Seminars in Cell and Developmental Biology xxx (xxxx) xxx

Contents lists available at ScienceDirect

Seminars in Cell and Developmental Biology

journal homepage: www.elsevier.com/locate/semcdb

Review

Composition and function of stress granules and P-bodies in plants

Alyssa Kearly [a], Andrew D.L. Nelson [a], Aleksandra Skirycz [a], Monika Chodasiewicz [b,*]

[a] The Boyce Thompson Institute, Cornell University, Ithaca, NY, USA
[b] Center for Desert Agriculture, Biological and Environmental Science and Engineering Division, King Abdullah University of Science and Technology (KAUST), Thuwal, Saudi Arabia

ARTICLE INFO

Keywords:
Stress Granules
P-bodies
Proteins
Metabolites
mRNA
Condensates

ABSTRACT

Stress Granules (SGs) and Processing-bodies (P-bodies) are biomolecular condensates formed in the cell with the highly conserved purpose of maintaining balance between storage, translation, and degradation of mRNA. This balance is particularly important when cells are exposed to different environmental conditions and adjustments have to be made in order for plants to respond to and tolerate stressful conditions. While P-bodies are constitutively present in the cell, SG formation is a stress-induced event. Typically thought of as protein-RNA aggregates, SGs and P-bodies are formed by a process called liquid-liquid phase separation (LLPS), and both their function and composition are very dynamic. Both foci are known to contain proteins involved in translation, protein folding, and ATPase activity, alluding to their roles in regulating mRNA and protein expression levels. From an RNA perspective, SGs and P-bodies primarily consist of mRNAs, though long non-coding RNAs (lncRNAs) have also been observed, and more focus is now being placed on the specific RNAs associated with these aggregates. Recently, metabolites such as nucleotides and amino acids have been reported in purified plant SGs with implications for the energetic dynamics of these condensates. Thus, even though the field of plant SGs and P-bodies is relatively nascent, significant progress has been made in understanding their composition and biological role in stress responses. In this review, we discuss the most recent discoveries centered around SG and P-body function and composition in plants.

1. Introduction

In nature, plants are exposed to a constantly changing environment and are often challenged by biotic and abiotic stresses. To survive unfavorable conditions, plants have developed mechanisms to adjust growth and metabolism. One example of such a mechanism is the formation of stress granules (SGs) [1] and processing bodies (P-bodies) [2]. These cytosolic foci are generally thought to be involved in the partitioning of protein-RNA-metabolite complexes in order to maintain proper balance between protection, degradation, and translation of mRNA under stress conditions and later on in the recovery phase [3,4]. This mechanism would theoretically prevent cells from investing energy into often counterproductive protein translation under stress conditions. The majority of research on SGs was performed in yeast and mammalian cells, however SGs have also been observed in plants and in chloroplasts. Their observation in chloroplasts suggests they may also exist in prokaryotes. In 2008, scientists identified SG-like structures in the model alga, *Chlamydomonas reinhardtii* [5], which also points towards the existence of such granules in a broader spectrum of single cell organisms. These organisms might be a useful tool to study SG-related processes in plants in light of evolution and development [6].

SGs represent a conserved transient mechanism of primary response to changes in the environment. In plants, they are formed in response to a variety of environmental stresses [7], including heat-shock [8], low oxygen levels (hypoxia) [9], salinity [10], hyperosmolarity [11], and viral infection [12], and usually disassemble during recovery. The recovery phase is the period during which stalled translation is being restored and plants restart growth processes. Studies in zebrafish revealed that SGs are crucial for their recovery from stress, and a zebrafish line engineered to lose the ability to form SGs could not recover once the stress was removed [13]. Several additional studies in human cell lines have shown that cell survival during stress or recovery afterwards are hindered when SG formation is prevented by the ablation of certain SG components [14–16]. Therefore, formation of SGs and selective sequestration of different components into those compartments is crucial for stress tolerance. In contrast to SGs, P-bodies are present independent of stress, but their dynamics and mobility might be affected by different environmental conditions. For example, in human cell lines,

* Corresponding author.
E-mail address: monika.chodasiewicz@kaust.edu.sa (M. Chodasiewicz).

https://doi.org/10.1016/j.semcdb.2022.11.008
Received 23 August 2022; Received in revised form 16 November 2022; Accepted 22 November 2022

oxidative stress increases the *de novo* formation of P-bodies while mitochondrial and heat stresses do not [4]. Similar increases in P-body synthesis have also been observed in yeast undergoing glucose starvation [17] and osmotic stress [18]. As the size and number of P-bodies depends on the status of translation machinery, and translation status might change in response to environmental stress, it is expected that stress will affect P-bodies composition or dynamics. In plants, both biotic and abiotic stress clearly have an impact on P-bodies [19]. For example, microbe-associated molecular patterns (MAMPs) were shown to modulate the dynamics of P-bodies [20] through the phosphorylation of mRNA decapping complex components DCP1 and DCP2 and selective regulation of mRNA decay. Overall, deviations from normal, homeostatic conditions influence the formation and dynamics of SGs and P-bodies that aid in the cellular response to such stresses.

The current understanding of SG and P-body dynamics is summarized in Fig. 1. In this review, we will discuss the most up-to-date discoveries on plant SG and P-body composition in terms of proteins, metabolites, and RNA. We will focus our discussion on key components and events associated with formation, sequestration into, and release from, SGs and P-bodies.

2. Proteins as key components of SGs and P-bodies

In general, the current model for SG assembly is composed of two steps. The first step is the formation of a dense core containing all the key components of SGs. The second step is the sequestration of components into the peripheral shell [8,21–23] that is considered to be more fluid and dynamic. Such condensation in the cell is possible due to liquid-liquid phase separation (LLPS) properties. LLPS highly depends on polymerization of proteins containing low complexity domains (LCDs) [24]. Among many proteins containing LCDs, RNA binding proteins are the majority. Transition of RNA binding proteins into SGs is driven by polymerization of proteins via their LCDs, which contributes to LLPS phenomena [25]. This event is crucial for SGs formation as well as for P-bodies [26].

