Stressful initiations

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Journal of Cell Science 115, 3227-3234 (2002) © The Company of Biologists Ltd

Summary

Stress granules (SGs) are phase-dense particles that appear in the cytoplasm of eukaryotic cells that have been exposed to environmental stress (e.g. heat, oxidative conditions, hyperosmolarity and UV irradiation). SG assembly is a consequence of abortive translational initiation: SGs appear when translation is initiated in the absence of eIF2-GTP-tRNA$_i^{Met}$, the ternary complex that normally loads tRNA$_i^{Met}$ onto the small ribosomal subunit. Stress-induced depletion of eIF2-GTP-tRNA$_i^{Met}$ allows the related RNA-binding proteins TIA-1 and TIAR to promote the assembly of eIF2-eIF5-deficient preinitiation complexes, the core constituents of SGs. The mRNP components that make up the SG are in a dynamic equilibrium with polysomes. As such, the SG appears to constitute a metabolic domain through which mRNPs are continually routed and subjected to triage – they are first monitored for integrity and composition, and then sorted for productive translational initiation or targeted degradation.

Key words: Stress granules, TIA-1, TIAR, Protein translation, eIF2alpha, Heat shock

Introduction

In response to environmental stress (e.g. heat, hyperosmolarity and oxidative conditions), eukaryotic cells shut down protein synthesis in a stereotypic response that conserves anabolic energy for the repair of stress-induced damage. The translational arrest that accompanies environmental stress is selective: whereas translation of constitutively expressed 'housekeeping' transcripts is turned off, translation of stress-induced transcripts encoding heat shock proteins and some transcription factors is maintained or enhanced. The stress-activated signaling cascades responsible for reprogramming translation in stressed cells are becoming apparent. At the apex of these signaling cascades is a family of serine/threonine kinases that serve as sensors of environmental stress. Included in this family are the following: (1) PKR[†], a double-stranded RNA-dependent kinase that is activated by viral infection, heat and UV irradiation (Williams, 2001); (2) PERK/PEK, a resident ER protein that is activated when unfolded proteins accumulate in the ER (Harding et al., 2000; Patil and Walter, 2001); (3) GCN2, a protein that monitors amino acid levels in the cell and responds to amino acid deprivation (Kimball, 2001); and (4) HRI, a heme-regulated kinase that ensures the balanced synthesis of globin chains and heme during erythrocyte maturation (Han et al., 2001; Lu et al., 2001). Each of these stress 'sensors' phosphorylates eIF2α, a critical regulatory component of the ternary complex (composed of eIF2αβγ bound to tRNA$_i^{Met}$ and GTP) that loads initiator tRNA$_i^{Met}$ onto the small ribosomal subunit to initiate protein synthesis (Dever, 2002; Kimball, 2001). Phosphorylation of eIF2α converts the eIF2 ternary complex into a competitive inhibitor of eIF2B, the GTP/GDP exchange factor that converts inactive ternary complex (GDP-associated) to active ternary complex (GTP-associated) (Krishnamoorthy et al., 2001). Thus, phosphorylation of eIF2α inhibits protein translation by reducing the availability of the eIF2-GTP-tRNA$_i^{Met}$ ternary complex.

When translation is initiated in the absence of eIF2-GTP-tRNA$_i^{Met}$, an eIF2/eIF5-deficient, 'stalled' 48S* preinitiation complex is assembled (Kedersha et al., 2002). These eIF2/eIF5-deficient preinitiation complexes and their associated mRNA transcripts are dynamically routed to cytoplasmic foci known as stress granules (SGs), in a process that requires the related RNA-binding proteins TIA-1 and TIAR (Kedersha et al., 2002; Kedersha et al., 2000; Kedersha et al., 1999). In stressed cells, mRNA is in a dynamic equilibrium between polysomes and SGs (Kedersha et al., 2000). Here we discuss the antagonistic roles of eIF2α and TIA-1/TIAR in the assembly of polysomes and SGs in the context of a translational checkpoint model, wherein TIA and eIF2 are functional antagonists whose relative activities determine how many times a given transcript is translated before it is degraded.

Stress granules: a historical perspective

When exposed to supra-ambient temperatures, eukaryotic cells exhibit characteristic morphological changes in both nuclear and cytoplasmic compartments. In the nucleus, actin filaments condense to form rod-shaped bodies, and the granularity of nucleoli is altered (Collier et al., 1988; Collier and Schlesinger, 1986). In the cytoplasm, the cytoskeleton is rearranged and the Golgi apparatus is disrupted (Collier et al., 1988; Collier and Schlesinger, 1986). The appearance of phase-dense cytoplasmic granules was first noted in cultures of Peruvian tomato cells subjected to heat shock (Nover et al., 1983). Low-

