

Beating the Heat: A Translation Factor and an RNA Mobilize the Heat Shock Transcription Factor HSF1

Cells respond to stress via orchestrated modifications in gene expression; in a recent publication in *Nature*, Shamovsky et al. (2006) report that an RNA and a translation factor regulate the activity of a transcriptional activator essential to the mammalian heat shock response.

Gene expression is differentially regulated at multiple levels during the response to heat shock (Morimoto, 1998; Panniers, 1994). For example, general translation and transcription of certain genes are repressed upon heat shock. By contrast, the transcription of heat shock protein genes is rapidly and highly activated under the same conditions. The key player in the activation of heat shock-responsive genes is the transcriptional activator HSF1 (heat shock factor 1). Prior to heat shock, HSF1 is predominantly monomeric and held in an inactive conformation by both intramolecular interactions and chaperone proteins (Voellmy, 2004). After heat shock, HSF1 is localized in the nucleus as a homotrimer bound to specific DNA elements termed HSEs (heat shock elements) located in the promoters of genes that are upregulated in response to heat shock. Studies of the activation of heat shock-responsive genes have been instrumental in defining fundamental principles of transcriptional regulation that are applicable to many genes. While this process has been extensively studied, surprises are still being revealed. In a recent *Nature* publication, Nudler and colleagues report that a translation factor and an RNA work together to regulate the trimerization and consequent activation of mammalian HSF1 (Shamovsky et al., 2006). This work provides new insight into how transcription is upregulated in response to heat shock and raises many new and exciting questions.

With the goal of identifying auxiliary factors that regulate HSF1 activity, Shamovsky et al. (2006) fished for factors in mammalian cell extracts that would associate with a monomeric HSF1 column and found the translation elongation factor eEF1A. Further experiments showed that HSF1 coimmunoprecipitated from cell extracts with eEF1A, with heat shock greatly enhancing the association. This discovery of a component shared between translation and transcription provides an intriguing link between the regulation of these two processes during heat shock. The Nudler group next asked whether eEF1A affected HSF1 activity. In vitro, the fraction containing eEF1A induced HSF1 trimerization and binding to HSE DNA. Surprisingly, more highly purified eEF1A did not induce DNA binding by HSF1, prompting the authors to search for an additional factor in the eEF1A fraction involved in this process. Because eEF1A is known to bind tRNA, they treated the eEF1A-

containing fraction with RNase A and discovered that it lost the ability to stimulate HSF1 DNA binding. This ultimately led to the cloning and identification of an RNA in the eEF1A fraction that they termed HSR1 (heat shock RNA 1).

Subsequent experiments showed that HSR1 is critical for HSF1 trimerization and transcriptional activity in response to heat shock (Shamovsky et al., 2006). In vitro, the addition of purified eEF1A and HSR1 was required for HSF1 trimerization and DNA binding activity. HSR1, like HSF1, coimmunoprecipitated from cell extracts with eEF1A in a heat shock-dependent manner; it isn't yet known whether HSR1 coimmunoprecipitates with HSF1 or if the RNA and HSF1 directly interact. Importantly, when cells were transfected with an antisense oligonucleotide or siRNA directed at HSR1, products from heat shock-responsive endogenous and transfected genes decreased. Moreover, cells could no longer survive a prolonged heat shock, thus clearly showing the importance of HSR1 in regulating the heat shock response.

Figure 1 depicts a model for the mobilization of HSF1 in response to heat shock. Previously, it had been shown that HSF1 trimerization is controlled by chaperone proteins, specifically, Hsp90, p23, and immunophilin (Zou et al., 1998). It is thought that these proteins are bound to HSF1 to maintain it in a monomeric state in non-stressed cells and then dissociate upon heat shock, thereby making it possible for HSF1 to trimerize. Shamovsky et al. (2006) have now shown that eEF1A and HSR1 associate upon heat shock and bring about the trimerization of HSF1. There could be interplay between the release of the chaperone proteins and the activity of eEF1A/HSR1, even though we depict these events sequentially. Once trimerized, HSF1 binds HSE sequences in the promoters of heat shock-induced genes. It is not yet known whether eEF1A and/or HSR1 dissociate from HSF1 trimers or if they bind along with the trimers at promoter DNA.