Recent research in mammals, yeast, and plants has shed light on the

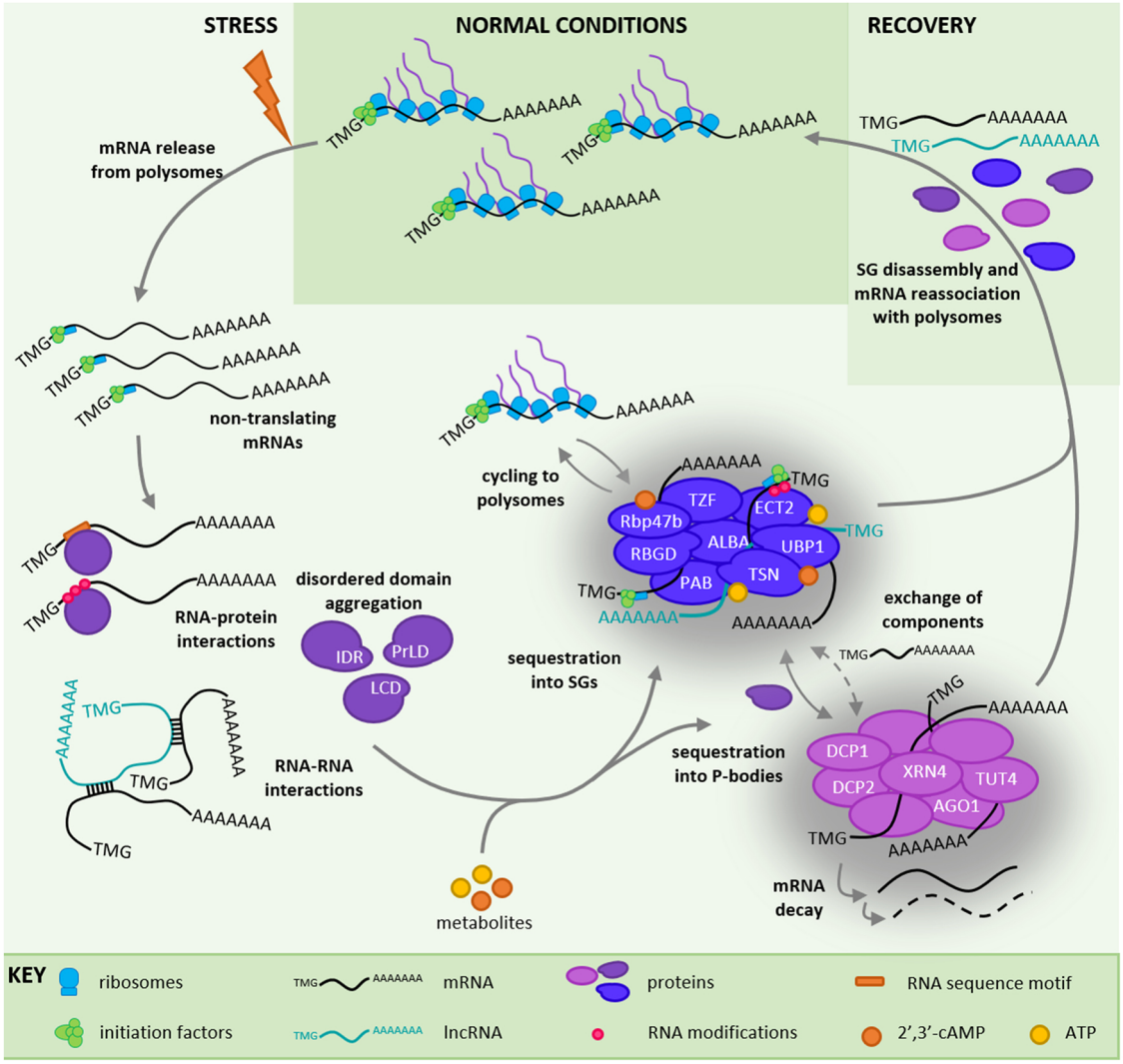

Fig. 1. Summary of current perspectives on SG and P-body dynamics. Normal protein translation is disrupted under stress conditions as mRNAs are released from polysomes. Free RNAs and proteins undergo LLPS through a combination of RNA-RNA interactions via complementary sequences; RNA-protein interactions via recognition of sequence motifs or base modifications; and protein aggregation via disordered domains such as IDRs, PrLDs, and LCDs. SGs are formed, a process regulated in part by small molecules like ATP and 2′,3′-cAMP, and P-bodies, which are already present in unstressed cells, increase in size and number. In these compartments, transcripts may be stored and preserved or subject to decay, respectively. Throughout the duration of stress, protein components are exchanged between SGs and P-bodies. SG-associated mRNAs cycle to polysomes and potentially to P-bodies, although there is conflicting evidence for this latter trafficking. Upon recovery, SGs and P-bodies disassemble, releasing their protein and RNA components, and the latter reassociate with polysomes as translation resumes. TMG, trimethylguanosine; IDR, intrinsically disordered region; PrLD, prion-like domain; LCD, low complexity domain; ARE, AU-rich element.

molecular composition of SGs and P-bodies [8,21,22]. To date, research on mammalian cells has identified around 500 SG-associated proteins [27], among which are proteins with domains that promote aggregation like LCDs, intrinsically disordered regions (IDRs), and prion-like domains (PrLDs), proteins involved in binding and regulation of RNA molecules, and also ATPases [3,27,28]. With the use of fluorescence-activated particle sorting (FAPS) of P-bodies, scientists identified around 125 proteins as stable components of P-bodies [29]. Interestingly, 28 of the identified P-body proteins were also present in the SG core proteome [21], perhaps reflecting the constant dynamic exchange between SGs and P-bodies [30]. Similarity in protein composition was further supported by computational analyses in which scientists compared SG and P-body proteomes by analyzing the proximity interactions of multiple baits for each foci [28]. Analysis revealed 106 proteins predicted to reside in SGs and 38 in P-bodies, with overlap of 16 proteins that were shared between both. Results were further validated using a colocalization approach, leading to 90% of positive hits [28,31]. This observed overlap in SG- and P-body-associated proteins may either attest to the exchange of components between the two foci or be indicative of the inherent propensity of these proteins to form aggregates.