[†]eIF, eukaryotic initiation factor; GCN2, general control nonrepressed 2; HRI, heme-regulated inhibitor; PERK, PKR-like endoplasmic reticulum kinase; PKR, double-stranded RNA-dependent protein kinase; RRM, RNA-recognition motif; TIA-1, T-cell intracellular antigen-1; TIAR, T-cell intracellular antigen-related.

molecular-weight heat shock proteins were identified as prominent components of tomato heat shock granules (Nover et al., 1983). Immunofluorescence microscopy revealed that they are also components of phase-dense granules observed in cytoplasm of heat-shocked mammalian cells (Arrigo et al., 1988). The use of immunofluorescence microscopy to detect stress-induced changes in the subcellular localization of cellular components has allowed the identification of cytoplasmic and nuclear microdomains that are assembled as part of the normal stress response. Heat-shock-induced transcription factors have been found to accumulate at discrete cytoplasmic and nuclear foci in response to stress (Cotto et al., 1997; Cotto and Morimoto, 1999; Holmberg et al., 2000; Jolly et al., 2002; Scharf et al., 1998). Similarly, the subcellular localization of mRNP particles involved in various aspects of mRNA metabolism is altered in cells subjected to environmental stress (Gallouzi et al., 2000; Krebber et al., 1999; Michael et al., 1995; van der Houven van Oordt et al., 2000). Thus, immunofluorescence microscopy can be used to identify subcellular domains that may regulate cellular metabolism during stress.

Stress granules: the mRNA connection

The identification of mRNA as a component of plant heat shock granules provided a clue to the possible function of these cytoplasmic structures (Nover et al., 1989). Remarkably, heat-shock-granule-associated mRNAs were found to encode constitutively expressed 'housekeeping' proteins but not newly synthesized heat shock proteins (Nover et al., 1989). This result suggested that translationally silenced mRNAs selectively accumulate at SGs, whereas translationally active mRNAs are excluded. These mRNAs could still be translated in vitro and could also be translated in vivo after the cells recovered from stress. Thus, the plant heat shock granule was proposed to serve as a storage repository for untranslated mRNAs.

Poly(A)$^+$ RNA is also a component of mammalian SGs (Kedersha et al., 1999), as demonstrated by in situ hybridization using oligo-dT (Fig. 1). Recently, sequence-specific in situ hybridization has shown that mRNA encoding inducible HSP70 is selectively excluded from mammalian SGs (P.A. and N.K., unpublished). Thus, the SGs found in the cytoplasm of both plant and animal cells are sites at which untranslated mRNA accumulates in cells subjected to environmental stress.

Stress granules: composition of the mRNP

Identification of the protein components of mammalian SGs was facilitated by the discovery that the related RNA-binding proteins TIA-1 and TIAR are robust markers of these cytoplasmic foci (Kedersha et al., 1999). TIA-1 and TIAR are concentrated in the nucleus of most cells, but heterokaryon analysis reveals that both proteins continuously shuttle between the nucleus and the cytoplasm (P.A. and N.K., unpublished). In response to environmental stress, TIA-1 and TIAR accumulate in the cytoplasm, where they rapidly aggregate to form mammalian stress granules (Fig. 1) (Kedersha et al., 1999). Following removal of a non-lethal stress, the SGs increase in size owing to fusion of smaller SGs, and then rapidly disperse [Fig. 1; see movie of this process in Kedersha et al. (Kedersha et al., 2000)]. TIA-1 (Tian et al., 1991) and TIAR (Kawakami et al., 1992) possess three RNA-recognition motifs at their N-termini and a glutamine-rich domain at their C-termini. TIA mutants lacking the RNA-recognition motifs function as transdominant inibitors of SG assembly in stressed cells (Kedersha et al., 2000) which suggests that the TIA proteins are required for SG assembly.

The discovery of TIA-1 and TIAR as markers of SGs allowed the use of dual-labeling studies to identify additional SG components. This analysis revealed that components of small, but not large, ribosomal subunits co-localize with TIA-1 and TIAR at SGs (Kedersha et al., 2002). The absence of the large ribosomal subunit eliminated the possibility that SGs are sites at which selected mRNAs are translated during stress. At the same time, the presence of the small ribosomal subunit indicated that the mRNPs making up SGs might be related to polysomes. Consistent with this premise is compositional analysis revealing that many of the protein and RNA components of SGs and polysomes are identical (Kedersha et al., 2002).

