Regulation of the Heat-Shock Response

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THE HEAT-SHOCK RESPONSE AND THERMOTOLERANCE

The heat-shock response is a conserved reaction of cells and organisms to elevated temperatures (heat shock or heat stress). Whereas severe heat stress leads to cellular damage and cell death, sublethal doses of heat stress induce a cellular response, the heat-shock response, which (a) protects cells and organisms from severe damage, (b) allows resumption of normal cellular and physiological activities, and (c) leads to a higher level of thermotolerance. Crucial to the survival of cells is the sensitivity of proteins and enzymes to heat inactivation and denaturation. Therefore, adaptive mechanisms exist that protect cells from the proteotoxic effects of heat stress. Owing to their sessile lifestyle, the acquisition of higher levels of environmental stress tolerance is of utmost importance to plants. It is not surprising that the heat-shock response is also linked to several other environmental stresses. Furthermore, an increasing number of studies indicate cross-protection between heat stress, dehydration/drought, cold/chilling/freezing, heavy-metal stress, and oxidative stress in plants.

HSPS ARE MOLECULAR CHAPERONES

At the molecular level the heat-shock response is a transient reprogramming of cellular activities featured by the synthesis of HSPs, concomitant with a cessation of normal protein synthesis. HSPs seem to accumulate in a dosage-dependent manner to amounts sufficient to protect cells and to provide a higher level of thermotolerance. In most organisms, the major groups of stress proteins, HSP100, HSP90, HSP70, HSP60, and small HSPs, are represented by a few members of each class. HSPs are functionally linked to the large and diverse group of molecular chaperones that are defined by their capacity to recognize and to bind substrate proteins that are in an unstable, inactive state. All cellular proteins probably have to interact with molecular chaperones at least once in their lifetime, such as during synthesis, subcellular targeting, or degradation. Owing to heat denaturation, the fraction of potential targets for molecular chaperones seems to dramatically increase upon heat stress and, consequently, the cellular chaperone pool has to be replenished. It is not surprising that, except for small HSPs and HSP100, each class of HSPs is matched by one or several HSCs expressed at normal temperatures. Different HSPs may have different functional properties but common to all of them is their capacity to interact with other proteins and to act as molecular chaperones in vitro (for overview, see Boston et al., 1996; Schöffl et al., 1998a, 1998b). The in vivo chaperone function of plant HSPs was recently demonstrated by the protection and reactivation of a luciferase reporter in Arabidopsis cells (Forreiter et al., 1997).

MUTATIONAL ANALYSIS AND GENETIC ENGINEERING

There is a striking correlation between the occurrence of HSPs and acquisition of thermotolerance, but there is little direct evidence for a causal relationship. Mutations would be required that result in a coordinate change in the expression of HSPs to study: (a) the signal pathway from stress to gene, (b) the mechanism of transcriptional regulation, and (c) the role of HSPs in thermotolerance. The effects of mutations in individual heat-shock genes have been investigated in different organisms. Analyses in yeast provided evidence for an important role of HSP104 and a minor, accessory role of HSP70 in thermotolerance (Sanchez et al., 1993). Mutations in Hsp26, the sole gene for a small HSP in yeast (Petko and Lindquist, 1986), overexpression of small HSPs, and antisense approaches in transgenic plants (Schöffl et al., 1987) had no obvious effects on the phenotype. The protective effect of HSPs is sometimes dependent on the physiological conditions of the cell, as has been shown shown by the disruption of a mitochondrial HSP30 gene in Neurospora crassa, which resulted in strains that were less thermostable under certain carbohydrate limitations (Plesovsky-Vig and Brambl, 1995). In other eukaryotes, other groups of HSPs seem to play important roles in thermotolerance; for example, HSP70 overexpression in mammalian cells and in Drosophila melanogaster (for overview, see Morimoto et al., 1990; Welte et al., 1993).

