Mechanisms and Roles of Small Heat Shock Proteins in Protein Folding

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Abstract

Small heat shock proteins (sHSPs; ~12–43 kDa) are ATP-independent molecular chaperones that act as first responders to proteotoxic stress. They bind non-native proteins, prevent irreversible aggregation, and triage clients for refolding or degradation. Their activities emerge from highly dynamic oligomer assemblies regulated by post-translational modification, hetero-oligomerization, and environmental signals. sHSPs cooperate with ATP-dependent systems—principally Hsp70 (and Hsp100 in bacteria, fungi, and plants)—to disaggregate and refold proteins after stress. Dysregulation of sHSP function is implicated in neurodegeneration, cardiomyopathy, cataract, and cancer. Here, we summarize current understanding of sHSP structure, client recognition and sequestration, interplay with other chaperones, and emerging concepts including phase-separation-like condensates and therapeutic targeting.

Keywords: sHSP, Protein,

Introduction

Proteins are fundamental biomolecules that perform a vast array of structural, enzymatic, and regulatory functions in all living organisms. To achieve their functional states, proteins must fold into precise three-dimensional conformations, a process that is inherently error-prone and sensitive to environmental fluctuations. Misfolding or aggregation of proteins can disrupt cellular homeostasis, leading to proteotoxic stress and contributing to various diseases such as neurodegeneration, cardiomyopathies, and cataracts. To counteract these challenges, cells have evolved an intricate molecular chaperone network that safeguards protein folding and maintains proteostasis. Proteostasis relies on coordinated chaperone networks that assist folding, suppress aggregation, and clear damaged proteins. sHSPs are ubiquitous across bacteria, plants, and animals and are rapidly induced under conditions such as heat shock,

oxidative stress, acidosis, and proteotoxic insults. Unlike Hsp70/Hsp90, sHSPs function without ATP, forming metastable complexes with partially unfolded clients—the “holdase” role—thereby preserving refoldability. Their oligomeric plasticity underpins substrate recognition breadth and regulatory control. In this review, we focus on the mechanisms by which sHSPs assist protein folding, their interaction partners, and the diverse roles they play in maintaining cellular homeostasis under stress and physiological conditions. Understanding these mechanisms not only provides insight into fundamental aspects of proteostasis but also opens avenues for therapeutic interventions in protein misfolding disorders.

Evolution and Diversity

sHSP families have expanded and diversified across taxa via gene duplication and specialization. Duplicate divergence in bacteria illustrates subfunctionalization in client recognition and stress responses, while in mammals the 10 HspB paralogs (HspB1–B10) exhibit tissue-specific expression and inducibility. Small heat shock proteins (sHSPs) are an ancient and evolutionarily conserved family of molecular chaperones found in all domains of life, from bacteria to plants and animals. Despite their relatively small size and lack of ATPase activity, sHSPs display remarkable structural and functional diversity, reflecting adaptations to organism-specific requirements and environmental pressures. At the core of their architecture lies the conserved α-crystallin domain, a structural hallmark that provides stability and substrate-binding capability. Flanking this domain, the N- and C-terminal extensions vary significantly among species, contributing to differences in oligomerization, substrate specificity, and regulatory mechanisms. This variability underlies the functional versatility of sHSPs across taxa. In prokaryotes, sHSPs often exist as single or few isoforms that protect essential enzymes during thermal or oxidative stress. In contrast, eukaryotes typically encode multiple sHSP isoforms, many of which are tissue-specific. For example, plants possess large sHSP families localized in different cellular compartments, reflecting their sessile lifestyle and constant exposure to abiotic stresses. In vertebrates, specialized sHSPs such as αA- and αB-crystallins maintain lens transparency and protect muscle and neuronal tissues. The evolutionary expansion and diversification of sHSPs highlight their crucial roles in proteostasis, stress adaptation, and organismal survival, while also linking their dysfunction to various human diseases.

Architecture and Oligomer Dynamics

All sHSPs share a conserved α-crystallin domain (ACD) flanked by variable N-terminal and C-terminal regions that mediate oligomerization and client binding. Oligomers exist as polydisperse ensembles (e.g., 12–48-mers) that interconvert on biologically relevant timescales; stress, pH, ionic strength, and phosphorylation shift these equilibria toward more active, client-binding subpopulations. Hetero-oligomerization (e.g., HspB1/HspB5) further tunes chaperone activity and client scope. Small heat shock proteins (sHSPs) possess a conserved α-crystallin domain flanked by variable N- and C-terminal regions, enabling structural plasticity. They assemble into large, dynamic oligomers that undergo constant subunit exchange. Stress triggers oligomer remodeling, exposing substrate-binding sites and enhancing chaperone activity, thereby stabilizing unfolding intermediates and preventing irreversible protein aggregation.