Translation is normally initiated when the small ribosomal subunit and its associated initiation factors are recruited to a capped mRNA transcript to form a 48S complex (Fig. 2, left branch of pathway; green arrows). Hydrolysis of eIF2-associated GTP by eIF5 displaces the early initiation factors, allowing the binding of the large ribosomal subunit. Repeated cycles of successful initiation convert an mRNA into a polysome. In stressed cells, activation of one or more eIF2α kinases (e.g. PKR, PERK/PEK, GCN2, HRI; Fig. 2, red box) results in the phosphorylation of eIF2α (Kimball, 2001; Williams, 2001), which consequently inhibits eIF2B, the

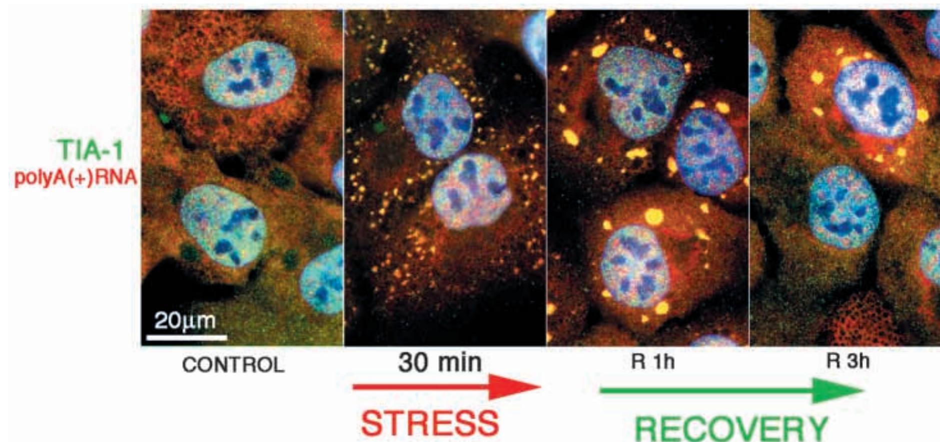

Fig. 1. Assembly and disassembly of arsenite-induced stress granules. DU145 cells were cultured in the absence (control) or presence of arsenite (1 mM) for 30 minutes (STRESS), washed and allowed to recover for 1 hour or 3 hours (RECOVERY) before processing for two-color immunofluorescent microscopy. TIA-1 protein is identified using a polyclonal antibody (green). Poly(A)+ RNA is revealed by in situ hybridation using an oligo-dT probe (red). Sites of co-localization of TIA-1 and poly(A)+ RNA appear yellow. Nuclei are counterstained using Hoechst dye (blue).

