plants. To reach this goal I am cloning miRNA precursor primary transcripts using 5'-RACE technique. Further analysis of miRNA precursors promoter

sequences and prediction transcription factor binding sites will be done using *in silico* approach. This work has been published: (Johnson et al., 2009).

Marie Curie Independent Early stage Fellowship: MolCALM Molecular and Computational analysis of the Leaf Margin, Department of Animal and Plant Sciences Sheffield University, UK

The main idea behind this individual research project was to understand cellular and molecular basis of the leaf margin growth. In particular I was interested in understanding how margin cells influence the final leaf shape. The main outcome of the project was understanding of the contribution of LAX genesin final leaf shape and form. In particular I have described their involvement in local accumulation of auxins in regulation of lobe outgrowth. Work has been published in The Plant Journal (Kasprzewska et al., 2015). I was also involved in work on local manipulation of cell growth in leaf margin, also published in The Plant Journal (Malinowski et al., 2011).

Currently works on the deciphering mechanisms governing flowering process in potato – IPG PAS

Technical skill:

Molecular biology:
preparation of small RNA libraries; small RNA isolation and detection; 3'- and 5'-RACE; designing and making vector constructions for plant transformation (incl. cloning, sequencing etc.); posttranscriptional gene silencing (PTGS) in *Arabidopsis*; DNA/RNA isolation, Southern and northern analysis; bacteria transformation; PCR analysis; radioactive probe preparation; basic and acidic native protein electrophoresis; SDS-PAGE electrophoresis; immunoblotting; in-gel chitinase activity assay; methods of extracellular protein isolation, purification and concentration; arabinogalactan proteins (AGPs) study, including protein detection with Yariv reagent.

In vitro culture:
Arabidopsis floral dip transformation and selection of transformants; generation and maintaining of *Arabidopsis* root culture in liquid medium; screening of *Arabidopsis* T-DNA mutants and RNA-i knock-outs using several *in vitro* culture approaches.

Bioinformatics:
Database homology searching (GenBank, EBI, Swiss Prot), gene architecture analyses (packages: EMBOSS, GENE BUILDER; programs: BLAST; cis acting elements data bases: PLANT CARE, SIGNAL SCAN), primer designing (Oligo 6.0), prediction of RNA secondary structures using mfold and Vienna RNA.

Microscopy:
Basic techniques of optical microscopy and sectioning, including tissue clearing techniques and staining. Epi-fluorescent microscopy of plant cells and organs. Imaging of auxin transport and gene activity with in transgenic lines with appropriate fluorescent markers. Basic skill on confocal microscopy.

OTHER EXPERIENCE

Courses:

- ✓ Training in the biochemical techniques of the plant cell wall study - Institute of Bioorganic Chemistry, Polish Academy of Sciences (under supervision of Prof. Przemyslaw Wojtaszek)
- ✓ Confocal Microscopy Training, 2012 University of York, UK

Meetings and conferences:

- ✓ Meeting of the Polish Frost Resistance Working Group, Skierniewice, Poland 2001 (oral presentation)
- ✓ 4th International Conference - "Ecophysiological Aspects of Plant Responses to Stress Factors", Krakow, Poland 2001 (oral presentation)
- ✓ I Meeting of Polish Plant Experimental Biology Society, Olsztyn, Poland 2003 (oral presentation)
- ✓ XIV Congress of the Federation of European Societies of Plant Biology, FESPB, Krakow, Poland 2004 (poster presenter)
- ✓ 13th New Phytologists Symposium - "The role of the extracellular matrix in the control of plant development", The Linnean Society, London, UK, 20-21 January 2005
- ✓ 16th Penn State Symposium in Plant Physiology – "RNA biology: novel insights from plants systems", May 18-20 2006

SUBJECT OF INTERESTS:

- ✓ Molecular mechanisms of plant growth and differentiation,
- ✓ Regulation of plant development by silencing pathways (miRNA, siRNA, ta-siRNA),
- ✓ Regulation of gene expression in plants,
- ✓ Molecular mechanisms of viral infection and propagation in plants,
- ✓ Mechanisms of plant resistance to biotic stimuli, especially plant-microorganism interactions,
- ✓ Molecular mechanisms governing morphogenesis in model and crop plants

HONORS and AWARDS

- The first prize honour for the best young scientists oral presentation during the 4th International Conference - "Ecophysiological Aspects of Plant Responses to Stress Factors", Krakow, Poland 2001

- M. Curie Independent fellow while wrking at Sheffield University

PUBLICATIONS

Blicharz, S., Stefanowicz, K., Truman, W., Basinska-Barczak, A., Singh, D., **Kasprzewska, A.**, *et al.* (2025) Laser dissection-assisted phloem transcriptomics highlights the metabolic and physiological changes accompanying clubroot disease progression in oilseed rape. ***Plant J***, **121,** e17156.

Malinowski, R., Singh, D., **Kasprzewska, A.**, Blicharz, S. and Basińska-Barczak, A. (2024) Vascular tissue – boon or bane? How pathogens usurp long-distance transport in plants and the defence mechanisms deployed to counteract them. ***New Phytologist***, **243,** 2075-2092.

Kasprzewska, A., Carter, R., Swarup, R., Bennett, M., Monk, N., Hobbs, J. K., *et al.* (2015) Auxin influx importers modulate serration along the leaf margin. ***The Plant Journal***, **83,** 705-718.

Malinowski, R*., **Kasprzewska, A*.** and Fleming, A. J. (2011) Targeted manipulation of leaf form via local growth repression. ***The Plant Journal***, **66,** 941-952. [*equal first authorship]

Johnson, C., **Kasprzewska, A.**, Tennessen, K., Fernandes, J., Nan, G.-L., Walbot, V., *et al.* (2009) Clusters and superclusters of phased small RNAs in the developing inflorescence of rice. ***Genome research***, **19,** 1429-1440.

Kasprzewska A. 2003. Plant chitinases - regulation and function. **Cellular and Molecullar Biology Letters** 8/3: 809-824