The most well studied integral components in mammalian SGs are eukaryotic translation initiation factors eIF3 and 4G, Cytotoxic Granule Associated RNA Binding Protein TIA1, Poly(A)-binding protein 1 (PAB1), and Ras-GAP SH3 domain-binding protein (G3BP1 and G3BP2) [21,32,33]. In plants, around 120 proteins were identified as components of heat stress-induced cytosolic SGs and a quarter of those proteins have homologs in mammalian and yeast SGs [8]. Among the conserved set of proteins were well-accepted SG markers RNA binding protein 47b (Rbp47b) [34] and Tudor-staphylococcal nuclease 1 and 2 (TSN1/2) [35], RNA-binding proteins, ATPases and chaperones, and translation initiation and elongation factors [8]. Interestingly, on the list of cytosolic proteins, a few chloroplast-derived proteins were observed. Five of these proteins were further confirmed to relocalize into heat-induced chloroplastic SGs [23].

In Arabidopsis, few proteins are well accepted as plant SG markers. These include the oligouridylate binding protein 1 (UBP1) family containing three members, UBP1a, UBP1b, and UBP1c. All three members were shown to relocalize into SGs under heat stress, and UBP1a and UBP1c also under hypoxia stress [9]. Interestingly, increased expression of UBP1b was shown to enhance tolerance to heat [36] likely through the protection of stress-related mRNAs sequestered in SGs. Plant SG markers also include RNA-binding proteins Rbp45 and Rbp47. These proteins relocate into SGs under heat, salinity, and hypoxia [19,37,38]. A third group of proteins contains two Tudor-staphylococcal nuclease proteins, TSN1 and TSN2 that are evolutionary conserved RNA-binding proteins [39] shown to localize into SGs under salt and heat stress [10]. Proteins from all three of these families are regularly used as markers in all research studies that require confirmation of SG localization.

Recently, two glycine-rich RNA-binding proteins RBGD 2 and 4 were identified as components of SGs formed under heat stress conditions [40]. The authors beautifully demonstrated that the heat-induced LLPS into SGs is driven by the presence of a LCD at the C-terminal part of the protein, and mutation of the tyrosine residue array (TRA) in the LCD leads to both abolishment of SG formation and decrease in stress resistance. This suggests that LLPS properties and SG formation are crucial for plant survival under heat stress. Other proteins that confer tolerance through formation of SGs include the small dimetric DNA/RNA-binding acetylation lowers binding affinity (ALBA) proteins [41]. By performing immunopurification coupled with mass spectrometry (IP-MS) experiments using different baits of ALBA proteins fused to GFP, the authors observed their interaction with SG and P-body proteins. This was further confirmed by inverted IP-MS using GFP-Rbp47b and DCP5-GFP baits and also by yeast two hybrid techniques. Co-localization studies revealed that ALBA proteins can be localized in both SGs and P-bodies. Localization into SGs and P-bodies was heat stress specific and did not occur under cold, osmotic, nor salinity stress [41]. As a complement to previously described research, Multiprotein Bridging Factor 1 (MBF1) was recently identified as a key player in thermotolerance in wheat (*Triticum aestivum*) through its localization into SGs [42]. Both overexpression and downregulation of TaMBF1 in wheat significantly affected plant height and fresh weight under heat stress suggesting that TaMBF1 is required for heat tolerance in wheat.

Interestingly, P-bodies also respond to different stresses such as cold, drought, and salt stress [43,44]. For example, the SM-like proteins (LSMs), which are implicated in numerous aspects of RNA metabolism in eukaryotes and are a known component of P-bodies, were shown to target selected stress-inducible transcripts for decapping and degradation depending on the abiotic stress. In that way, LSMs can control transcript expression levels and thus lead to adequate transcriptomic response [43]. Plants lacking LSM1a and LSM1b were hypersensitive to salt and osmotic stress, confirming that selective regulation of mRNA decay is crucial for stress tolerance.

Both foci are very dynamic and any stimulus that affects the dynamics of one might have an effect on the other as well. As an example, 2′,3′-cyclic adenosine monophosphate (2′,3′-cAMP), a signaling molecule that interacts with the SG marker Rbp47b, was shown not only to induce SG formation [34], but also to increase the speed and distance of P-body movement within the cell [45]. Direct interaction between SGs and P-bodies can be inferred by looking at RNA-binding proteins such as tandem zinc finger proteins (TZFs), which have diverse roles in plant growth and response to stress. Arabidopsis TZF1 was found to be shuttled between the nucleus and P-bodies under control conditions and relocalized to SGs under heat stress [46]. TZF4, 5 and 6, which are involved in regulation of seed germination, were shown to be involved in response to light, abscisic acid (ABA), and gibberellic acid (GA) [47]. In rice, OsTZF1 showed increased co-localization with SGs and P-bodies under drought stress [48] and in doing so, conferred delayed senescence and stress tolerance. Despite the fact that SGs can interact with P-bodies and might exchange their components, one of the biggest differences between them is that SGs uniquely contain translation initiation factors [49]. In contrast, the majority of proteins that are associated with P-bodies are involved in RNA degradation, including decapping complexes, 5′ to 3′ exoribonucleases, small-RNA-dependent slicer Argonaute 1 (AGO1), and proteins involved in general translational repression [50,51]. Recently, more novel components are being identified, such as proteins involved in RNA modification (TUT4, APOBEC3F), proteins regulating post-translational modifications, and proteins such myosin, or microtubule filaments pointing towards a connection between P-bodies and the cytoskeleton [29,52]. The proteins enriched in SGs and P-bodies reflect the generally suggested roles of these structures in mRNA storage and decay. As many of these proteins and their interactions with SGs and P-bodies are conserved across species, they likely play key roles in the generation of these structures or in the recruitment or processing of associated RNA transcripts.

