NEW INSIGHTS OF \textit{Yarrowia lipolytica} MORPHOGENESIS MODULATED BY POLYAMINES

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Polyamines modulate fungal dimorphism by mechanisms that remain elusive. Here, we investigated whether polarized growth of \textit{Yarrowia lipolytica} regulated by polyamines occurs through activation of plasma membrane H\textsuperscript{+}-ATPase and the recruitment of auxin-dependent pathways. The involvement of polyamines and auxin (IAA) in hyphal growth was investigated by culturing cells with or without spermidine (Spd), spermine (Spm), polyamines biosynthesis inhibitor (CHA), IAA, auxin transport inhibitor (TIBA), and auxin signaling inhibitor (PCIB). H\textsuperscript{+} pumping, ATPase activity and expression of P-type H\textsuperscript{+}-ATPase were analyzed from isolated membrane vesicles. Yeast-hypha transition of \textit{Y. lipolytica} was stimulated with 1 mM polyamines, since only 30-40\% of Spd/Spm-cultivated cells remained in the yeast form versus 50-55\% of the control cells at 36 h. Culture of 2 mM CHA prevented transition in 80\% of cells. On agar, Spd/Spm resulted in wrinkled colonies surrounded by hyphal fringes longer than those of control colonies, whereas CHA produced smooth colonies with the shortest fringes. Determination of H\textsuperscript{+} pumping and ATPase activity of total membranes from \textit{Y. lipolytica} cells revealed that P-type H\textsuperscript{+}-ATPase activity increased with morphogenesis. ATP-dependent and vanadate-sensitive H\textsuperscript{+}-pumping in membranes isolated from cells grown in Spm was 2.5-fold higher than in the control membranes. Although Spm did not significantly increase ATPase activity compared to the control, the expression of Pma1p was nearly 1.5-fold higher over control values at 20 h. Exogenous IAA largely reversed the inhibitory effect of CHA, whereas the inhibitory effects of TIBA and PCIB on dimorphic transition were not reversed by exogenous Spd or Spm. Additionally, the stimulatory effect of Spm on H\textsuperscript{+}-pumping was abolished by TIBA and PCIB. Polyamines and IAA markedly stimulated extracellular alkalization by \textit{Y. lipolytica} cells on agar plates, whereas their inhibitors prevented it. Altogether, the data suggest the involvement of auxin signaling in polyamine-induced plasma membrane H\textsuperscript{+}-ATPase during the yeast-to-hyphal transition.

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