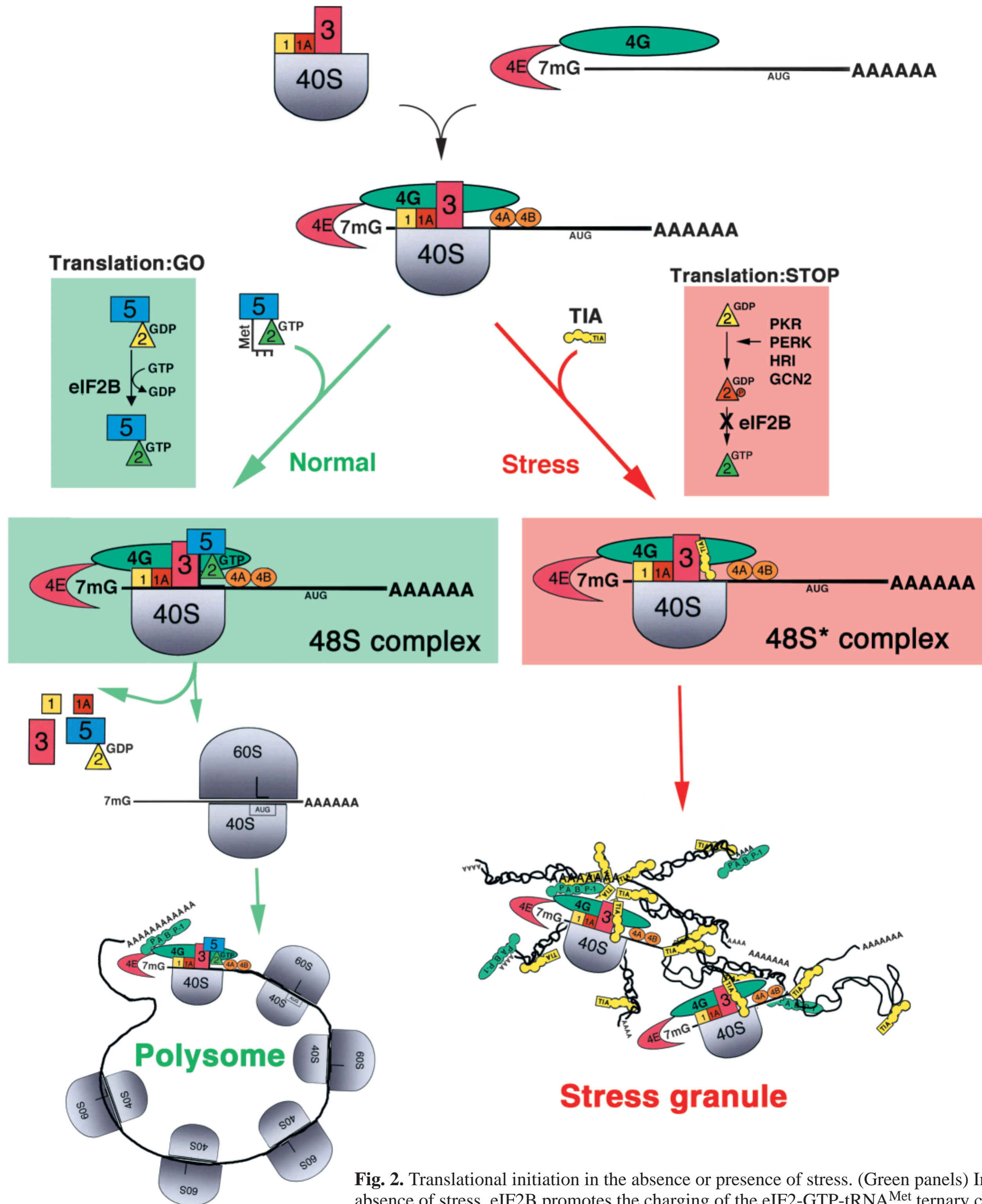

Fig. 2. Translational initiation in the absence or presence of stress. (Green panels) In the absence of stress, eIF2B promotes the charging of the eIF2-GTP-tRNA^{Met} ternary complex by exchanging GDP for GTP. When the eIF2-GTP-tRNA^{Met} ternary complex is available, a canonical 48S preinitiation complex is assembled at the 5′ end of capped transcripts (green arrow: Normal) and scanning begins. Upon recognition of the initiation codon by the anticodon of tRNA^{Met}, eIF5 promotes GTP hydrolysis, and early initiation factors are displaced by the 60S ribosomal subunit. As additional ribosomes are added to the transcript, the mRNA is converted into a polysome. (Red panels) In stressed cells (red arrow: Stress), phosphorylation of eIF2α by PKR, PERK, HRI or GCN2 converts eIF2 into a competitive antagonist of eIF2B, depleting the stores of eIF2/GTP/tRNA^{Met}. Under these conditions, TIA-1 is included in a non-canonical, eIF2/eIF5-deficient 48S* preinitiation complex (composed of all components of the 48S pre-initiation complex except eIF2 and eIF5) that is translationally silent. TIA-1 self-aggregation then promotes the accumulation of these complexes at discrete cytoplasmic foci known as stress granules. Blue square, eIF5; green triangle, eIF2 bound to GTP; yellow triangle, eIF2 bound to GDP; red triangle, phospho-eIF2 bound to GDP.

GTP/GDP exchange factor that charges the eIF2 ternary complex. The ensuing depletion of eIF2-GTP-$tRNA_i^{Met}$ prevents productive translation initiation. Under these conditions, TIA-1 and TIAR promote the assembly of an eIF2/eIF5-deficient preinitiation complex (denoted 48S* in Fig. 2) that is routed to SGs (Fig. 2, right branch of pathway, red arrows) (Kedersha et al., 2002). RNA-binding proteins that either promote [human autoantigen R (HuR) (Gallouzi et al., 2000)] or inhibit (tristetraprolin; P.A. and N.K., unpublished) mRNA stability are also recruited to SGs. This suggests that the SG is a site where the fates of specific transcripts are determined by the activity of different RNA-binding proteins. Whether the SG is also a site of mRNA processing (e.g. through degradation by exosomes) remains to be determined.

Stress granules and polysomes: a dynamic equilibrium

Earlier models suggested that heat-shock-granules are storage depots for untranslated mRNAs, but mammalian SGs exhibit behavior inconsistent with such a model. The antagonistic effects of different pharmacological inhibitors of protein translation on SG assembly have revealed that SG-associated mRNA is in a dynamic equilibrium with polysomes (Fig. 3) (Kedersha et al., 2000). Drugs that stabilize polysomes by freezing ribosomes on translating mRNAs (e.g. cycloheximide and emetine) inhibit the assembly of SGs and actively dissolve SGs in the continued presence of both stress and eIF2α phosphorylation (Kedersha et al., 2000). Conversely, drugs that destabilize polysomes by releasing ribosomes from mRNA transcripts (e.g. puromycin), promote the assembly of SGs (Kedersha et al., 2000). This behavior suggested that SG-associated poly(A)+ RNA is in equilibrium with polysomes. Direct evidence for the dynamic nature of SGs was obtained in experiments using green fluorescent protein (GFP)-TIA-1 and a construct in which GFP was fused to poly(A)-binding protein (PABP-GFP) to monitor the assembly and disassembly of SGs in living cells (Kedersha et al., 2000). In response to