3. The role of metabolites in regulating SG dynamics and function

Recent findings have convincingly demonstrated the role of metabolites in the dynamics and function of SGs. In addition to proteins and RNAs, metabolites were also shown to sequester within SGs. Examples include nucleotides, amino acids, and lipids [8,23]. We anticipate that the occurrence of metabolites within SGs can be both “co-incidental” (i.e. proteins being sequestered together with their small-molecule ligands without any apparent function) as well as contributing to SG dynamics and function, which is well illustrated by adenosine triphosphate (ATP). SGs require ATP to fuel the activity of RNA and protein chaperone complexes, which might directly affect the SGs dynamics [21]. Chaperones are a critical component of the SGs, and among other functions, they counter the propensity of the IDR-containing proteins to form insoluble aggregates. In addition to being an energy currency, ATP is a

hydrotrope that prevents the formation of and dissolves previously formed protein aggregates [53]. Intriguingly, amino acids such as proline found in the SG isolations also can act as small-molecule chaperones, possibly further contributing to the chaperoning properties of the SGs [54]. Treatments that lead to a rapid drop in ATP levels prevent SG assembly, and once SGs are formed impede their liquid-liquid disassembly and prevent dissociation [21]. In the recent paper, Cereghetti and colleagues, using an elegant combination of genetics and cell biology, demonstrated that SGs are not only dependent on ATP but are also involved in fine-tuning ATP levels [55]. In glucose-grown yeast, stress is associated with reduced glycolysis and, consequently, a decrease in fructose 1,6 bisphosphate (FBP). FBP is an allosteric ligand of a glycolytic enzyme pyruvate kinase (CDC19) that catalyzes the final ATP-producing step of glycolysis. FBP binding to CDC19 promotes its active tetrameric structure. The drop in the FBP leads to the tetramer disassembly; the monomeric CDC19 then has the propensity to aggregate and sequester within SGs, where it remains inactive. When the stress ceases, FBP levels go up, and FBP binding to CDC19 promotes recruitment of chaperones and CDC19 re-solubilization. In turn, once released from SGs, CDC19 contributes to the increase in the ATP levels required for SG disassembly. Based on the evolutionary conservation of the SG core components and functionalities, we expect that ATP will be equally important for the SG dynamics in plants.

A different nucleotide compound associated with SGs is RNA degradation product 2′,3′-cAMP. 2′,3′-cAMP binds to the RNA-binding motif, dubbed RRM, present in the core SG proteins, and importantly Rbp47b [34]. 2′,3′-cAMP supplementation is associated with large changes at transcriptome, proteome, and metabolome levels, reminiscent of plant stress responses and it also promotes SG assembly and changes mobility of the P-bodies [34,45]. However, in contrast to ATP, 2′,3′-cAMP is not required for SG formation and plants impaired in stress-induced increase in the 2′,3′-cAMP levels are not compromised in SG formation [8,56]. Whereas ATP and 2′,3′-cAMP affect SG dynamics, a different metabolite, S-adenosylmethionine (AdoMet), was shown to suppress SG formation in response to acute stress, and also affect both the expression and recruitment of the specific SG components, such as SG nucleator Ded1 [57]. AdoMet is a co-substrate involved in methyl group transfers, however protein-methylation does not seem to have an effect on the AdoMet function. AdoMet could also alter RNA methylation. Intriguingly, S-adenosylmethionine synthase, an enzyme responsible for AdoMet production sequesters within SGs in yeast during post-diauxic, ethanol-fueled growth, pointing to an intricate interplay between metabolism and SG dynamics, composition, and function. Analogously to yeast, diverse metabolic enzymes and metabolic regulators such as SNF1-related kinase 1 (Snrk1) [8,56] have also been reported in plants, opening a new avenue of SG research. Further examination of this overlooked, yet critical, component of SGs will likely influence our understanding of the dynamic sequestration and release of protein and RNA components.

4. RNA transcripts as participants in SG formation and SG-mediated stress responses

Historically, the focus of SG and P-body research has been the protein composition of these granules and the impact that sequestering these proteins has on cellular responses to stress. However, recent work has highlighted the importance of RNA molecules in the nucleation and maintenance of such membraneless aggregates and has sparked an interest in identifying the specific RNAs present within them. The RNA composition and fate in P-bodies has been discussed extensively elsewhere [2,50,58,59]. Here, we focus on the RNA species of SGs—their common characteristics, how they may be directed to these aggregates, and what happens to them once they are associated. Where possible, we detail insights into the specifics of plant SGs, and then supplement these findings with information from other systems. As SGs appear to play conserved roles in mRNA biology and stress tolerance across eukaryotes, we envision overlap in key concepts and pathways.

4.1. SG RNA composition and targeting

Despite a global decrease in translation under stress conditions, only a fraction of RNA transcripts localize to stress granules, begging questions of whether there is any specificity in this localization and, if so, how this specificity is managed. The formation of P-bodies and SGs depends on a mass release of mRNA transcripts from polysomes. Cycloheximide treatment, which stalls ribosomes on transcripts without releasing them, prevents the generation of SGs in response to stress [60]. In contrast, complete dissociation of ribosomes from their transcripts by puromycin treatment promotes SG formation [60]. These observations suggested two fates for RNAs that escape decay mechanisms during cellular stress responses, whereby one set of transcripts remains actively translated while another is sequestered into SGs. This puzzling dichotomy has sparked interest in determining factors that dictate either state (Fig. 2).

Certain RNAs may associate with SGs due to intrinsic properties that inherently enhance their capacities for self-aggregation and LLPS. Both coding and non-coding SG-enriched RNAs tend to be longer than SG-depleted transcripts [61,62], likely reflecting the increased capacity of longer transcripts for RNA-RNA and RNA-protein interactions. Indeed, Van Treeck et al. [63] determined that out of total cellular RNA, longer RNA transcripts are more likely to self-aggregate, even in protein-free systems. SG-associated transcripts were found to be enriched in this list of self-aggregating RNAs, highlighting a model in which RNA recruitment into SGs may not be as protein-dependent as previously thought.

