glabrata response and tolerance to acetic acid thereby enhancing colonization of vaginal epithelium

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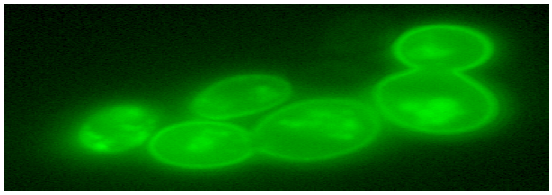
To successfully colonize the vaginal tract *Candida glabrata* has to cope with various stresses including the presence of acetic acid at a low pH that is produced by the bacteria that co-colonize this niche. The genes/pathways involved in *C. glabrata* tolerance and response to acetic acid are largely unknown, although these are a highly interesting set of novel targets to control vaginal infections caused by this yeast. *Saccharomyces cerevisae* response and tolerance to acetic acid was found to be largely mediated by the ScHaa1 transcription factor [1,2,3]. In this work the involvement of CgHaa1 in *C. glabrata* tolerance and response to acetic acid is demonstrated. Elimination of *CgHAA1* gene from *C. glabrata* genome dramatically increased susceptibility of this pathogenic yeast to acetic acid (30 mM at pH 4.0). Around 140 genes were found to be up-regulated, directly or indirectly, by CgHaa1 in response to acetic acid stress, based on

results of a transcriptomic analysis. Functional clustering of the genes activated by CgHaa1 under acetic acid stress shows an enrichment of those involved in carbohydrate metabolism, transport, cell wall maintenance, regulation of internal pH and nucleic acid processing. At least five of the CgHaa1-regulated genes were found to increase *C. glabrata* tolerance to acetic acid including *CgGAD1*, encoding a glutamate decarboxylase; *CgTPO2/3*, encoding a drug efflux pump of the Major Facilitator Superfamily; *CgYPS1*, encoding a cell wall aspartyl protease; and *CAGL0H04851* and *CAGL0E03740*, encoding two uncharacterized ORFs. Altogether our results are consistent with the concept that the CgHaa1-signalling pathway increases *C. glabrata* tolerance to acetic acid by reducing the internal accumulation of the acid and by up-regulating the activity of the plasma membrane proton pump H^+-ATPase CgPma1, two essential features for a robust weak acid response.

The role exerted by CgHaa1 in the ability of *C. glabrata* to colonize reconstituted vaginal human epithelium (RVHE) in the presence of acetic acid (30 mM at pH 4.0) was also investigated in this work. In the absence of acetic acid wild-type and Δ*CgHaa1* mutant cells were able to colonize RVHE at a similar rate, however, in the presence of acetic acid colonization of the vaginal tissue was markedly reduced in the mutant background. The reduced colonizing capacity of Δ*CgHaa1* mutant cells was correlated with a reduced expression of the adhesin-encoding genes *EPA6*, *EPA7* and *EPA1* and with a lower adhesiveness to the extracellular matrix proteins fibronectin and vitronectin.

[1] Mira NP, Becker J and Sá-Correia I, OMICS:14, 587-601, (2010)
[2] Mira NP, Teixeira MC and Sá-Correia I, OMICS:14, 525-40, (2010)
[3] Mira NP et al., Nucleic Acids Res., 16, 6896-907, (2011)



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