IUBMB *Life*, 60(1): 10–18, January 2008

Critical Review

Chaperones as Integrators of Cellular Networks: Changes of Cellular Integrity in Stress and Diseases

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Summary

The complex integrity of the cells and its sudden, but often predictable changes can be described and understood by the topology and dynamism of cellular networks. All these networks undergo both local and global rearrangements during stress and development of diseases. Here, we illustrate this by showing the stress-induced structural rearrangement of the yeast protein–protein interaction network (interactome). In an unstressed state, the yeast interactome is highly compact, and the centrally organized modules have a large overlap. During stress, several original modules became more separated, and a number of novel modules also appear. A few basic functions such as the proteasome preserve their central position; however, several functions with high energy demand, such the cell-cycle regulation loose their original centrality during stress. A number of key stress-dependent protein complexes, such as the disaggregation-specific chaperone, Hsp104 gain centrality in the stressed yeast interactome. Molecular chaperones, heat shock, or stress proteins became established as key elements in our molecular understanding of the cellular stress response. Chaperones form complex interaction networks (the chaperome) with each other and their partners. Here, we show that the human chaperome recovers the segregation of protein synthesis-coupled and stress-related chaperones observed in yeast recently. Examination of yeast and human interactomes shows that chaperones 1) are intermodular integrators of protein–protein interaction networks, which 2) often bridge hubs and 3) are favorite candidates for extensive phosphorylation. Moreover, chaperones 4) become more central in the organization of the isolated modules of the stressed yeast protein–protein interaction network, which highlights their importance in the decoupling and recoupling of network modules during and after stress. Chaperone-mediated evolvability of cellular networks may play a key role in cellular adaptation during stress and various polygenic and chronic diseases, such as cancer, diabetes or neurodegeneration.
IUBMB *Life*, 60(1): 10–18, 2008

Keywords Alzheimer's disease; cancer; chaperones; diabetes; evolution; evolvability; heat shock proteins; networks; Parkinson's disease; stress.

Received 7 October 2007; accepted 9 October 2007
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ISSN 1521-6543 print/ISSN 1521-6551 online
DOI: 10.1002/iub.8

CELLULAR NETWORKS: TOPOLOGY AND DYNAMISM

Structural Properties of Cellular Networks

The network approach dissects the cellular complexity to various elements (also called as vertices, see Table 1), such as proteins, cytoskeletal filaments, cellular organelles, signaling components, or enzyme reactions, and tries to catalog those interactions of these elements, which have a relatively high affinity, and, therefore, are measurable with our current, "traditional" biochemical or high-throughput methods. The interactions (also called as links or edges) often have weights, which reflect their affinity, propensity or probability, and directions, which become especially important in signaling and metabolic networks [Fig. 1; (*1–3*)].

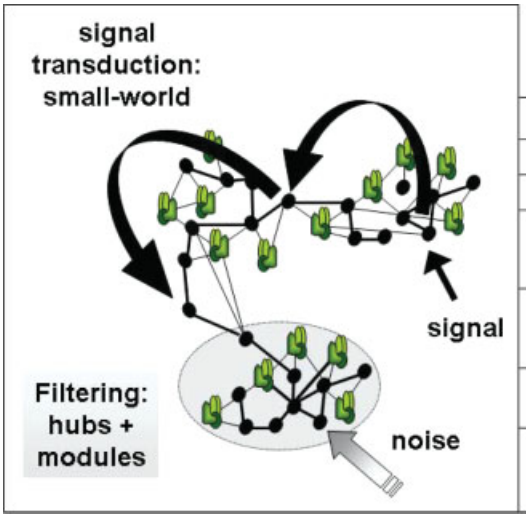
Cellular networks often form small worlds, in which two elements of the network are separated by only four to five other elements in an average (*4*). This proverbial "six steps of separation" is a key feature to limit the distortion of the information, which becomes unmanageable after six transferring steps (*5, 6*). "Information" in the cellular context is often a conformational change of the participating proteins or other macromolecules, such as RNAs. Networks of our cells contain hubs, that is, elements, which have a large number of neighbors. These networks can be dissected to overlapping groups or communities [which we will call modules in this review; (*7–10*)]. Both hubs and network modules are efficient in screening and filtering of the extensive

