Summary

Formins are multidomain proteins containing a conserved formin-homology 2 (FH2) domain, which catalyzes *de novo* nucleation of actin filaments. In yeast and animal cells, both mechanisms and regulation of formin function have been extensively studied, yet much less is known about action of plant formins, which considerably differ from yeast and animal ones in the domain composition. In higher plants, formins are classified into two groups, Class I and Class II, and so far, experimental data are available only for the first group members. Here I present results of experimental study of several members of the large formin family in *Arabidopsis*, including the characterization of a Class II formin AtFH16.

Arabidopsis genome contains 21 formin-encoding genes, and though they greatly differ in their expression levels and pattern, all of them are transcriptionally active. We selected 17 homozygous T-DNA insertional mutants in 14 formin genes. Under standard cultivation conditions, no obvious phenotypic discrepancies between wild type and mutant plants were found. To impair two dominant pollen formins, an *atfh3atfh5* double-mutant was prepared and even in this case, both microspore development and pollen tube growth remained unaffected. Consistently, polarized growth of tobacco pollen tubes was not altered after the targeting of Class I formins by antisense oligonucleotides (ODNs). I also cloned cDNA of *AtFH3* and revised existing gene predictions (Cvrcková et al., 2004).

To study *in situ* localization of pollen-specific AtFH3 together with other formins and cell polarity regulators, a set of rat polyclonal antibodies against synthetic KLH-conjugated oligopeptides was prepared and tested. However, most rat antisera recognized the same background KLH-related plant antigen (KRAP) in *Arabidopsis* and tobacco. We characterized KRAP with respect to size and cellular localization and examined possible antigen-specific reasons for the failure of most immunizations (Oulehlová et al., 2009).

A Class II formin AtFH16 with an unusual domain composition was closely characterized. Under specific conditions (hard tilted agar or continuous darkness), an *atfh16* null mutant exhibited moderate phenotypical changes, especially shortening and waving of etiolated hypocotyls that was further emphasized after treatments with cytoskeletal drugs. Our results indicate involvement of AtFH16 in some aspects of cell expansion in *Arabidopsis* seedlings. Furthermore, we cloned full-lenght cDNA of *AtFH16* and characterized subcellular localization of AtFH16-derived variants *in vivo*. Co-localization studies revealed that AtFH16 can associate with filamentous cytoskeletar structures; in some cases, it labels stabilized actin cables via its N-terminal part, but mostly, AtFH16 decorates microtubules. We identified that a presence of the conserved FH2 domain is required for AtFH16 localization on microtubules. Our results suggest that *Arabidopsis* Class I formins might not be essential for microspore development and for subsequent polarized growth of pollen tubes. Further, Class II formin AtFH16 is capable of microtubule association and could be a potential cytoskeletar crosslinker