In addition to their longer length, SG-associated transcripts have been found to exhibit less secondary structure compared to excluded transcripts [64], a property that has been implicated in RNA aggregation. In both *Ashbya gossypii* cells and a cell-free LLPS system, Langdon et al. [65] demonstrated that certain mRNAs discriminately associate with distinct phase separated droplets, and that this preferential association is dependent upon specific secondary structure conformations. Thus, similar to their protein counterparts, SG-associated transcripts have less structural complexity, and this influences their ability to interact with other molecules. In general, SG-associated transcripts are longer and more disordered, promoting intermolecular interactions and aggregation.

Aside from a natural penchant for aggregation, RNAs may be recruited to SGs by sequence and structure driven protein-binding. For example, motif analysis of SG-associated transcripts in mouse fibroblasts showed an enrichment of AU-rich elements (AREs) under multiple stresses [62]. A number of ARE-binding proteins are enriched in SGs across eukaryotes and have been implicated in recruiting bound transcripts. The previously mentioned UBP1c, an RNA-binding protein associated with hypoxia tolerance in Arabidopsis, binds AREs in the 3′ UTRs of mRNA transcripts and localizes to SGs during hypoxic stress [9], while its fellow UBP1 homolog, UPB1b, has been shown to associate with SGs and protect bound transcripts from degradation during heat stress [36]. Additionally, ARE-binding TZF proteins colocalize with SG and P-body markers, and have been genetically implicated in abiotic and biotic stress responses in Arabidopsis [46,66] and rice [48,67]. However, while their importance for P-body-mediated mRNA turnover has been established [68], no direct role for these ARE-binding proteins in SG mRNA sequestration has been demonstrated.

In addition to AREs, the 5′ terminal oligopyrimidine (5′TOP) cis regulatory element, common in mammalian mRNAs encoding ribosomal proteins and translation factors [69], is associated with SG recruitment [70]. Single molecule imaging of HeLa cells under arsenite stress revealed that reporter mRNAs harboring the 5′TOP motif were more likely to localize to SGs than those lacking it, and their recruitment depended on binding by the La-related protein 1 (LARP1) protein [70]. In plants, only a fraction of ribosomal protein mRNAs contains the

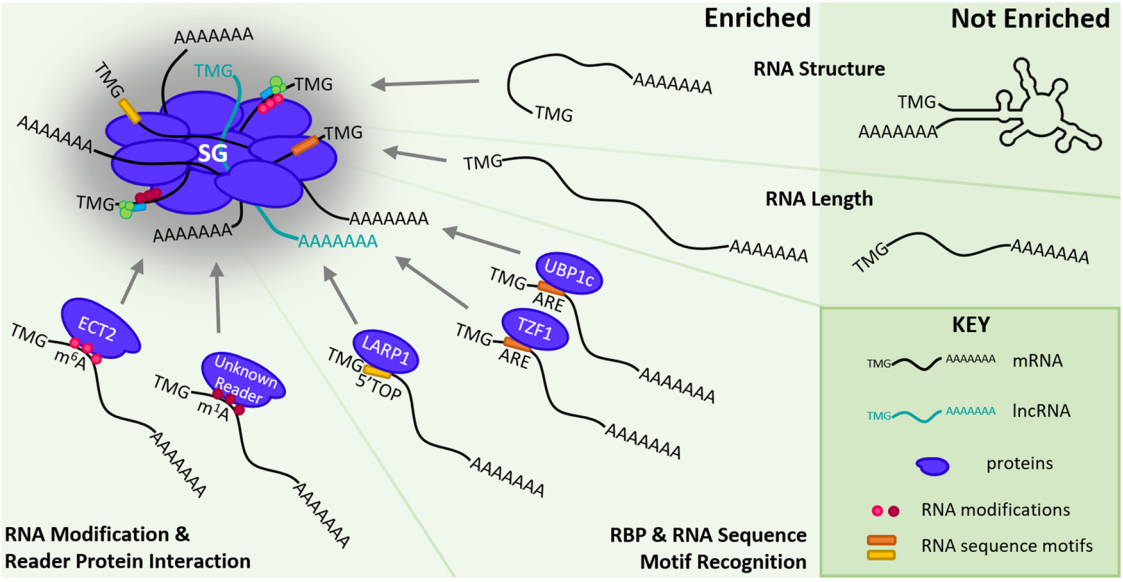

Fig. 2. Factors that influence RNA enrichment in SGs. Transcripts enriched in SGs tend to have less secondary structure and a longer length than those not enriched. SG RNAs frequently have AREs and 5′TOP elements, and may be recruited through the binding of RBPs like UBP1c, TZF1, and LARP1, which have been shown to localize to SGs. Interactions between reader proteins and RNA modifications like m^6A or m^1A may also influence SG localization. TMG, trimethylguanosine; ARE, AU-rich element; 5′TOP, 5′ terminal oligopyrimidine.

5′TOP motif. However, there is another plant-specific set of 5′TOP transcripts involved in ribosome biogenesis, and these transcripts are translationally regulated by LARP1 under normal conditions [71]. In Arabidopsis, LARP1 colocalizes to SGs in response to hypoxic stress, and although it has been shown to target transcripts for decay in response to heat stress, this appears to be independent of P-bodies [72]. Thus, in addition to RNA intrinsic sequence and structural features, many RNAs are likely specifically recruited into SGs through their protein-interaction partners.