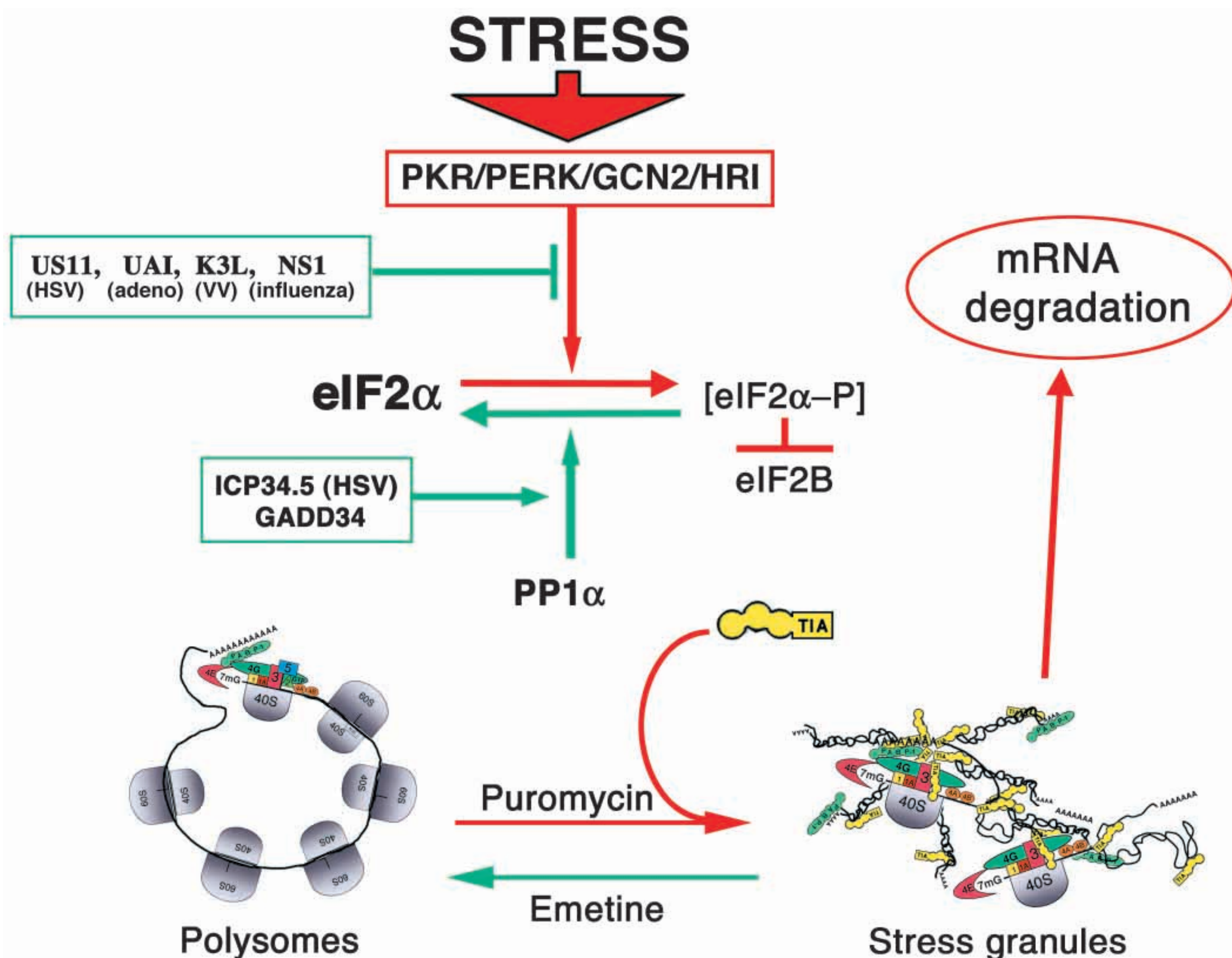

Fig. 3. The dynamic equilibrium between polysomes and stress granules is regulated by the availability of eIF2-GTP-$tRNA^{Met}$ and TIA proteins. Stress-induced activation of eIF2α kinases reduces the concentration of eIF2-GTP-$tRNA^{Met}$, allowing TIA proteins to promote the assembly of stress granules. Several eukaryotic viruses [e.g. herpes simplex virus (HSV), adenovirus (adeno), vaccinia virus (VV) and influenza virus] prevent stress-induced translational arrest by inhibiting the activity of eIF2α kinases or activating eIF2α phosphatases. Green events and arrows indicate routes leading to translational activation; red events and arrows indicate events leading to translational inhibition.

arsenite-induced oxidative stress, GFP-TIA-1 rapidly moves from the nucleus to the cytoplasm, where it is evenly and diffusely distributed. After ~10 minutes, the cytoplasmic GFP-TIA-1 aggregates into discrete foci that coalesce and slowly enlarge over the next 20 minutes. When arsenite is washed out of the cells, the SGs slowly disassemble with similar kinetics. However, this slow and steady accumulation of GFP-TIA-1 at SGs is misleading. Fluorescence recovery after photobleaching (FRAP) analysis reveals that GFP-TIA-1 shuttles in and out of SGs very rapidly such that 50% of SG-associated GFP-TIA-1 is replaced every 2 seconds. Although the rate at which mRNA shuttles in and out of SGs was not determined directly, GFP-PABP was used as a surrogate marker for its associated mRNA. Interestingly, GFP-PABP shuttles in and out of SGs at a rate that is ten times slower than that of GFP-TIA-1 (i.e. 50% of SG-associated GFP-PABP is replaced every 20 seconds). Given these kinetics, and considering that the dominant negative mutant of TIA-1 (e.g. TIA-1ΔRRM) prevents SG assembly altogether (Kedersha et al., 2000), it appears that TIA-1 actively escorts untranslated mRNA to SGs. These data reveal that SGs are highly dynamic structures despite their apparent stability in real-time microscopy.