Using genetic engineering of Arabidopsis as a model for higher plants, dominant regulatory mutations were generated that showed a constitutive synthesis of HSPs at normal temperatures (Lee et al., 1995; Prändig et al., 1998). In these transgenic plants the fundamental role of HSPs in stress tolerance is indicated by significantly higher levels of

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Abbreviations: HSC, heat-shock constitutive or cognate protein; HSE, heat-shock consensus element; HSF, heat-shock transcription factor; HSP, heat-shock protein.
The expression of the heat-shock genes encoding the different HSPs in plants is similar to the situation in other eukaryotes, that is, it is primarily regulated at the transcriptional level. The thermonducibility is attributed to conserved cis-regulatory promoter elements (HSEs) located in the TATA-box-proximal 5'-flanking regions of heat-shock genes. The occurrence of multiple HSEs within a few hundred base pairs is a signature of most eukaryotic heat-shock genes. The eukaryotic HSE consensus sequence has been ultimately defined as alternating units of 5'-nGAAn-3'. In plants the optimal HSE core consensus was shown to be 5'-nGAAnTTCnnGAAn-3'. HSEs are the binding sites for the trans-acting HSF, and efficient binding requires at least three units, resulting in 5'-nGAAnTTCnnGAAn-3'. Plant HSF1 from Arabidopsis has been shown to bind consensus tripartite HSE sequences and HSE-containing regions of the D. melanogaster HSP70 promoter (Hübel and Schöffl, 1994; Hübel et al., 1995). Arabidopsis HSF1 expressed in Escherichia coli has been also shown to form trimers in vitro. Trimerization is required for efficient DNA binding and is a key step in regulating HSF activity in metazoans.

The importance of HSE for heat-dependent transcriptional regulation in plants has been verified by promoter deletions and by the capacity of synthetic HSE sequences, integrated in a truncated cauliflower mosaic virus 35S promoter, to stimulate heat-inducible reporter gene expression in transgenic tobacco (Schöffl et al., 1989).

In addition to HSEs, a number of sequence motifs were found to have quantitative effects on the expression of certain heat-shock genes. In plants there is evidence for involvement of CCAAT-box elements, AT-rich sequences, and scaffold-attachment regions (Czarnecka et al., 1989; Rieping and Schöffl, 1992; Schöffl et al., 1993). These data suggest that sequences affecting the chromatin structure may be important for efficient access of transcription factors (e.g. the TATA-box binding protein) and/or the transcriptional activator proteins (e.g. HSF). The following model integrates the current knowledge about the activation of heat-shock gene expression: The binding of a chromatin-modifying factor, e.g. the GAGA-sequence binding factor (Giardina et al., 1992; Tsukijama et al., 1994), or scaffold attachment affects chromatin structure in a way that provides TBP access to the TATA box, which is a prerequisite for subsequent assembly of the basal transcription complex. In this “stand-by” mode, heat-shock genes are primed for transcriptional activation upon heat stress, and this is mediated by the trimerization and binding of HSF to the HSE sequences.

In many organisms, including plants, the expression of heat-shock genes is not only triggered by a number of environmental stresses but also by developmental cues. In plants certain stages of male gametogenesis and embryogenesis are accompanied by an accumulation of HSPs. This suggests that the requirements for protein chaperoning and catabolism are altered during development, and this alteration is compensated for by the induction of heat-shock gene expression.

In this paper we try to relate recent progress in studying plant HSF gene structure, modification, transgenic expression, and developmental regulation to other eukaryotic systems, and to draw a picture about the possible molecular mechanisms and pathway of regulation and signaling in plants.

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**THE REGULATION OF HSF**

A Conserved Mechanism of HSF Activation

In response to heat stress, HSF of higher eukaryotes is converted from a monomeric to a trimeric form capable of high-affinity binding to HSE and transcriptional activation. In *Saccharomyces cerevisiae* and *Kluyveromyces lactis*, HSF is bound to heat-shock promoters in the absence of stress, indicating that the primary level of regulation involves the acquisition of trans-activating competency (Mager and De Kruijff, 1995; Wu, 1995).

Comparative DNase I footprinting analysis using *D. melanogaster* DmHSF and Arabidopsis AtHSF1 revealed an almost identical pattern of protected sequences comprising the HSE-containing region of a *D. melanogaster* heat-shock promoter. However, differences in the patterns of DNase-I-hypersensitive sites flanking the protected region suggest differences in the conformation of the DNA-to-protein interaction between *D. melanogaster* and Arabidopsis HSFs (Hübel et al., 1995). However, these subtle differences in DNA recognition do not interfere with the conservation of mechanism exemplified in the regulation of gene expression via the *D. melanogaster* hsp70 promoter in plants (Spena et al., 1985) or in the ability of transiently expressed Arabidopsis AtHSF1 to activate heat-shock gene expression in *D. melanogaster*, albeit constitutively, at normal temperatures (Hübel et al., 1995).