Recognition and Sequestration

sHSPs preferentially engage destabilized intermediates via multiple low-affinity, multivalent contacts distributed across flexible N-terminal regions and exposed ACD interfaces. Recent single-particle and integrative studies indicate "client-induced remodeling," where binding polarizes and locally destabilizes sHSP subunits to grow protective assemblies around clients—stabilizing them in a refolding-competent state ("sequestration"). sHSPs recognize partially unfolded or misfolded proteins through exposed hydrophobic regions. They rapidly bind and sequester these non-native substrates into stable, soluble complexes, preventing aggregation. This sequestration maintains substrates in a refolding-competent state, enabling subsequent transfer to ATP-dependent chaperones such as HSP70 for proper folding and functional restoration.

Regulation of Activity

Phosphorylation (e.g., HspB1 Ser15/78/82; HspB5 Ser19/45/59) modulates oligomer size and exchange, generally favoring smaller, more active species with increased client affinity; redox modifications and metal ions can also alter activity. Stress-specific cues and client load determine ensemble distribution and thereby functional output. sHSP activity is tightly regulated by post-translational modifications, oligomeric rearrangements, and cellular

conditions. Phosphorylation modulates oligomer size and substrate affinity, enhancing chaperone function under stress. Redox state and pH also influence dynamics, while interactions with partner proteins fine-tune activity, ensuring effective protein quality control and adaptation to environmental challenges.

Cooperation with Hsp70 (± Hsp100) Systems

After stress subsides, sHSP-bound clients are transferred to Hsp70 co-chaperones for iterative unfolding and refolding; in bacteria, fungi, and plants, Hsp100 (ClpB/Hsp104) collaborates with Hsp70 to thread and disentangle aggregates. Genetic and biochemical evidence supports a functional triad: sHSPs organize and solubilize misfolded proteins; Hsp70/Hsp40 coat and remodel; Hsp100 powers extraction/disaggregation. Metazoan cytosol lacks Hsp100, relying on Hsp70-based disaggregation with lower maximal capacity. sHSPs cooperate with Hsp70 (and, in some organisms, Hsp100) systems to ensure efficient protein quality control. sHSPs first capture unfolding substrates in stable complexes. These are then transferred to Hsp70, which, with ATP hydrolysis and co-chaperones, refolds proteins. In bacteria and yeast, Hsp100 collaborates to resolubilize aggregated substrates.

Pathophysiology and Therapeutic Targeting

Altered sHSP expression and mutations are linked to cataract (HspB4/HspB5), desmin-related and cardiomyopathies (HspB5), neuropathies and myopathies (HspB1/HspB8), and cancer, where sHSPs can confer apoptosis resistance and therapy tolerance. Pharmacological strategies include modulating phosphorylation, disrupting pathogenic sHSP–client interactions, or enhancing protective functions. Dysfunction or mutations in sHSPs are implicated in diverse pathologies, including cataracts, neurodegenerative disorders, cardiomyopathies, and skeletal myopathies, largely due to impaired chaperone activity or toxic oligomer formation. Aberrant regulation of sHSPs can disrupt proteostasis and cellular resilience to stress. Therapeutic strategies aim to modulate sHSP expression, stabilize protective oligomers, or enhance interactions with partner chaperones. Pharmacological inducers, gene therapies, and engineered variants hold promise for targeting sHSP-related diseases and restoring protein homeostasis.

Conclusion

sHSPs are central to early proteotoxic defense, capturing unfolding proteins in dynamic, regulatable assemblies that preserve refolding potential. Their collaboration with Hsp70 (± Hsp100) completes the rescue cycle, and their misregulation reverberates across disease contexts. New mechanistic work—especially on client-induced remodeling and condensate biology—offers routes to rational therapies that either amplify cytoprotective functions or blunt pathological interactions. Small heat shock proteins are versatile ATP-independent chaperones central to proteostasis. Through dynamic oligomerization, substrate sequestration, and cooperation with other chaperones, they safeguard cells against stress-induced damage. Their evolutionary diversity highlights specialized functions across organisms, while their involvement in human diseases positions them as promising targets for therapeutic interventions.

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