The dynamic movement of mRNA into and out of SGs argues against a model in which SGs are passive repositories of untranslated mRNAs that accumulate in stressed cells. Rather, the SG is more likely to serve as a 'way station' through which untranslated mRNAs pass before being translated or degraded (Kedersha et al., 2000). Taken together, the data suggest that TIA-1 and TIAR act downstream of the stress-induced phosphorylation of eIF2α to drive mRNA from polysomes to SGs (Fig. 3). The central importance of the PKR/eIF2α pathway in the cellular response to stress is indicated by the number of eukaryotic viruses that must disable PKR or reverse the phosphorylation of eIF2α to effect productive infection (Barber, 2001). If TIA-1 is an important component of the PKR/eIF2 stress response pathway, viruses might target TIA-1 to subvert this protective response. Indeed, TIA-1 and TIAR have been found to bind to specific sequences encoded by both West Nile Virus RNA (W. Li, Y. Li, N.K. et al., unpublished) and Sendai virus RNA (F. Iseni, D. Barcin, M. Nishhio et al., unpublished). Fig. 3 depicts the dynamic equilibrium between polysomes and SGs and summarizes the molecular pathways that regulate this equilibrium.

Translational triage model

Fig. 4 shows a model depicting the molecular stages in the conversion of a polysome into an SG and the subsequent fate of its component mRNAs. The formation of an eIF2-deficient 48S* complex at the 5′ end of the polysomal mRNA allows TIA to bind to RNA in lieu of ternary complex (stage 1), resulting in an abortive initiation event. This allows the previously initiated ribosomes to run off the transcript (stages 2 and 3) until only the 48S* complex remains bound to the 5′ end of the RNA (stage 4). Should an eIF5-eIF2-containing ternary complex become available at any time during this process, the transcript can still be reinitiated (green arrow) by displacement of TIA with eIF5-eIF2-GTP-$tRNA_i^{Met}$. However, as stress conditions disrupt TIA shuttling such that the normally nuclear TIA accumulates in the cytoplasm, more TIA becomes available to bind nonspecifically to the mRNA. The self-aggregation of TIA drives SG assembly (stages 5 and 6). Other RNA-binding proteins such as HuR and tristetraprolin (TTP) are recruited to the SG to determine the fate of specific transcripts. ATP depletion promotes the assembly of SGs without increasing phospho-eIF2α (Kedersha et al., 2002), and overexpression of HSP70 results in the dispersal of the prion-like domain of TIA-1 in vivo (P.A. and N.K., unpublished). Therefore, we propose that HSP70 and ATP are required to extricate mRNAs from the SG, probably by altering the conformation of the TIA prion-like domain. Thus, according to this model, phospho-eIF2α initiates the formation of SGs, a sudden flood of untranslated polyA(+) mRNA is released from polysomes, TIA proteins aggregate this RNA, and HSP70 (and ATP) is required to dis-aggregate the TIA proteins.

Translational silencing without visible stress granules

Overexpression of recombinant TIA-1 represses the production of co-expressed reporter genes in the absence of exogenous stress (Kedersha et al., 2000). It is therefore likely that some eIF2/eIF5-deficient 'abortive' preinitiation complexes are assembled under normal conditions, and that their frequency determines how often mRNA transcripts are subjected to triage. Consistent with this view, endogenous TIA-1 and TIAR repress the translation of TNFα transcripts in the absence of stress. Both TIA-1 and TIAR are components of a regulatory complex that binds to an adenine/uridine-rich element found in the 3′ untranslated region of TNFα transcripts (Gueydan et al., 1999; Piecyk et al., 2000). LPS-activated macrophages derived from mice lacking either TIA-1 or TIAR overexpress TNFα compared with wild-type controls. In macrophages lacking TIA-1, the polysome profile of TNFα transcripts is shifted such that the percentage of TNFα transcripts associated with polysomes is increased compared with that of wild-type macrophages (Piecyk et al., 2000). This suggests that TIA-1 represses the translation of TNFα by promoting the assembly of non-polysomal mRNP complexes. Sucrose-density-gradient-analysis reveals that TIA-1 is found in low-density fractions that contain soluble proteins, as well as higher density fractions that contain 40-60S mRNPs, but not in polysomes (Kedersha et al., 2000). It is likely that the TIA-mediated shift of TNF-α mRNA transcripts away from polysomes occurs through the assembly of abortive eIF2/eIF5-deficient pre-initiation complexes that are similar or identical to those that comprise the core units of SGs.