Table 1
A glossary of network-specific expressions

Expression	Short explanation
Centrality	Centrality of a network *element* or an interaction defines relative importance of the *element* or the interaction within the network (for example, how important a person is within a social network or a protein in a cellular network). There are various measures of centrality in network analysis giving numerical figures to characterize this importance from the local structure of the network interactions, global properties of the whole network or both. In this work, we refer to the latter, that is, complex understanding of centrality, which takes into account all levels of network structure.
Chaperome	The protein–protein interaction network of molecular chaperones with each other and with their targets. The chaperome is a segment of the *interactome*, which contains all protein–protein interactions in the respective cell type.
Element (node, vertex or vertices)	The element is a single building block of a network. The element is also called a vertex in graph theory, site in physics, or actor in sociology. Most of the times, the element itself is a complex network again, such as the elements of cellular protein–protein interactions networks, the individual protein molecules can be perceived as networks of their constituting amino acids or atoms.
Fractal	Fractal objects are generated by recursive process, in which self-similar objects of different size are repeated and repeated again. In nature, we are often talking about fractal-like behavior, in which the extent of self-similarity is not complete as in pure (and many times extremely beautiful) mathematical fractals.
Hierarchical network	A hierarchical organization arises in a network, when an *element* has a "parent" and this "parent" also has a "grandparent," like in a family tree. Networks may contain more subtle hierarchies, wherein network *modules* form a network, where again, network *modules* can be defined, which also form a network, etc.
Hub	A hub is a highly connected *element* of the network. Usually, a hub has more than 1% of total interactions.
Hub-repulsive	A network is called as hub-repulsive, when its *hubs* are connected to each other with a smaller probability than in a network, which contains the same number of *hubs*, but they are connected randomly. The opposite of a hub-repulsive network contains a *rich-club*.
Interactome	The protein–protein interaction network of a respective cell type. Please note that the interactome always contains data that were averaged from a high number of individual experiments involving myriads of cells. We do not have yet the means to uncover the interactome of a specific, single cell.
Modules (network communities, groups)	Modules are groups of network *elements* that are relatively isolated from the rest of the network, and where the *elements* inside the module are functionally and/or physically linked to each other.
Rich-club	A network has a rich-club, when its *hubs* are connected to each other with a higher probability than in a network, which contains the same number of *hubs*, but they are connected randomly. The opposite of a rich club-containing network is a *hub-repulsive* network.

information a cell receives and generates in each second. Hubs can transmit only a minority of the continuously bombarding pieces of information at a given time. Network modules, by definition, have denser intramodular connections than intermodular contacts, therefore, keeping the incoming information "trapped" inside the module and allowing its preferential passage to the next module only in special cases, when the network has already been trained to provide a fast transmission of that particular change by previous experience. In this sense, all our cells have a unique and special "personal history," which developed their network configuration to its current status [Fig. 1; (*6*)].

Cellular networks and their modules often form a hierarchical structure. These "chain-of-command-type" or pyramid-type organizations are very useful to allow a fast and efficient integration of signaling steps [*e.g.*, in transcription factor networks; (*11*)] and are helpful in the compartmentalization of cellular metabolism (*12, 13*). Hierarchical networks often display self-similarity, which makes them fractal-like objects (*14*). Cellular networks may possess a "rich-club," in which the hierarchy is configured via preferential direct interactions between hubs, or just oppositely, may be "hub-repulsive," in which hubs are separated from each other by other elements. The former is often



Network	Elements	Links
Interactome	Proteins	Interactions
Cytoskeleton	Filaments	Interactions
Organelle network	Membranes, organelles	Proteins, membranes
Signalling network	Proteins, siRNA	Specific interactions
Metabolic network	Metabolites	Enzymes
Gene transcription network	Transcription factors and DNA	Interactions

Figure 1. Cellular networks and their major functions: information propagation by small worldness and filtering by hubs and network modules.

characteristic to transportation networks, such as metabolic networks, whereas the latter is often typical to structural networks, such as protein–protein interaction networks (*15, 16*). To add to the complexity of the phenomenon, protein–protein interaction networks may reveal a rich-club phenomenon at an intermediate, but at neither low nor high levels of their hierarchy, which may reflect their intermediate state between an information transportation network and a structural network (*17*).

