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The role of Cdc42p in cell polarization of the yeast *Saccharomyces cerevisiae*

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We were able to further elucidate the mechanism of Cdc42p polarization in the yeast *Saccharomyces cerevisiae*. Two distinct pathways for the polarization of Cdc42p were established. One involves acto-myosin based transport of membrane bound Cdc42p. The second pathway polarizes cytosolic Cdc42 molecules and requires the function of the Rho GDP dissociation inhibitor Rdi1.

We found that polarization of either GDP- or GTP-locked Cdc42p was completely actin dependent. These cycling-deficient mutants of Cdc42p could not cycle through the cytoplasm and remained membrane bound. For membrane extraction cycling between the GDP- and GTP-bound forms of Cdc42p was necessary.

The second pathway for Cdc42p polarization is Rdi1 dependent. Without Rdi1p function, Cdc42p remained mostly membrane bound and the polarization of Cdc42p became completely actin dependent. Overexpression of Rdi1 correspondingly led to increased membrane extraction of Cdc42p.

We also found that actin dependent transport was important for maintaining the site of polarization during bud formation. Inhibition of transport led to unstable polarization.

For Cdc42p targeting, the C-terminal end with its CAAX-box and polylysine domain is of high importance. The geranylgeranylation of the Cystein in the CAAX box appeared necessary for membrane association and the CAAX-box together with the polylysine domain was sufficient for plasma membrane association and was polarized during bud formation. Mutations in the polylysine domain led to reduced membrane association and to reduced spontaneous polarization of activated Cdc42p. Change of charge mutations in the polylysine domain most likely leads to an increased rate of endocytosis, possibly by increased Rdi1 membrane extraction.

Future research should target the exact pathways for endocytic recycling and its importance and relevance for polarization. One important aspect of the recycling should be to find out the mechanism of Rdi1p function. Also how the Rdi1p recycling is organized and directed. This should involve research if there are specific release factors for Rdi1.

For the polarization of Cdc42p it is important to find out further parameters, like the diffusionrate, transportrate and endocytosisrate and how these are affected.

Especially the exact role of the polybasic region will be interesting with its affects on the stated parameters. Also the function and interaction of other membrane proteins and negatively charged phospholipids for Cdc42p will need further research.