Nuclear history

Their visible roles in the cytoplasm notwithstanding, TIA-1 and TIAR normally predominate in the nucleus, where they have been shown to act as selective regulators of alternative mRNA splicing (Del Gatto-Konczak et al., 2000; Forch et al., 2000; Le Guiner et al., 2001). The binding of either TIA-1 or TIAR to uridine-rich elements found in intronic sequences located downstream of weak 5′ splice sites promotes the recruitment of U1snRNP and the inclusion of 'cryptic' alternative exons that are otherwise excised from the heteronuclear RNA. In their dual ability to regulate both mRNA splicing and translation, TIA-1 and TIAR resemble

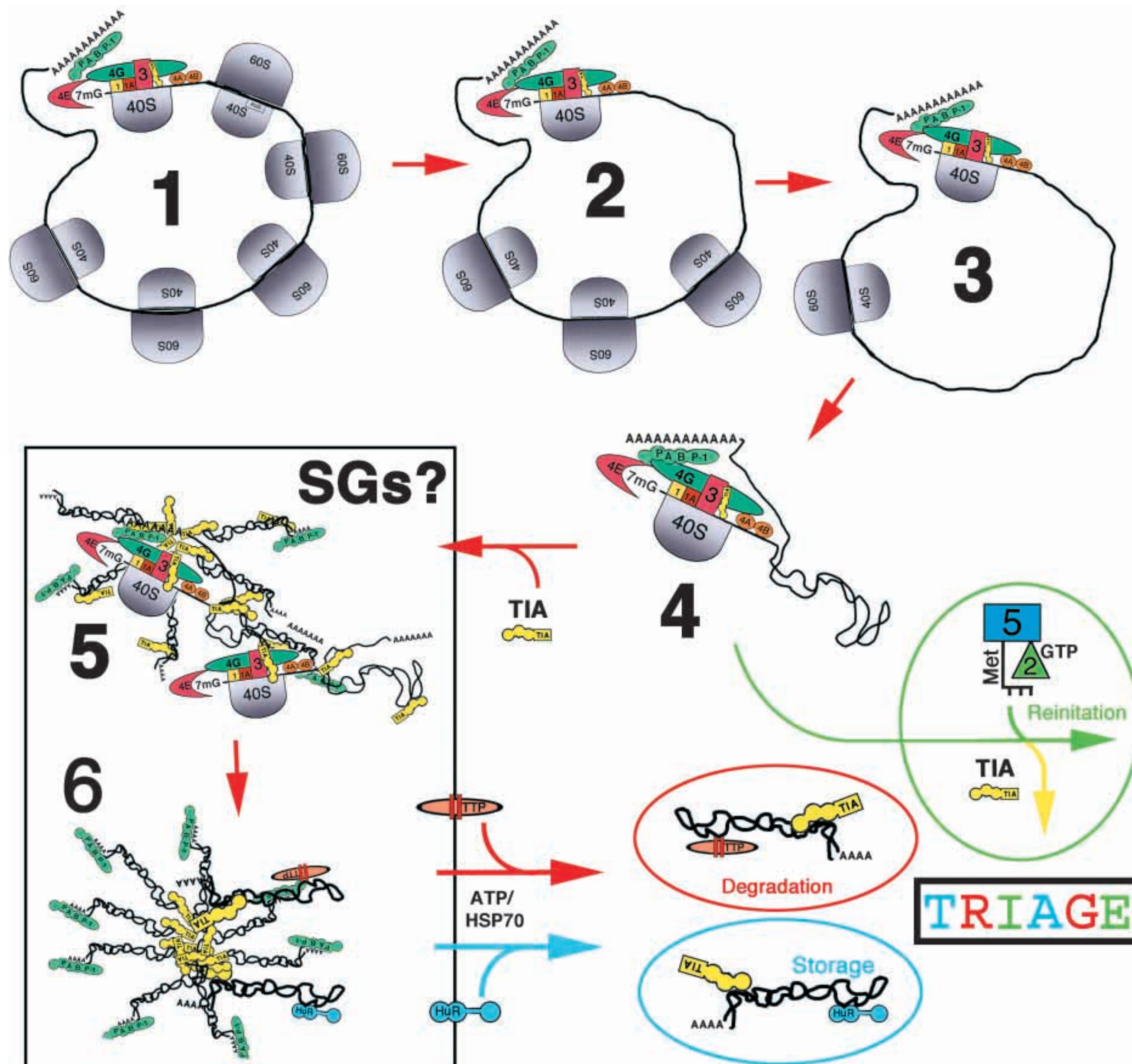

Fig. 4. Proposed mechanism for the assembly of stress granules. Assembly of an eIF2/eIF5-deficient preinitiation complex at the 5′ end of a polysome results in translational arrest (1). As elongating ribosomes 'run-off' the mRNA (2,3), the polysome is converted into a 48S* preinitiation complex (4) that is routed by TIA-1 into stress granules (5,6) or productively initiated by the replacement of TIA with eIF5/eIF2/GTP/tRNA^{met} (green circle). A requirement for HSP 70 and ATP in removing mRNAs from the SG is indicated. Destabilizing elements (red) such as tristetraprolin (TTP) are proposed to direct selected stress granule mRNAs to sites of degradation, whereas stabilizing elements such as HuR (blue) are proposed to direct selected mRNAs to sites of storage and/or reinitiation. By this triage process, the SG may monitor the structure and integrity of mRNP complexes and determine the fate of specific RNAs.

several other multifunctional RNA-binding proteins, including PTB, CUB-BP-related proteins, La, hnRNPK and hnRNP A1 (Wilkinson and Shyu, 2001; Ladomery, 1997). The process of mRNA splicing has been shown to 'mark' selected mRNA transcripts for both quality control in the nucleus and translational control in the cytoplasm (Le Hir et al., 2000a; Le Hir et al., 2000b). It is therefore possible that heteronuclear mRNAs encoding introns that are recognized by TIA-1/TIAR retain these proteins at the exon-exon junction following the removal of the intron upon splicing. Upon arrival in the cytoplasm, transcripts that are 'marked' by TIA-1/TIAR could be selectively regulated at the level of mRNA stability or translatability. Such an explanation would allow for the nuclear loading of TIA-1 and TIAR onto selected transcripts that become subject to translational silencing once in the cytoplasm. It also suggests a mechanism whereby stress-induced transcripts might be excluded from SGs: mRNAs transcribed during stress would escape being marked by TIA proteins, whose normal shuttling appears to be disrupted when they are routed into cytoplasmic SGs.

Conclusions/perspectives

Molecular modifications (particularly phosphorylation) are widely appreciated to regulate many distinct steps of the translational initiation process (Clemens, 2001; Lawrence and