At the end of the initial view, we must warn the readers that this field is new. Cellular networks have been uncovered only in the last 8 years (*1–3*), which may sometimes lead to overgeneralizations led by the joy and excitement of a novel understanding of cellular complexity. Moreover, most of our current methods allow only a sampling of the cellular networks resulting in an average network topology from millions of individual cells with opposing "personal histories." Therefore, each statement needs to be validated through critical scrutiny of the datasets, sampling procedures and methods of data analysis at each network examined (*18, 19*). As an example of the controversies, which may arise during our current, initial status of understanding, we cite the current "stratus-(alto)cumulus debate" (*20–23*), where opinions differ, whether the overall organization of the *Saccharomyces cerevisiae* protein–protein interaction network connects network modules by their central, denser core leading to a compact organization, which resembles to the stratus-type clouds, or yeast network modules are connected via their peripheral layers resulting in a more structured organization resembling the cumulus-type clouds.

Dynamic Changes of Cellular Networks

Cellular networks display an extremely high dynamism. Not only their links are often rearranged, but also because of the synthesis and proteolysis of cellular proteins, a large amount of network elements is continuously vanished and reappeared again. An initial and currently often-quoted example of this scenario is the existence of date hubs and party hubs, in which the date hubs form complexes with different subsets of their partners at different times and cellular locations, whereas party hubs collect their partners and form complexes with all of them simultaneously (*20, 24–26*). Date hubs—logically—usually have a single-binding surface, whereas party hubs are multi-interface proteins (*24*). Date hubs contain more disordered regions, whereas party hubs have a larger tendency to form a rich club (*25*). Network modules of the yeast interactome may be dissected to static and dynamic modules, when gene expression changes are taken into account. The pathway structure of static modules is more redundant, which allows a faster evolution and larger tolerance of gene expression noise. On the contrary, dynamic modules help the condition-dependent, flexible regulation of cellular responses (*26*).

Stress may induce a decrease in the strength and number of links, which leads to a gradual detachment of network modules from each other. The overlap decreases between modules leading to simpler, less regulated and more specialized cellular functions (*27, 28*). In a related study, Luscombe *et al.* (*29*) examined the topology of yeast transcriptional signaling subnetworks of 142 transcription factors and 3,420 target genes in five different cellular conditions. The stress response was governed by a simplified subnetwork, which had a shorter diameter and was characterized by large hubs, which probably behaved as integrators of the reprogrammed cellular response. On the contrary, the cell cycle was governed by a highly interwoven, complex structure indicating a multistage internal program (*29*).

The above, stress-induced topological differences were largely recovered, when we compared the overlapping modular