Domain Structure of HSF

Similar to vertebrates, all plant species investigated so far contain multiple HSFs, in contrast to the single HSF genes reported for yeast and *D. melanogaster*. To date, four HSFs have been described from Arabidopsis (Hübel and Schöffl, 1994; Nover et al., 1996; Prändl et al., 1998), six from soybean (Czarnecka-Verner et al., 1995), three from tomato (Scharf et al., 1990), and three from maize (Gagliardi et al., 1995). Molecular masses of plant HSFs are in the range of 31.2 to 57.5 KD. Based on sequence homology and domain structure, plant HSFs can be subdivided in the two classes, A and B (Nover et al., 1996). Structural features of plant HSFs, exemplified for Arabidopsis HSF1, HSF3, and HSF4, are compared with the sole HSF of *D. melanogaster* in Figure 1A. The DNA-binding domain and the oligomeriza-
tion domain are located in the N-terminal region of HSF (Fig. 1A). Both domains are conserved in primary structure throughout the HSF protein family. Other regions show significant homology only between closely related HSFs. Nuclear localization signals, hydrophobic heptad repeats localized in the C-terminal region, and activation domains have been identified by functional studies in several HSFs (for overview, see Mager and De Krujiff, 1995; Wu, 1995), including those from tomato (Treuter et al., 1993; Lyck et al., 1997).

**DNA-Binding Domain**

HSFs carry a conserved DNA-binding domain consisting of an antiparallel four-stranded $\beta$-sheet packed against a bundle of three $\alpha$-helices, as determined for HSFs from K. lactis, D. melanogaster, and tomato (for overview, see Mager and De Krujiff, 1995; Wu, 1995; Nover et al., 1996). The second and the third helices form a typical helix-turn-helix motif, with the third helix establishing specific nucleic acid contacts with the HSEs. A distinguishing feature of unknown significance between nonplant and plant HSFs is an 11-amino acid deletion in a solvent-exposed loop between two $\beta$-sheets in plant HSFs.

**Oligomerization Domain**

The oligomerization domain is characterized by a hydrophobic-repeat region A/B, which is separated from the DNA-binding domain by a linker of variable length and sequence. Region A of the hydrophobic repeats is based on a seven-amino acid repetition of hydrophobic amino acids, whereas region B is composed of two overlapping seven-amino acid repeats. In class-A plant HSFs, these arrays are separated by three seven-amino acid repeats, whereas plant HSFs of class B lack this subdomain. It is assumed that the function of the hydrophobic-repeat A/B region is to allow homotrimer formation through a triple-stranded, $\alpha$-helical coiled-coil structure (for overview, see Mager and De Krujiff, 1995; Wu, 1995; Nover et al., 1996).

In higher eukaryotes the formation of trimeric HSFs requires heat stress, but how is the suppression of HSF trimerization achieved under nonstress conditions? The C-terminal hydrophobic repeats is involved in the regulation of trimerization of animal HSFs, in which mutations in this region lead to constitutive trimerization and DNA-binding capacity for D. melanogaster HSF, chicken HSF1 and HSF3, and human HSF1 (Nakai and Morimoto, 1993; Rabindran et al., 1993; Zuo et al., 1994). Although the
C-terminal hydrophobic region is well conserved in animal HSFs, it is poorly conserved in plant and yeast HSFs. A model proposes that intramolecular coiled-coil interactions between the hydrophobic regions A/B and C suppress trimer formation under normal growth conditions; however, deletion mapping of D. melanogaster HSF has revealed larger portions of HSF involved in the negative control of trimer formation (Orosz et al., 1996). The role of the C-terminal hydrophobic repeats has not been established for plant HSFs.

**Nuclear Localization**

HSFs carry two clusters of basic amino acids that have been proposed to function as nuclear localization sequences. A highly conserved cluster of basic amino acids is located at the C terminus of the DNA-binding domain, and a second cluster resides C-terminally from the A/B hydrophobic region (Sheldon and Kingston, 1993; Wu, 1995). In functional studies with two class-A tomato HSFs, the more C-terminal nuclear localization sequence was found to be exclusively required for nuclear import (Lyck et al., 1997). In contrast, vertebrate HSFs require either both or only the N-terminal nuclear localization sequence for translocation. It has been shown that the nuclear localization sequence is sufficient for stress-induced nuclear entry, supporting the view that nuclear import is one layer of HSF regulation by stress (Zandi et al., 1997).