Transcripts may also be targeted to SGs via post-transcriptional processes such as RNA base modifications. These modifications can impact an mRNA's propensity to form LLPS aggregates or to interact with SG-associated proteins. The methylation of adenosine at either the N6 (m^6A) or N1 (m^1A) position has been implicated in RNA recruitment to SGs in response to stress. Anders et al. [73] demonstrated that levels of m^6A modification significantly increase in human cell lines undergoing oxidative stress, mainly in the 5′ UTR and 5′ region of the coding sequences of SG-associated transcripts, while actively translated transcripts experienced minimal changes in m^6A levels. Heat and endoplasmic reticulum (ER) stress also resulted in increased m^6A modifications in human and mouse cell lines [74–76]. The m^6A reader YTHDF3 was shown to be required for m^6A detection in SGs [73], and interestingly, the reciprocal has been observed for YTHDF2, in that lack of m^6A in loss of function methyltransferase mutants or lack of reading capacity in YTHDF2 mutants resulted in hindered recruitment of YTHDF2 protein to SGs [77]. Taken together, these observations suggest that the modification of transcripts and the resulting interactions with readers can be important for both transcript and protein recruitment. Importantly, m^6A and the interaction of modified transcripts with readers is associated with enhanced LLPS, as heavily modified transcripts bind multivalently to readers with disordered domains [77]. Multiple m^6A reader proteins localize to SGs in plants, including YTH domain proteins ECT2 and ECT4 during heat stress [8,78], suggesting that their potential role in directing m^6A modified transcripts to SGs may be conserved. Conflicting evidence on the influence of m^6A on transcript recruitment to SGs was found by Khong et al. [79], who noted that a subset of poly-m^6A modified transcripts were recruited to SGs similarly in wild type and methyltransferase mutant cells, and that in general, modification alone was a poor predictor of SG localization. The authors suggest that the increased modification of SG-localized transcripts may simply be due to their longer length relative to excluded transcripts [79]. It is possible that m^6A and other modifications are crucial for only a specific set of transcripts or that they contribute to a more complex network of interactions that influence the overall likelihood of SG recruitment.

Similar to m^6A, the m^1A modification has also been shown to increase in response to oxidative and heat stress in human cell lines [80,81], particularly on SG-associated transcripts [80]. Interestingly, the addition of the m^1A motif from a SG-enriched transcript onto a reporter transcript led to a faster decrease in reporter translation upon stress induction and a more rapid increase in translation during recovery [80]. These observations suggest that in this context, the presence of m^1A allows for a more efficient regulation of translation in response to stress. Taken together, these data implicate RNA modifications in translational regulation and RNA targeting to SGs during stress. The exact mechanistic and regulatory connections between the epitranscriptome and SGs remain to be uncovered.

Across plant species, SG-associated RNA populations are composed of both translation-associated transcripts (e.g. ribosomal proteins and translation factors) and, interestingly, stress-response associated transcripts. For example, in response to heat stress in Arabidopsis, the interaction of ribosomal protein mRNAs with polysomes decreases with a concomitant increase in interaction with SG marker UBP1a, while their abundance in total RNA remains constant [82]. This change is reversed upon recovery, suggesting that these transcripts are sequestered in SGs during stress and released upon its resolution. Additionally, Juntawong & Bailey-Serres [83] showed that unanticipated darkness induces polysome dissociation from mRNAs associated with translation and ribosome biogenesis, which is rapidly reversed upon reillumination. The authors speculate that these transcripts are sequestered during light stress, as their abundance in the total RNA population remains unchanged throughout. While no direct connection was made in this work, a likely hypothesis is that these transcripts are sequestered into SGs or P-bodies for storage. The storage of transcripts associated with translation is likely to allow for efficient re-initiation of translation upon recovery [82].

The sequestration of stress-responsive transcripts seems more counter-intuitive, as their translation would likely promote survival

during stress or recovery. However, evidence suggests that stress-responsive mRNAs may indeed localize to SGs in certain contexts. For example, Tong et al. [41] recently demonstrated that ALBA proteins play a critical role in Arabidopsis thermotolerance by recruiting heat response-related transcripts to SGs, including several heat shock factors and dehydration-response element-binding protein 2 A (DREB2A). Interestingly, when these transcripts cannot be recruited into SGs in an *alba456* triple mutant, they are subsequently degraded by the mRNA decay machinery, likely in a P-body-dependent manner [41]. These observations indicate that ALBA-directed mRNA sequestration into SGs enhances transcript stability. Additionally, maize seedlings experiencing ER stress strongly induce the transcription of genes related to the unfolded protein response (UPR). For a portion of these upregulated transcripts, polysome association does not increase proportionally and the translation efficiency decreases [84]. In this same study, UPR-upregulated mRNAs were detected in SG-enriched fractions [84], suggesting that excess transcripts may be stored in SGs. Furthermore, Yan et al. [38] noted that the mRNA encoding gibberellic acid biosynthetic enzyme GA200×3, which is upregulated in response to salt stress and is required for growth under high salinity conditions, is bound and regulated by TSN, which localizes to SGs during salt stress. The authors surmise that TSN recruits GA200×3 mRNA to SGs to stabilize it and promote its optimal translation, though localization of the transcript to SGs was not examined [38]. In these instances, SG-sequestration of stress-responsive transcripts may serve to stabilize transcripts as they cycle between SGs and polysomes or to fine-tune protein expression levels when mRNAs are strongly upregulated. In contrast, Sorenson & Bailey-Serres [9] observed that hypoxia-related transcripts were upregulated under hypoxic conditions but continued to associate with polysomes rather than SG marker UBP1c. Therefore, the SG recruitment or continued translation of stress-responsive transcripts is likely context dependent.

Generally, characteristics like length, secondary structure, sequence motifs, and base modifications appear to influence RNA-RNA and RNA-protein interactions that result in any given RNA's recruitment to or exclusion from SGs. The specificity of these interactions and localizations is likely context-dependent. Overall, the mechanisms underlying the discriminant incorporation of mRNAs into SGs in response to various stresses remain in need of investigation.

4.2. LncRNAs in SGs

A relatively unexplored area in SG biology is the potential for long non-coding RNAs (lncRNAs) as core constituents, recruiters, or recruited transcripts. lncRNAs have been shown to be under-represented in SG-enriched RNA species [62], however this may be due to limitations in detection. LncRNAs expression levels are generally low compared to other RNA classes, and they tend to be tissue or cell-type specific [85,86]. Thus, studies examining SG RNA populations may have overlooked or not had the sequencing depth to monitor lncRNA constituents. Indeed, Khong et al. [61] found that, of the lncRNAs detected in cells undergoing oxidative stress, those that are enriched in SGs exhibit lower cellular expression levels than those depleted from SGs. In addition, many immunoprecipitation methods for isolating and analyzing SG-associated RNAs rely on stable interactions between RNAs and the SG proteins used for purification. However, it has been noted that lncRNA interactions with P-bodies are relatively short-lived [87], and similarly transient interactions may occur between lncRNAs and SGs. Coupling these brief interactions with the generally low and tissue-specific expression of most lncRNAs may make their SG association difficult to determine, and more targeted approaches may be necessary.