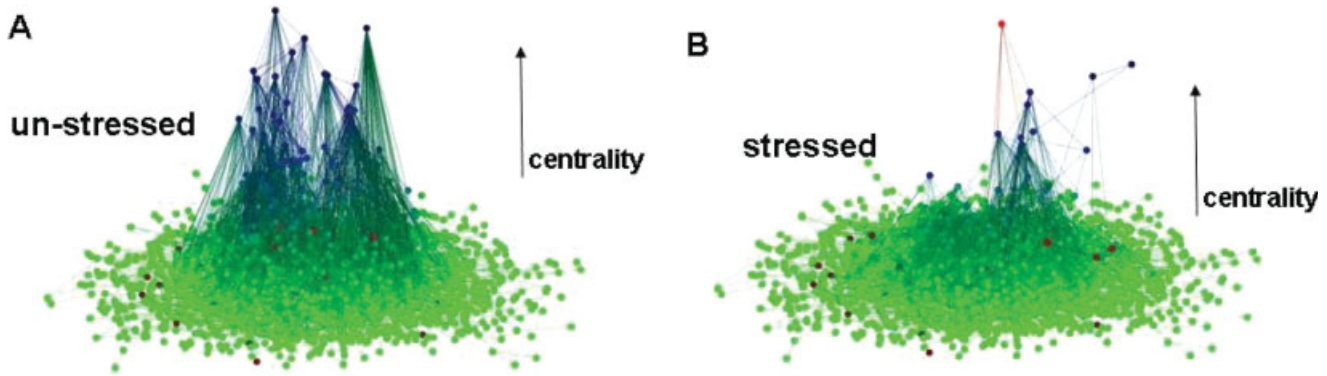


Figure 2. Rearrangements of the yeast interactome, and changes of yeast chaperone positions in stress. The figure shows the yeast protein–protein interaction network using the high-confidence dataset of 2,640 yeast proteins and their 6,600 interactions (*25*). The peaks represent network modules detected by the ModuLand method (*10*), and the vertical position corresponds to the centrality of the given protein. The color scale of light green to dark blue represents increasing centralities, while molecular chaperones were coded by the red color. Colored lines represent the interactions (links) between proteins. Line colors were set according to the color of the end points. The network was visualized with the modification of the Pajek program (*55*). (A) The unstressed yeast interactome. (B) The stressed yeast interactome. Stress was modeled by the readjustment of the uniform link-weight of 1.0 of the unstressed data set taking into account the average of gene expression changes in 65 experiments of 13 diverse stress conditions from the data of Gasch *et al*. (heat shock 5, 15, 30, 40 and 80 min; 37 → 25°C 15, 30, 45, 60, and 90 min; 0.32 mM hydrogen peroxide 10, 30, 50, 80, and 120 min; 1 mM menadione 10, 30, 50, 105, and 160 min; 2.5 mM dithiothreitol 15, 30, 60, 120, and 480 min; 1.5 mM diamide 5, 20, 30, 50, and 90 min; 1 M sorbitol 5, 15, 45, 60, and 120 min; hypo-osmotic shock 5, 15, 30, 45, and 60 min; amino-acid starvation 0.5, 1, 2, 4, and 6 hours; nitrogen depletion 0.5, 2, 8, 24, and 72 hours; diauxic shift 9.5, 11.5, 13.5, 15.5, and 20.5 hours; YPD growth 2, 6, 10, 24, and 72 hours; YPD stationary phase 0.33, 1, 3, 7, and 22 days; as described in detail in ref. *30*). The signed sum of minimum twofold changes in gene expression of the 65 experiments was counted. Weights of respective proteins were assigned as 0.25 or 0.5, if the minimum twofold decrease was observed between 29 and 22, or between 21 and 12 experiments, respectively. Weights were set as 2 or 4, if the minimum twofold increase was observed between 10 and 20, or between 21 and 40 experiments, respectively. Link-weights were calculated as the products of their two endpoint weights.

structure of the yeast protein–protein interaction network using the NodeLand version of our recently developed ModuLand method family [(*10*) and Kovacs *et al*., in preparation] and a high-confidence dataset of 2,444 yeast proteins and their 6,271 interactions (*25*), in which stress was modeled by the readjustment of protein abundance values taking into account the average of gene expression changes in 65 experiments of 13 diverse stress conditions (such as heat shock, oxidative and reductive stress, hypo- and hyper-osmotic stress, diauxic shift, nitrogen, and amino-acid starvation) from the data of Gasch *et al*. (*30*) as described in the legend of Fig. 2. The height of the peaks on Fig. 2 represent the centrality of the given yeast protein in the yeast interactome. Under normal growth conditions ("unstressed state," Fig. 2A), the interactome is highly compact, and the centrally organized modules have a large overlap. On the contrary, in the stressed state (Fig. 2B, which is an interactome reflecting the average changes of protein abundance in 13 specific stressful conditions), modules become separated, and their overlap decreases. In the stressed interactome, peaks (meaning local centralities) appear at several novel positions. Consequently, the unstressed interactome contains much less modules than the stressed interactome (42 compared to 117 modules in the current analysis). This reflects the emergence of specific protein complexes providing the adaptation to one or another specific stressful event. Applying the "stratus/cumulus nomenclature," the unstressed yeast interactome resembles more to a stratus type, whereas the stressed interactome resembles to a cumulus-type organization. This suggestion is strengthened by the fact that the number of hub–hub interactions (in which hub is defined as an element having eight or more weighted interactions after ref. *25*) is decreased to less than half in the interactome of the stressed yeast cell, when compared with the interactome of the yeast cell under normal growth conditions (182 hub–hub interactions *vs*. 494 in the unstressed yeast interactome). This dynamism may explain the variable results obtained before (*20–23*). Furthermore, more detailed examinations are necessary to prove or disproof this assumption.