**Activation Domain**

The activation domains of HSFs of higher eukaryotes are localized C-terminally, whereas the HSFs of S. cerevisiae and K. lactis carry activation domains at C- and N-terminal sites of the protein (for overview, see Mager and De Krujiff, 1995; Wu, 1995; Nover et al., 1996). The activation domains of human HSF1 and D. melanogaster HSF show limited sequence identity and are rich in hydrophobic and acidic amino acids (Newton et al., 1996; Wisniewski et al., 1996). In yeast HSF is assumed to be regulated primarily at the level of trans-activating competence. A specific amino acid in the DNA-binding domain, hydrophobic region B, and a yeast-specific control element (CE2) have been shown to be involved in the repression of the activation domain under nonstress conditions (Bonner et al., 1992; Chen et al., 1993). Close inspection of amino acid sequences in the C-terminal part of tomato HSFs suggests that aromatic, bulky hydrophobic, and acidic residues may play a role in transcriptional activation (Treuter et al., 1993). Similar clusters are also present in other HSFs and other transcription-activator proteins (for overview, see Nover et al., 1996).

**REGULATORS OF HSFs ACTIVITY**

**Negative Regulation of HSFs by HSP70**

There is genetic evidence for an autoregulation of the heat-shock response in E. coli, yeast, and higher eukaryotes (for overview, see Mager and De Krujiff, 1995; Wu, 1995). In S. cerevisiae, mutations in two constitutively expressed HSC70/HSP70 genes activate a β-galactosidase reporter gene in an HSE-dependent manner but in the absence of heat stress (Boorstein and Craig, 1990). These data suggest that HSF activity is regulated by HSP70 directly or indirectly. According to the chaperone-titration model, the pool of free HSC70/HSP70 is depleted during heat shock due to binding of HSC70/HSP70 to unfolded proteins, thereby relieving the repression of HSC70/HSP70 on HSF. In a negative feedback loop, the synthesis of excess levels of HSP70 shuts off HSF activity and, consequently, the heat-shock response. With respect to trimer formation, HSC70/HSP70 may maintain HSF in a monomeric state or may participate in the disassembly of trimeric HSFs. Stoichiometric complexes between nonactivated HSF1 and HSP70 have been described previously, as well as inhibition of heat activation of HSF1 in mammalian cells that transiently overexpress HSP70 (Baler et al., 1996).

In plants there is also genetic evidence for a negative regulation of HSF activity and feedback control. Arabidopsis HSF1 is repressed under nonstress conditions and trimerizes upon heat shock. A heat-stress-independent derepression of Arabidopsis HSF1 was obtained by constitutive overexpression of HSF1-GUS fusion proteins (Lee et al., 1995). The molecular mechanism of derepression is still unknown but seems not to be restricted to GUS fusions of HSF. The conformation of the fusion protein may be inaccessible to a negative regulatory molecule, or overexpression of this protein may titrate a transacting negative regulator. It is interesting that, unlike AtHSF1, overexpression of AtHSF3, another Arabidopsis HSF, appears to be sufficient for derepression of the heat-shock response in transgenic Arabidopsis (Prändl et al., 1998). On the other hand, overexpression of AtHSF4 (a class-B HSF) or AtHSF4-GUS fusion proteins in transgenic Arabidopsis was not sufficient to derepress the synthesis of HSFs at normal temperatures (Prändl et al., 1998).

Arabidopsis HSF1 shows also a constitutive DNA binding upon heterologous expression in D. melanogaster and human cells and was able to activate transcription of a suitable reporter gene in D. melanogaster (Hubel et al., 1995). Thus, the negative control of HSF in homologous plant cells seems to depend on a factor that is obviously absent in cultured animal cells. Involvement of HSP70 as a negative regulator of HSF in Arabidopsis is indicated by the analysis of transgenic Arabidopsis plants carrying a heat-inducible HSP70 antisense gene (Lee and Schöffl, 1996). In antisense plants, endogenous HSC70/HSP70 levels are reduced, and during the recovery from heat shock, HSF1 trimer formation is present longer than in control plants.

