

Formins are evolutionarily conserved proteins participating in actin and microtubule organisation, affecting thus also intracellular transport, cell growth, morphogenesis and cell polarity. All formins contain FH2 domain, known to dimerize and act as a nucleator of actin. Angiosperms have two formin clades, Class I and Class II, which are distinguished by domain organisation. Based on knowledge from animal models and protein sequence homology, two groups of candidate membrane-associated formin interactors have been proposed in Arabidopsis (Cvrčková, 2013).

First group of candidates consists of FYVE domain-containing proteins FAB1A (At4g33240) and FAB1B (At3g14270), the other contains proteins with BAR and SH3 domains AtSH3P1 (At1g31440), AtSH3P2 (At4g346600) and AtSH3P3 (At4g18060).

Yeast two hybrid assay was used to examine protein interactions of selected proteins from both candidate groups (FAB1A, SH3P2 and SH3P3) with FH2 domains representing both plant formin clades. The same experimental setup was also used to test dimerization among FH2 domains of plant formins. Translational fusions of FH2 domains from Class I formins AtFH1 (At3g25500), AtFH5 (At5g54650) and Class II representatives AtFH13 (At5g58160) and AtFH14 (At1g31810) with the GAL4 activation domain have been co-expressed in yeast with GAL4 DNA binding domain fusions of the candidate interactors or other plant formin FH2 domain.

Strong interaction between the FH2 domains of AtFH5 with AtSH3P3 protein has been confirmed, while the other candidates did not interact in this experimental setup.

Homodimerization of FH2 domain of AtFH13 has been confirmed as well as heterodimerization of FH2 domains of AtFH13 with AtFH14, showing that heterodimerization between FH2 domains of closely related non-identical formins may take place. However, no other dimerization was observed, albeit this does not rule out the possibility that other interactions may take place under different experimental conditions.