The central (peak) protein is identical in several modules of the unstressed and stressed interactomes. Modules, which preserve their centrality in both unstressed and stressed conditions, are organized around the proteasome, the nuclear transport com-

plex and actin-regulatory proteins. During stress, the regulatory complex of the proteasome becomes rearranged, and the cell cycle and rRNA synthesis become suppressed, whereas the GCN4 stress-dependent transcription complex, the damaged protein label, ubiquitin, the chemical chaperone, trehalose-synthesizing complex, the disaggregation-specific chaperone, Hsp104, and the stress surviving cAMP-kinase pathway gain centrality in the interactome (Fig. 2 and data not shown). All these rearrangements emphasize the energy-sparing behavior and point toward the activation and emerging centrality of a multifaceted protection machinery of the yeast cell during stress. However, we must warn that the illustrative analysis mentioned earlier took into account approximately the strongest 6% of known, and possibly 3% of total yeast protein–protein interactions. The comparison of unstressed and stressed states probably cancels some of the errors caused by this biased dataset; however, the results certainly do not reflect the whole interaction complexity. Additionally, the illustrative analysis gave an averaged picture of 13 divergent stress conditions, in which individual states may vary. A more detailed analysis (Csermely and coworkers, in preparation) will certainly reveal more details than the initial attempt presented in this review to illustrate the dynamism of modular organization during stress.

The above "simplification/specialization" duo of the yeast interactome during stress resembles to an accelerated and reversible version of the reductive evolution of symbiotic organisms. In this latter scenario, the engulfment by the host provides a safe and stable environment for the "guest," for example, a parasite (*31*). In both processes, major segments of the original networks become attenuated parallel with a specialization of the network structure for a specific set of environmental conditions provided by either the stress or the host. This network simplification gives a more rigid structure, in which most of the original universal and flexible adaptation strategies were temporarily or irreversibly lost.

If the cell experiences an increasing amount of stress, its network may undergo topological phase transitions. Plenty of resources allow a high link-density keeping contact-preference low, and resulting in a random network-type final configuration. During stress, discrimination between network elements, and contact preferences will occur, and increasingly strong hubs will appear. In an extreme case, the network may be switched to a star network, where the "winner hub takes all," and an extremely centralized, highly hierarchical structure develops. With a further reduction of the resources, the star network collapses, and a number of isolated, small subgraphs will be formed. This corresponds to the death of the former gross structure. The latter, disintegration-type topological phase transition may be preceded by quarantining the most damaged modules of the network and might accompany various forms of programmed cell death (*6, 28, 32, 33*). Currently, the above scenario awaits support by experimental evidence. However, the appearance of central, starlike hubs in stress-related subnetworks of yeast transcription factor networks (*29*) supports the possible existence of these rearrangements.