**Negative Regulation of HSFs by Phosphorylation**

Phosphorylation has been proposed to play a role in activation and inactivation of HSFs (for overview, see Mager and De Krujiff, 1995; Wu, 1995). However, recent functional studies suggest that phosphorylation is primarily involved in repression of HSF. In yeast phosphorylation of CE2-adjacent Ser residues has been shown to enhance deactivation of HSF after heat shock (Hoj and Jakobsen, 1994). In human cell cultures HSF1 is phosphorylated at normal
growth temperatures at two Ser residues in the regulatory domain that modulate the activation domain. These two Ser residues are involved in maintaining human HSF1 in the repressed state under basal conditions (Kline and Moyer, 1997). Phosphorylation of these residues is increased upon stimulation of the Raf/ERK pathway, a mitogen-activated protein kinase pathway responsive to growth factors, and results in inhibition of HSF1 activity in mammalian cells (Chu et al., 1996; Knauf et al., 1996).

In plants phosphorylation of HSF has been demonstrated for recombinant AtHSF1 in extracts of Arabidopsis suspension-cultured cells. AtHSF1 became phosphorylated at Ser residues and, consequently, its capacity for HSE binding decreased. Immunological characterization of the kinase activity has identified CDC2a kinase, a cyclin-dependent kinase regulating the cell cycle (Reindl et al., 1997).

Therefore, in human cells as well as in Arabidopsis, phosphorylation of HSF through various kinases may integrate growth signals. As yet it is unknown whether cyclin-dependent kinases are involved in HSF phosphorylation in animals or whether mitogen-activated protein kinases play a role in HSF regulation in plants. It is conceivable that in growing cells phosphorylation of HSF is required for repression of the heat-shock response that might otherwise interfere with proliferation. This interpretation is supported by growth inhibition of D. melanogaster cells overexpressing HSP70 at normal temperatures (Feder et al., 1992).

**DEVELOPMENTAL REGULATION OF THE HEAT-SHOCK RESPONSE**

**Expression of Small HSPs in the Absence of Environmental Stress**

Induction of heat-shock gene transcription, independent of environmental stress, is evident during meiosis in various organisms. In maize, mRNAs of ZmHsp18–1 and ZmHsp18–9 accumulate during meiosis and at the binucleate stage of the gametophyte, but with different timing of maximal expression (Atkinson et al., 1993). A third gene encoding a small HSP (ZmHsp18–3) is not expressed at all. Recently, HSPs of different classes have been verified in maize microspores (Magnard et al., 1996).

Expression of heat-shock genes occurs during embryogenesis from somatic cells, microspores, and developing pollen in alfalfa and tobacco (Györgyey et al., 1991; Zársky et al., 1995). Changes in concentrations of artificial phytohormones, heat shock, and starvation are known inducers of somatic or microspore embryogenesis. Despite these largely different conditions, microspore-derived embryos from tobacco and somatic embryos from alfalfa express small HSPs during the globular and heart stages but not during the following torpedo stage. These data raise the question of whether heat-shock gene expression during early somatic embryogenesis is a general phenomenon that is also relevant to zygotic embryogenesis.

In zygotic embryos expression of heat-shock genes occurs during the maturation stage of the seed, when cell division has ceased and seeds adapt to desiccation and long-term survival. In sunflower, expression of class II small HSPs seems to parallel roughly storage protein and lipid accumulation, whereas expression of class I coincides with seed desiccation (Coca et al., 1994). It has been proposed that HSPs are important for desiccation tolerance of the embryo or are required for germination upon rehydration. Similar to other plants, Arabidopsis accumulates a specific set of HSPs (AtHSP17.4 and AtHSP17.6) during seed maturation, whereas AtHSP18.2 is not expressed (Wehmeyer et al., 1996).

The expression of subsets of heat-shock genes during gametogenesis and embryogenesis suggests that the developmentally expressed HSPs serve certain functions that may differ to some extent from those required for coping with environmentally stressed vegetative tissue. Furthermore, these findings may indicate differences in the signal-transduction pathway.