Despite these limitations, lncRNA interactions with SGs have been observed. For example, in human cell lines, Van Treek et al. [63] determined that natural antisense transcripts corresponding to SG-enriched sense transcripts were also enriched in SGs. These sense-antisense interactions may enhance LLPS propensity or actively recruit specific transcripts to SGs. Additionally, Khong et al. [61] found that the lncRNA non-coding RNA activated DNA damage (NORAD) is particularly enriched in SGs, with about 60% of its transcripts localizing to SGs. NORAD is known to sequester and consequently inhibit the function of PUMILIO (Pum) proteins, which repress translation of target mRNAs to promote cell division [88,89]. Pum2 has been shown to colocalize with SG markers during oxidative stress [90], though whether it is repressing translation of bound transcripts or being inactivated by NORAD remains to be seen. In another example, glutamine deprivation induces the formation of a ribonucleoprotein complex containing the glutamine insufficiency regulator of glutaminase lncRNA (GIRGL) and cell cycle associated protein 1 (CAPRIN1), which then sequesters glutaminase 1 (GLS1) mRNA in SGs [91]. Taken together, lncRNAs clearly have the potential to be key players in the formation of SGs or recruitment of target mRNAs, but intensive investigation will be required to fully parse their involvement.

4.3. The RNA experience in SGs

Stress granules have long been thought to be aggregates of translationally inactive mRNA transcripts, being sorted to P-bodies or awaiting stress relief before being reintroduced to the cytosol. The RNA population was considered to be composed of mRNAs bound by stalled ribosomal subunits primed to reenter the translational pool [92]. However, recent evidence shows that these structures are quite dynamic and may not be as translationally inactive as once thought. Transcripts have been shown to cycle from SGs to polysomes [93], and the authors suggest that cycling into SGs is important for transcript stability and may involve RNA modifications or protein-RNA interactions. This phenomenon may account for why stress-responsive transcripts are found in SGs, as they may constantly cycle between polysomes for translation and SGs to preserve their integrity and subsequently help mediate the stress response. There is conflicting evidence on whether transcripts migrate between SGs and P-bodies where they are thought to be degraded. Wilbertz et al. [70] reported that very few RNA molecules trafficked from SGs to P-bodies and none traveled in the reverse direction. In contrast, Moon et al. [94] observed bidirectional movement of transcripts between these two structures. This discrepancy may be due to experimental differences, such as cell type, single molecule tracking strategy, or mRNA reporter design. With the dynamic nature of these granules and their importance to RNA lifecycles, it is not unlikely that bidirectional RNA shuttling occurs, but more evidence is needed before a definitive conclusion can be made.

Recent evidence has challenged the assumption that SGs contain exclusively translationally stalled mRNA transcripts. Using integrated reporter systems and live cell imaging to simultaneously track transcripts, SGs, and active translation, Mateju et al. [95] observed translation of SG-associated reporter transcripts. They also noted that actively translated transcripts can move from the cytosol to SGs and vice versa while still polysome associated. In contrast, translationally inactive transcripts were less dynamic, generally remaining localized to either the cytosol or SGs [95]. These observations suggest that translation is not always repressed in SGs and that, contrary to the prevailing theory, transcripts do not need to dissociate from polysomes in order to associate with SGs. This idea is not entirely without precedent. Although it is not nearly as strong as the colocalization of small ribosomal subunits, overlap of large ribosomal subunits and SG marker foci has been detected in cells undergoing thapsigargin-induced ER stress [96]. Additionally, in response to combined arsenite stress and disrupted ribosomal degradation pathways, HeLa cell SGs accumulate the large ribosomal subunit component RPL19 [97]. These observations run counter to the current paradigm of SG biology, but may be biologically intuitive. In several instances described above, stress-responsive transcripts localize to SGs, and their continued translation may aid in the overall stress response. Further investigation into the regulation of

SG-based translation, as well as the extent to which this process occurs under biologically relevant contexts, is needed and may continue to reshape our understanding of SG biology.

Upon resolution of stress, SGs disperse and associated mRNAs are released back into the cytosol where they can more freely associate with polysomes. Several studies have suggested that the sequestration of transcripts in SGs during stress does not affect their ability to be translated or their stability once SGs have disbanded [70,82,94]. In particular, Merret et al. [82] noted that the release of transcripts related to translation and ribosome biogenesis that were sequestered in SGs during heat stress aided in the recovery following stress resolution. Importantly, N15 incorporation assays in the presence of transcription inhibitor actinomycin D demonstrated that newly generated ribosomes account for recovery-associated increase in polysomes, independent of newly transcribed ribosome-related transcripts [82]. These data indicate that transcripts stored in SGs during stress are translated upon their release. Interestingly, the m^6A modification implicated in transcript recruitment to SGs may also influence transcript translation upon recovery. Of all SG-associated transcripts identified in an arsenite-treated human cell line, a larger proportion of m^6A -modified transcripts were found to associate with ribosomes after stress relief than unmodified transcripts [98]. Overall, evidence points toward SGs serving as sites for the storage, stabilization, and oddly enough, translation, of associated transcripts, promoting survival during periods of stress and allowing for recovery when favorable conditions return.

5. Conclusions and open questions

SGs and P-bodies play critical roles in stress tolerance, an increasingly important concept for plants as climate change influences environmental factors like temperature, hydration, and salinity. How cellular components, through their association with these foci, contribute to stress tolerance has historically been understudied. However, paradigm shifting work in the past decade has added granularity to this field. For instance, we now better appreciate that SGs are not just composed of translationally stalled mRNAs, but also consist of stress-responsive transcripts and lncRNAs. Similarly, investigations into the protein composition revealed the presence of not only stress-responsive proteins, but also proteins that are inhibited under stress by association with SGs. In addition, many of the findings in plants resonate with those from mammals, demonstrating the deep conservation of these stress-response mechanisms. Many questions still remain, such as how proteins and RNA are selectively recruited into SGs and P-bodies through common protein or RNA motifs, intrinsically disordered protein domains, or promiscuous protein-RNA interactions; how the myriad RNA modifications found within the epitranscriptome influence transcript fate during and after stress; and why transcripts cycle between SGs, P-bodies, and polysomes. Our concept of what SGs and P-bodies are, what specific purposes they serve, and what happens to their associated proteins, metabolites, and RNAs is likely to continue to change as new work challenges established assumptions and addresses these outstanding questions.