The studies of Shamovsky et al. (2006) add another layer of understanding of how the mammalian heat shock response is regulated and identify a confluence of three biological systems: transcription, translation, and functional RNAs. Translation shuts down in response to heat shock (Panniers, 1994). The authors postulate that, upon shut down of translation, eEF1A will become available to facilitate HSF1 trimerization. Perhaps HSR1 also plays a role in regulating translation before or after heat shock via its interaction with eEF1A. It is not yet clear how HSR1 is regulated. Its levels do not change with heat shock; however, Nudler and colleagues propose that the RNA itself might serve as a thermosensor via a heat-induced change in conformation. Similarly, it is not yet known where HSR1 is localized in the cell; perhaps its intracellular localization is regulated in response to heat shock. Lastly, HSF1 trimerizes and activates transcription in response to a variety of other environmental stresses and pathophysiological states (Morimoto, 1998). It will be interesting to determine whether HSR1 and eEF1A mobilize HSF1 in response to stresses other than heat shock.

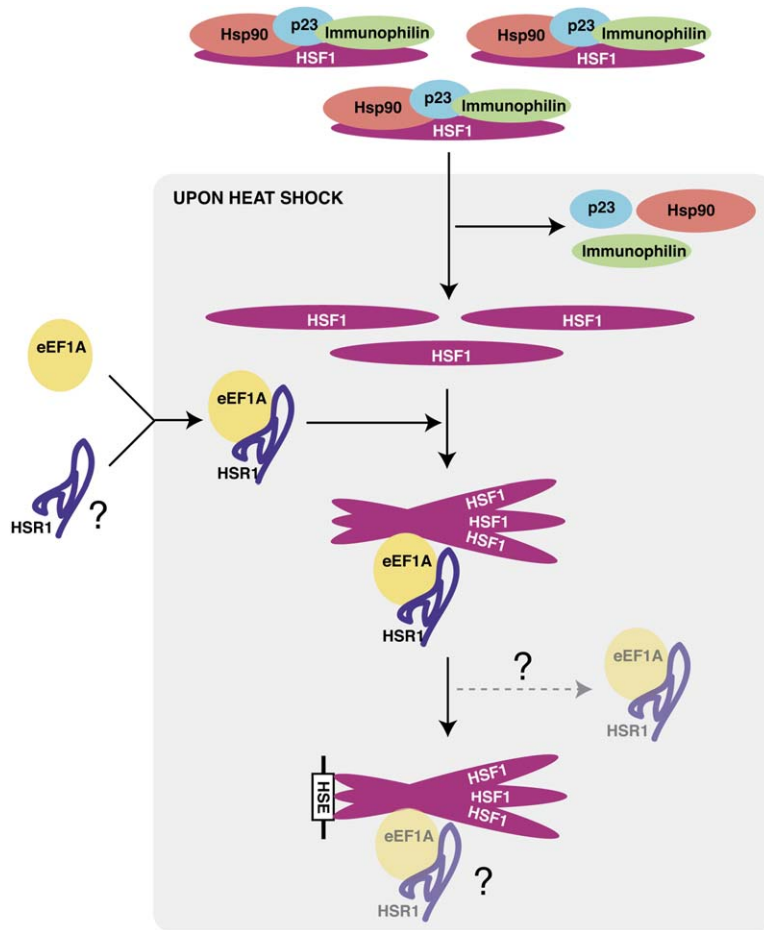


Figure 1. A Model for the Heat Shock-Induced Trimerization and Activation of HSF1 Prior to heat shock (outside of the gray area), HSF1 is predominantly monomeric, held in this conformation by the combined actions of Hsp90, p23, and immunophilin. Under this same condition, HSR1 and eEF1A are present in the cell, with eEF1A functioning as a translational elongation factor and the role of HSR1 being unknown. Upon heat shock, two events lead to the trimerization of HSF1, although the precise order and interplay between these two events is not yet understood. (1) Hsp90, p23, and immunophilin dissociate from HSF1, thereby freeing the monomers to ultimately trimerize. (2) eEF1A and HSR1 associate and bring about the trimerization of HSF1. HSF1 trimers are then able to bind HSE sequences in heat shock-induced promoters and activate transcription. The fate of eEF1A and HSR1 after HSF1 trimerization is unknown. Two possibilities are shown: eEF1A/HSR1 might dissociate from HSF1 trimers or eEF1A/HSR1 might bind to promoter DNA with HSF1 and function in transcriptional activation.

With the discovery of HSR1, there are now three RNAs found to regulate transcription during the mammalian heat shock response, the other two being 7SK RNA (Nguyen et al., 2001; Yang et al., 2001) and mouse B2 RNA (Allen et al., 2004; Espinoza et al., 2004). While all are controlled by cell stress, these three RNAs differentially regulate transcription with respect to the genes they target and their mechanisms of action. Perhaps cells have evolved to use RNAs as regulators of transcription during the stress response; time will tell whether this mechanism is widely used outside of the heat shock response.

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Selected Reading

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