**On the Mechanism of Developmental Regulation**

In plants the regulation of developmental expression of HSPs has not yet been investigated in great detail. The analysis of a developmentally regulated soybean heat-shock promoter in transgenic tobacco suggests participation of HSE sequences and, consequently, binding and involvement of HSF (Prándl and Schöffl, 1996). However, it cannot be excluded that other sequences and trans-active factors are involved in seed-specific expression of HSPs. The control of this expression by a developmental program rather than by a stress signal is indicated by the negative effect of the abi3 mutation in Arabidopsis on seed-specific expression of sHSP (Wehmeyer et al., 1996). ABI3, originally identified as an ABA-insensitive mutant allele in Arabidopsis, appears to have a dominant regulatory effect on the developmental expression of heat-shock genes in the embryo.

Recent models for the action of VP1 (Hill et al., 1996; Quatrano et al., 1997), the structural-functional homolog of ABI3 in maize, suggest that VP1 and ABI3 act in the stabilization and activation of regulatory complexes involved in the transcription of target genes. Further investigation of the activation of heat-shock promoters during seed maturation will be required to test the hypothesis that ABI3, directly or via the action of secondary factors, is a regulator of HSF activity. It should be noted that in D. melanogaster, developmental regulation of certain heat-shock genes, such as the expression of HSP82 and HSP26 in oocytes and early larval stages, seems to be regulated by steroid hormones and does not involve an HSE:HSF interaction. In addition, the sole HSF of D. melanogaster plays an essential role at this stage of development, although this function does not appear to be directly related to the expression of HSPs (Jedlicka et al., 1997).

**CONCLUSIONS AND PERSPECTIVES**

Some of the plant responses to heat stress show certain characteristics that are unique to plants, that were originally discovered in plants, or, more importantly, that are
more important to plants than to other organisms. Future research will focus on the roles of HSP100, HSP90, HSP70, and small HSPs in an effort to identify specific determinants involved in protection from the deleterious effects of heat, cold, heavy metal, desiccation, reactive oxygen species, and other stresses in plants.

The regulation of HSF activity and the multiplicity of HSFs in plants are problems of continuing scientific interest. The mechanism of derepression of HSF activity is still not understood. HSF1 protein fusions and HSF3 of Arabidopsis are constitutively active upon transgenic overexpression, suggesting that negative regulation and/or conformational changes are involved in the mechanism of activation (Fig. 1B). Up to six HSF-like genes were identified in plants, including tomato, Arabidopsis, maize, and soybean. The question of whether the genetic redundancy of HSF reflects diversification of functions has to be addressed and answered by future research. It seems possible that some HSFs, classified by the criterion of structural features in the DNA-binding and multimerization domains (Fig. 1A), may work as repressor proteins that counteract transcriptional activation. Preliminary results suggest that this may be true for certain HSFs in subclass B (Fig. 1A) (Czarnecka-Verner et al., 1998). Such proteins could act through DNA binding, either as repressors or through protein:protein interaction as modulators of HSF activity.

Is there a signal pathway that senses stress from external sources and triggers the heat-shock response via HSF? Components in the pathway upstream of HSF are not yet known. It is conceivable that HSF itself or its interaction with HSC70 and other proteins (Fig. 1B) is the sensor of heat stress and results in an activation of HSF via conformational changes involving monomer-to-trimer transition, nuclear targeting, DNA binding, and transcriptional activation. An alternative model for temperature sensing and regulation of the heat-shock response integrates observed membrane alterations (for overview, see Wu, 1995; Carratì et al., 1996).

Developmental signaling seems to be responsible for the expression of HSPs during seed maturation. The involvement of HSF is indicated by the dependence of HSE promoter sequences, and signaling through ABA pathways is suggested by the negative effect of an abi3 mutation in Arabidopsis (Fig. 1B). However, neither the responsible HSF nor the level of control by ABI3 has been identified. ABA does not seem to be involved in microspore development, so it can be concluded that this pathway is probably not involved in the meiosis-dependent activation of heat-shock gene expression. Yet another pathway may exist that integrates signals of cell proliferation and results in cell-cycle-dependent phosphorylation of HSF via Cdc2a (Fig. 1B). It will be important to find out whether HSF phosphorylation also occurs in vivo, whether phosphorylation blocks the activity of native HSF, and whether HSF has as-yet-unknown biological functions in cell growth and development. Although up to now the cellular targets of developmentally expressed HSPs are unknown and no mutants in the developmental heat-shock response are available, the current data suggest that HSFs are important to cells in certain stages of development.

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LITERATURE CITED


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