

Characterization of the Heat Shock Transcription Factor Hsf1 of *Saccharomyces cerevisiae* in Response to High Hydrostatic Pressure

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Eukaryotes have evolved mechanisms to survive to environmental challenges, as fluctuations in hydrostatic pressure (HP). Previous results have shown that the stress transcription factors Msn2/4 were mainly activated when *S. cerevisiae* was exposed to 50MPa. However, Msn2/4 were not activated by higher pressures, suggesting that other factors might be involved in HP gene regulation. The heat shock transcription factor HSF1, which is conserved from yeast to humans, binds to heat shock element (HSE) and activates transcription under normal and stressful conditions. HSE is formed by different patterns of inverted repeats of nGAAn sequences. In this work, we investigated the role of Hsf1 in response to high hydrostatic pressure stress. Once Hsf1 becomes hyperphosphorylated by heat shock, we investigated whether HP would also induce phosphorylation. Western blot analysis of Hsf1-myc from cells subjected to 100MPa for 30 minutes revealed a distinct phosphorylation pattern that observed after heat shock, indicating that Hsf1 could be differently activated by pressure. As the phosphorylation state of Hsf1 was directly implicated in HSE recognition, we investigated which type of HSE was activated by pressure. Therefore, plasmids containing the different architectures 4Ptt, 3P, Gap and Step of HSE fused to lacZ gene (HSE-CYC1-lacZ) were inserted in yeast. 100MPa treatment induced a 5 fold activation of beta-galactosidase activity of HSE-4Ptt transformed yeast when compared to cells grew at atmospheric pressure. Activation of stress responsive genes under control of the different HSE types, such as *HSP104*, *HSP26*, *CUP1* and *SSA3*, will be analyzed by RT-PCR. These results bring new insights to regulation of Hsf1 in lower eukaryotes and permits the comparative study of adaptive mechanisms used in yeast as well as mammals. Keywords: *Saccharomyces cerevisiae*, stress response, Hsf1, HSE, hydrostatic pressure. Support: CNPq/FAPERJ.