CHAPERONES AS NETWORK INTEGRATORS

Stress provokes the activation and extensive synthesis of molecular chaperones, many of which are also called heat shock proteins, or stress proteins, abbreviated as "Hsp"s. Hsps provide a general response to stress by repairing damaged proteins. Several chaperones are often abbreviated as "Hsc"s, referring to their heat shock cognate protein status. These chaperones are continuously present in the cells and assist in protein synthesis, the unfolding/refolding steps of protein transport and structural rearrangements of, for example, the nucleus, as well as participate in the triggering of various signaling steps by releasing the respective kinases or other signaling proteins. Chaperones never work alone, but form large complexes with each other and with their cochaperones (which we call as chaperome after ref. *37*; *34–39*).

The currently available human chaperome is shown in Fig. 3. The core domain contains 14 chaperone molecules, whereas the periphery has 15 chaperone isoforms. Most of Hsp60 chaperones are in the core, while most of the small heat shock proteins are in the periphery. The Hsp70 and Hsp90 chaperones are divided between the two. Analysis of the individual chaperones reveals that the core domain of the human chaperome corresponds to the yeast "CLIPS-chaperones" (chaperones linked to protein synthesis), whereas the various subgroups of the periphery reflect the functional divergence of "HSP-chaperones" (stress-induced chaperones) as dissected by Albanese *et al*. (*38*). Human CLIPS-like chaperones are mostly cognate proteins, which are either expressed continuously in human cells or are brain/testis-specific isoforms of their major counterparts. Human HSP-type chaperones are involved in the stress response and in the reprogramming the cells in malignant transformation or viral infection. The recovery of the major division of the yeast chaperone arrangement in the human chaperome suggests the evolutionary conservation of this functional distinction.

Chaperones as Intermodular Connections Between Hubs

Molecular chaperones bind and release a large variety of damaged proteins. This is made possible by a large promiscuity in the chaperone-client interactions. Consequently, chaperones form low affinity, dynamic temporary interactions called weak links in cellular networks (*6, 39, 40*). Chaperones have a large number of hubs among their neighbors in the yeast interactome (*40*). The abundance of hubs as chaperone neighbors gives a central position to molecular chaperones in the protein–protein interaction network, which may help the chaperone-mediated crosstalk between divergent cellular pathways. Most importantly, chaperones are intermodular elements of both protein–protein interaction and membrane organelle networks, assembling the modular structure of the cell (*28, 40*). This notion is supported by the observation that chaperones are enriched in date hubs (*26*), pointing toward their integrative role of various cellular functions. The maximal number of potential phosphorylation sites on various chaperones is color coded in Fig. 3.

