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Cell wall maintenance in fungi

Hans de Nobel, Herman van den Ende and Frans M. Klis

To resist internal turgor pressure a fungal cell needs a strong wall. When confronted with cell-wall-degrading enzymes, fungal cells swell and burst. This explains why organisms often defend themselves against fungal invasion by producing such enzymes. The need for a sturdy wall has to be balanced against the need for growth, branching, cell fusion and other morphogenetic events, which all require drastic remodeling of the wall and therefore temporarily increase the risk of cell lysis. To cope with these contradictory demands, fungi possess a signaling pathway to maintain cell wall integrity. This pathway has been particularly well studied in the model fungus *Saccharomyces cerevisiae* and is known as the cell-wall-integrity pathway, or Slt2p/Mpk1p pathway after the name of the corresponding mitogen-activated protein kinase (MAPK). In a recent report, Jung and Levin[1] described >20 genes controlled by this pathway, most of which are cell-wall-maintenance genes. Using DNA-array filters, they studied gene expression in response to constitutive activation of the cell-wall-integrity pathway and found all but one of the affected genes to be dependent on a single transcription factor, Rlm1p.

Cell wall damage triggers a salvage mechanism

The cell walls of *S. cerevisiae* and *Candida albicans* consist of only four classes of macromolecules, namely cell wall proteins, β1,6-glucan, β1,3-glucan and chitin, which are interconnected by covalent bonds[2–4]. Mutants defective in the synthesis of particular cell wall components show characteristic alterations in the composition and architecture of their walls that seem to compensate for the loss in strength caused by the mutation. More chitin is deposited in the lateral walls, and the expression of several cell wall proteins, and of the alternative subunit of β1,3-glucan synthase, Fks2p, is upregulated. Increased chitin deposition and Fks2p expression have also been reported for yeast cells challenged with wall-perturbing agents[5]. It has been proposed that these changes are part of a salvage mechanism that is activated in response to cell wall weakening[2,4–7]. The recent work by Jung and Levin confirms these earlier results and presents a much more complete picture of how a fungus copes with cell wall stress, thereby vividly illustrating the power of global transcript analysis.

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Conservation of the cell-wall-integrity pathway in other fungi

The cell-wall-integrity pathway seems to be conserved in several yeasts and filamentous fungi. The best example of functional conservation is provided by the fission yeast *Schizosaccharomyces pombe*. Similar to the GTPase Rho1p in *S. cerevisiae*, *S. pombe* Rho1p stimulates protein kinase C activity. Additionally, loss of this protein kinase, Pck2p, results in a severely weakened cell wall[8,9]. Pck2p might exert its effect on cell wall construction partly via activation of a downstream MAP kinase cascade consisting of *MKH1*, *PEK1/SKH1* and *PMK1/SPM1*, which encode typical MAPK kinase kinase (MEKK), MAPK kinase (MEK) and MAPK activities, respectively. Loss of any of the kinases in this pathway results in the appearance of rounded instead of rodlike cells that are more sensitive to β-glucanase, suggesting that this pathway is involved in cell wall maintenance[8,10]. Components of a cell-wall-integrity pathway have also been identified in the human pathogenic dimorphic fungus *C. albicans*, where loss of the protein kinase C, Pkc1p, results in osmotically remedial cell lysis of both the yeast and hyphal form[11]. Effectors directly downstream of Pkc1p in *C. albicans* have not yet been isolated, but a functional homolog of *S. cerevisiae SLT2/MPK1* is present and deletion of this MAP kinase, Mkc1p, results in defects in cell wall construction[12]. Homologs of *S. cerevisiae MPK1* with a function in cell wall maintenance have also been identified in *Aspergillus nidulans*, which is genetically well accessible and an important model system for food-spoilage fungi, and in the plant pathogenic fungus *Magnaporthe grisea*[13,14].

Alternative signaling pathways

Many cell-wall-maintenance genes are probably under the control of multiple signaling pathways. Transcript studies have indeed identified cell wall proteins whose expression is either cell-cycle-regulated and/or depends on nutrient or oxygen availability, underscoring the dynamic nature of cell wall assembly (reviewed in Ref. 4). In addition, growth at elevated temperatures activates Mpk1p but results in various expression patterns for the Rlm1p-dependent genes, suggesting that multiple signaling events take place in response to heat stress[1]. One possible explanation is provided by the presence of stress-response elements (STREs) in the promoters of several cell-wall-maintenance genes[15].

Conclusions and perspectives

The identification by Jung and Levin[1] of at least 18 cell-wall-maintenance genes that are controlled by the Slt2p/Mpk1p pathway and the transcription factor Rlm1p confirms the dynamic nature of the cell wall and the importance of this pathway in cell wall maintenance. Cell wall assembly is likely to be affected by additional signal transduction pathways depending on the environmental conditions. Together, they allow fungal cells to sense and adapt to various stress conditions. This raises the question how these pathways interact and whether they operate in a hierarchical manner. Transcript analysis does not necessarily give the full picture in the case of the cell-wall-integrity pathway. As cell-wall-maintenance proteins generally follow the secretory pathway, which is a comparatively slow process, one may expect the cell-wall-integrity pathway to also include a set of rapid responses, possibly involving local activation of cell wall synthases. This putative rapid response could then be backed up by a secondary response involving transcriptional activation of cell-wall-maintenance genes, as described by Jung and Levin[1].

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