Brunn, 2001; Mahalingam and Cooper, 2001). The importance of subcellular compartmentalization of translational components is an important mode of regulation that occurs in living cells but is less understood. Key components of the translational apparatus (e.g. mRNA and its associated proteins, ribosomal subunits and translation initiation factors) move between the nucleus and the cytoplasm in a regulated manner. The localized availability and hence the activity of these components is regulated by interactions with organelles and with the cytoskeleton. The rapid assembly and disassembly of mammalian SGs is a striking illustration of this type of regulation. SGs coalesce into existence as mRNAs are released from polysomes during stress-induced translational arrest and melt away like snowflakes as the cell adapts or recovers from the stress.

The molecular trigger for SG formation is the abortive translation that occurs when a transcript is initiated without the eIF2-GTP-$tRNA^{Met}$ ternary complex. The assembly of translationally inactive initiation complexes lacking eIF2, coupled with increased levels of cytoplasmic TIA, allows the RNA-binding proteins TIA-1 and/or TIAR to redirect untranslated mRNAs from polyribosomes to SGs (Fig. 4). Thus, TIA-1/TIAR and eIF2-GTP-$tRNA_i^{Met}$ appear to act as antagonists that regulate the equilibrium between polysomes and SGs (Figs 3, 4). It remains to be determined whether TIA-1 and eIF2-GTP-$tRNA^{Met}$ compete for binding to a common site on the preinitiation complex. Regardless of the details of molecular mechanism, the functional antagonism between eIF2 and TIA may determine the frequency with which a given mRNA transcript is initiated before being subject to a checkpoint at which mRNP structure and composition is monitored. If the ratio of TIA-1/TIAR to eIF2-GTP-$tRNA^{Met}$ were 1:10, one might predict that, on average, ten productive initiation events would occur before a TIA-1/TIAR-containing translationally incompetent pre-initiation complex is assembled. As translating ribosomes 'run off' this mRNA, the eIF2/eIF5-deficient complex would be directed to an SG, where the integrity and composition of the mRNP might determine whether the transcript is reinitiated, degraded or packaged into an untranslated mRNP particle.

Although considerable progress has been made in elucidating the relationship between SGs and translational initiation, we have much to learn about the connection between SGs and molecular chaperones. The aggregation domain of TIA is its glutamine-rich C-terminus, which resembles prion protein, and is essential for SG assembly. Specific molecular chaperones are required to disperse aggregated TIA-1 in living cells (P.A. and N.K., unpublished). These chaperones are also required for the adaptive response to stress, which suggests that SGs constitute a point of crosstalk between molecular chaperones and eIF2 kinases. Do SGs function as signal transduction domains? And what else is in SGs? Plant SGs contain not only RNA and small HSPs but also heat shock transcription factors. Do mammalian SGs also contain stress-related transcription factors and, if so, which ones? The end of stress is not yet in sight!

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