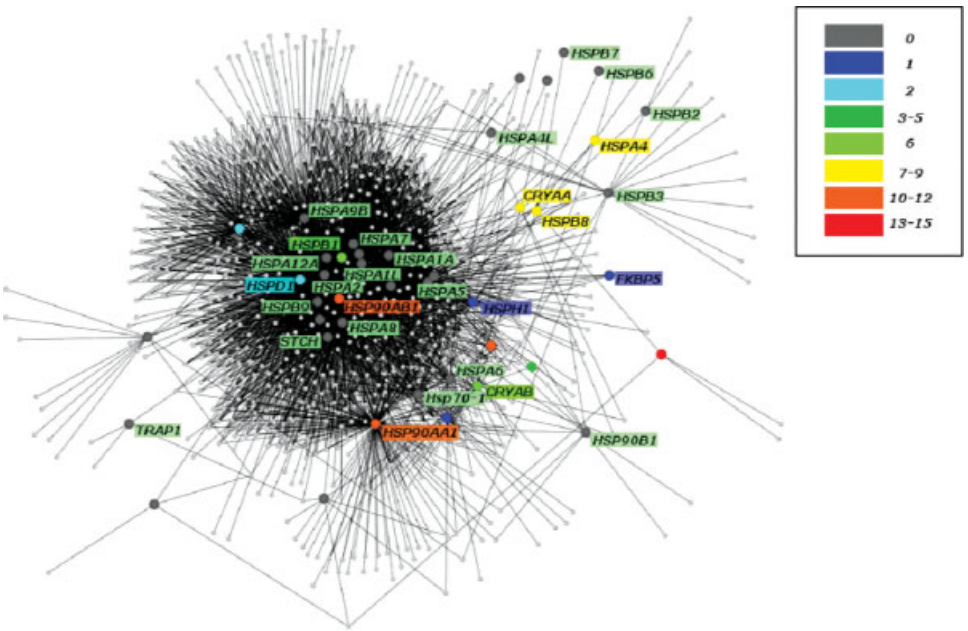


Figure 3. The phosphorylated human chaperome. Interacting neighbors of human molecular chaperones were identified from the 16.09.07. release of the pSTIING database (*56*, pstiing.licr.org). Potential phosphorylation sites were obtained from the same release of the NetworKIN database (*57*, networkin.info). Interacting proteins are marked as white small circles, cochaperones as large grey circles, while chaperones are color-coded according to the number of their potential phosphorylation sites shown in the inset. Grey lines represent protein–protein interactions. The abbreviated names of human molecular chaperones are given with the respective phosphorylation-specific color. The network was visualized with the Pajek program (*55*).

The multitude of interacting kinases and potential phosphorylation sites is an additional piece of evidence indicating the regulatory role of chaperones in the organization of human protein–protein interaction networks.

Role of Chaperones in Stress-induced Network Rearrangements

During stress, chaperones become increasingly occupied by damaged proteins causing a so-called "chaperone overload" (*41*). Chaperone inhibition may lead to a decoupling of network modules both in protein–protein interaction networks and in the mitochondrial-ER organelle network. These provide additional safety measures for the cell, because decoupling of modules may stop the propagation of network damage at the modular boundaries (*6, 28, 42, 43*). Chaperones are marked with red color in Fig. 2. In average, molecular chaperones had an ~20% higher position in the stressed yeast interactome (Fig. 2B) than in the unstressed network (Fig. 2A), indicating an increased chaperone-centrality during stress. In other words, chaperones may gain an even more important role in the connection of various segments of the interactome during stress, which suggests an additional importance of the regulatory role of the "chaperone-overload," that is, the balance between damaged proteins and available chaperones (*41*). The most central protein of the stressed interactome in Fig. 2B is the damaged protein disaggregating chaperone, Hsp104. Hsp104 forms a complex with a number of other chaperones and cochaperones, interacts with the Sup35 yeast prion, associates with the ERAD protein degradation pathway of the endoplasmic reticulum and interacts with the nuclear pore complex. This set of key partners propels Hsp104 as a connector of two of the three most central modules (the proteasome and the nuclear complex), which explains its extreme centrality. Although the role of Hsp104 in Sup35 regulation and in ERAD protein degradation is well known (*44, 45*), the demonstration of the functional role of Hsp104 in the protection of nuclear pore complexes during stress awaits experimental evidence.

Importantly, while this review was under preparation, a comprehensive study of yeast Hsp90 networks under normal growth conditions and elevated temperature (*36*) recovered many key features of our former and current, illustrative analyses showing that 1) Hsp90 neighbors contained a higher than expected number of hubs (*39*), 2) Hsp90 complexes were rather labile pointing out a preponderance of low-affinity interactions (*6, 39, 40*), and 3) stressed Hsp90 network was more diverse and structured than that under normal growth conditions (Fig. 2), where it is centered around transport processes (data not shown).

When the stress is over, the low affinity and promiscuity of chaperone interactions may provide an efficient tool for the remodeling of the modular structure of the reassembling cellular networks. This may be an underlying reason for the regulatory

role of Hsp90 and other chaperones in the evolvability of complex systems (*6, 39, 46*). The recent data of Bobula *et al.* (*47*), which show that a genome-wide mutagenesis with a synthetically harmful mutation screen with the yeast Hsp40/Hsp70 chaperone complex did not recover the usual substrates of this chaperone machinery, also point toward a network-type explanation of evolvability regulation.

IMPLICATIONS IN THE THERAPY OF CHRONIC DISEASES: CANCER, DIABETES, AND NEURODEGENERATION

Repeated stress induces an accumulating damage in cellular networks and parallel with this "wears out" the cellular adaptation capacity. This exceedingly happens in chronic disease and during aging (*6, 41*). An additional type of danger is raised by the fact that disease and aging induce a generally higher noise level (*48*). If disease and age-induced noise is accompanied by the extra, stress-generated noise, it may well go beyond the tolerable threshold and may induce an "error-catastrophe."

