Cell-specific regulation of HSF1-dependent pro-death response to proteotoxic stress.

Accumulation of damaged proteins (e.g. due to cellular stress like heat shock) leads to the activation of the Heat Shock Factor 1 (HSF1). In general, stress-activated HSF1 induces the expression of genes coding for heat shock proteins (HSPs), which are molecular chaperones preventing cell death. However, activated HSF1 can also induce cell death in so-called heat-sensitive cells, which include male germ cells, certain neurons, preovulatory oocytes, and some stages of embryonic development, as well as T lymphocytes, and certain types of cancer cells. We found that in heat-sensitive cells, activated HSF1 can up-regulate the expression of the proapoptotic *Pmaip1* (*Noxa*) gene. This induces cell death and is the primary stress-induced mechanism of cell elimination.

In the current project, we aim to elucidate the tissue-specific mechanisms of *Pmaip1* activation by HSF1. Specifically, we will look for the answer to the following question: what are the mechanisms of cell-specific activity of HSF1, and why does HSF1 bind to *Pmaip1* regulatory sequences and activate *Pmaip1* transcription only in heat-sensitive cells? We plan to test the hypothesis that the ability to induce the *Pmaip1* expression in heat-sensitive cells depends on post-translational modifications and protein partners of HSF1 as well as the accessibility of chromatin to HSF1-containing complexes. Therefore, we plan to search for modifications and binding partners of HSF1 as well as for differences in chromatin accessibility at HSF1 targets in heat-sensitive and heat-resistant cells that determine its tissue-specific activity. Moreover, we will search for hypothetical genes that could be regulated by HSF1 in heat-sensitive cells in a similar manner to *Pmaip1* and validate the generality of mechanisms that regulate differential activation of HSF1 in different cells.

The elucidation of the mechanisms leading to cell death under cellular stress is of great general importance. Impaired spermatogenesis related to elevated temperature is observed in several disorders (e.g. cryptorchidism) or as a result of occupational exposure to heat. Moreover, hyperthermia can also cause neurological and cognitive dysfunction, embryonic death, growth retardation, and developmental defects, while heat shock-induced T cell death is implicated in immune deficiency. Furthermore, due to the supporting action of HSF1 in cancers and impaired activity in aging cells, HSF1 is considered a potential target for the treatment of cancers and disorders associated with protein aggregation (that include Alzheimer's disease, Parkinson's disease, and prion diseases). However, considering the discovery of alternative pro-death signaling of HSF1, its therapeutic targeting may have opposite consequences in cells that differ in stress sensitivity (and may not be appropriate in some types of target cells).

Our grant proposal addresses the basic mechanisms of transcription regulation in general, with a particular focus on mechanisms that cause cell/tissue-specific action of the transcription factor. We expect to uncover mechanisms responsible for different HSF1-dependent transcriptional programs in stress-sensitive and stress-resistant cells. In addition, a novel marker(s) for monitoring the pro-death mode of HSF1 activation/activity will be identified that could be used to predict cell response to HSF1 targeted therapies. Finally, new genes regulated by the pro-death variant(s) of HSF1 will be detected (similar to *Pmaip1*).