Efficient repair of the multiple rearrangements and defects of disease-, aging-, and stress-affected cellular networks are better provided by multitarget drugs than by the "magic bullets" of traditional drug design. Moreover, the low-affinity binding of multitarget drugs eases the constraints of druggability and significantly increases the size of the druggable proteome. These effects tremendously expand the number of potential drug targets and will introduce novel classes of multitarget drugs with smaller side effects and toxicity. In fact, many herbal teas, traditional medicines, nutrients, micronutrients, vitamins, and phytonutrients act as multitarget compounds interacting with various cellular networks with a low affinity (*49, 50*).

Because of the multitude of chaperone-mediated interactions and their central role in network integration, chaperone modulators are excellent, bona fide examples of multitarget drugs. Indeed, chaperone substitution [in the form of chemical chaperones, (*51*)], the help of chaperone induction (*52*), and chaperone inhibition (*53*) are all promising therapeutic strategies involving an increasing number of multitarget drugs acting on the chaperome (*54*).

SUMMARY AND PERSPECTIVES

The plethora of information in various datasets and our emerging knowledge on network topology and dynamics provide a unique chance to understand the rearrangements of cellular networks during stress and disease. It is especially intriguing to assess the long-thought integrative role of molecular chaperones at the level of the whole cell and organism. The highlights of our current knowledge can be summarized as follows:

- In an unstressed state, the yeast interactome is highly compact, and the centrally organized modules have a large overlap. During stress, the original modules become separated, and novel modules appear.
- Examination of yeast and human interactomes shows that the chaperones 1) are intermodular integrators of protein–protein interaction networks, which 2) often bridge hubs and 3) are favorite candidates for extensive phosphorylation. Moreover, chaperones 4) become more central in the organization of the isolated modules of the stressed yeast protein–protein interaction network, which highlights their importance in the decoupling and recoupling of network modules during and after stress. The human chaperome recovers the segregation of protein synthesis-coupled and stress-related chaperones observed in yeast recently.

However, a number of key issues have not been tackled, yet both from the theoretical and the experimental points of view.

- We are at the very beginning to understand stress-induced network rearrangements: the exploration of various stress conditions, as well as parallel datasets showing the differences between protein–protein interaction, organelle, and functional cellular networks are missing. The exploration of topological phase transitions of cellular networks by comparing their topology in extremely resource-rich and resource-poor environments awaits experimentation. Our knowledge on the reestablishment or rebuilding of cellular networks after stress is practically zero.
- The comparison of chaperones of various organisms will reveal a lot of exciting and heretofore uncovered functions of these key proteins. Even more importantly, we will have a novel view on their integrative functions, which is a typical emergent network phenomenon, which cannot be guessed from our current, fragmented knowledge but needs a global picture of the whole interactomes and other cellular networks.
- We need a much better understanding of cellular network changes in disease and aging. Besides chaperones-related therapies, the design of efficient therapeutic interventions to help cellular networks to cope with stress is missing.

We are quite certain that the rearrangements of stressed networks and the emergent properties of chaperomes will give a lot of excitement and pleasure in the near future. As a result of these studies, the emergence of network-based therapies is expected, in which entirely novel target sets of multitarget drugs will be identified using our knowledge on the vulnerable points (hotspots) of cellular networks in stress, disease, and aging.

ACKNOWLEDGEMENTS

The authors thank Huba J. Kiss (University of Pécs) for collecting the human chaperome data, the anonymous referee and members of the LINK-group (www.linkgroup.hu) for helpful suggestions. Work in the authors' laboratory was supported by

research grants from the Hungarian National Science Foundation (OTKA-K69105), from the EU (FP6-016003) and by the Hungarian National Research Initiative (NKFP-1A/056/2004 and KKK-0015/3.0).

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