Funding acknowledgement

This work was supported by NSF-PGRP 2023310 awarded to ADLN, NSF 2226270 awarded to AS and KAUST support for MC.

Data availability

No data was used for the research described in the article.

References

[1] D.S.W. Protter, R. Parker, Principles and properties of stress granules, Trends Cell Biol. 26 (2016) 668–679.
[2] Y. Luo, Z. Na, S.A. Slavoff, P-Bodies: composition, properties, and functions, Biochemistry 57 (2018) 2424–2431.
[3] N. Kedersha, P. Anderson, Stress granules: sites of mRNA triage that regulate mRNA stability and translatability, Biochem. Soc. Trans. 30 (2002) 963–969.
[4] N. Kedersha, G. Stoecklin, M. Ayodele, P. Yacono, J. Lykke-Andersen, M.J. Fritzler, D. Scheuner, R.J. Kaufman, D.E. Golan, P. Anderson, Stress granules and processing bodies are dynamically linked sites of mRNP remodeling, J. Cell Biol. 169 (2005) 871–884.
[5] J. Uniacke, W. Zerges, Stress induces the assembly of RNA granules in the chloroplast of Chlamydomonas reinhardtii, J. Cell Biol. 182 (2008) 641–646.
[6] F. Rafique, K.J. Lauersen, M. Chodasiewicz, N.E. Figueroa, A. New, Approach to the study of plastidial stress granules: the integrated use of and as model organisms, Plants 11 (2022), https://doi.org/10.3390/plants11111467.
[7] I. Maruri-López, N.E. Figueroa, I.E. Hernández-Sánchez, M. Chodasiewicz, Plant stress granules: trends and beyond, Front. Plant Sci. 12 (2021), 722643.
[8] M. Kosmacz, M. Gorka, S. Schmidt, M. Luzarowski, J.C. Moreno, J. Szlachetko, E. Leniak, E.M. Sokolowska, K. Sofroni, A. Schnittger, A. Skirycz, Protein and metabolite composition of Arabidopsis stress granules, N. Phytol. 222 (2019) 1420–1433.
[9] R. Sorenson, J. Bailey-Serres, Selective mRNA sequestration by OLIGOURIDYLATE-BINDING PROTEIN 1 contributes to translational control during hypoxia in Arabidopsis, Proc. Natl. Acad. Sci. U. S. A. 111 (2014) 2373–2378.
[10] E. Gutierrez-Beltran, P.N. Moschou, A.P. Smertenko, P.V. Bozhkov, Tudor staphylococcal nuclease links formation of stress granules and processing bodies with mRNA catabolism in Arabidopsis, Plant Cell 27 (2015) 926–943.
[11] L. Arribas-Hernández, S. Bressendorff, M.H. Hansen, C. Poulsen, S. Erdmann, P. Brodersen, An mA-YTH module controls developmental timing and morphogenesis in arabidopsis, Plant Cell 30 (2018) 952–967.
[12] H. Reuper, K. Amari, B. Krenz, Analyzing the G3BP-like gene family of Arabidopsis thaliana in early turnip mosaic virus infection, Sci. Rep. 11 (2021) 2187.
[13] C. Zampedri, M. Tinoco-Cuellar, S. Carrillo-Rosas, A. Diaz-Tellez, J.L. Ramos-Balderas, F. Pelegri, E. Maldonado, Zebrafish P54 RNA helicases are cytoplasmic granule residents that are required for development and stress resilience, Biol. Open 5 (2016) 1473–1484.
[14] A. Baguet, S. Degot, N. Cougot, E. Bertrand, M.-P. Chenard, C. Wendling, P. Kessler, H. Le Hir, M.-C. Rio, C. Tomasetto, The exon-junction-complex-component metastatic lymph node 51 functions in stress-granule assembly, J. Cell Sci. 120 (2007) 2774–2784.
[15] S. Kwon, Y. Zhang, P. Matthias, The deacetylase HDAC6 is a novel critical component of stress granules involved in the stress response, Genes Dev. 21 (2007) 3381–3394.
[16] T.S.K. Eisinger-Mathason, J. Andrade, A.L. Groehler, D.E. Clark, T.L. Muratore-Schroeder, L. Pasic, J.A. Smith, J. Shabanowitz, D.F. Hunt, I.G. Macara, D. A. Lannigan, Codependent functions of RSK2 and the apoptosis-promoting factor TIA-1 in stress granule assembly and cell survival, Mol. Cell. 31 (2008) 722–736.
[17] D. Teixeira, R. Parker, Analysis of P-body assembly in Saccharomyces cerevisiae, Mol. Biol. Cell. 18 (2007) 2274–2287.
[18] S. Huch, T. Nissan, An mRNA decapping mutant deficient in P body assembly limits mRNA stabilization in response to osmotic stress, Sci. Rep. 7 (2017) 44395.
[19] C. Weber, L. Nover, M. Fauth, Plant stress granules and mRNA processing bodies are distinct from heat stress granules, Plant J. 56 (2008) 517–530.
[20] X. Yu, B. Li, G.-J. Jang, S. Jiang, D. Jiang, J.-C. Jang, S.-H. Wu, L. Shan, P. He, Orchestration of processing body dynamics and mRNA decay in arabidopsis immunity, Cell Rep. 28 (2019) 2194–2205, e6.
[21] S. Jain, J.R. Wheeler, R.W. Walters, A. Agrawal, A. Barsic, R. Parker, ATPase-modulated stress granules contain a diverse proteome and substructure, Cell 164 (2016) 487–498.
[22] S. Markmiller, S. Soltanieh, K.L. Server, R. Mak, W. Jin, M.Y. Fang, E.-C. Luo, F. Krach, D. Yang, A. Sen, A. Fulzele, J.M. Wozniak, D.J. Gonzalez, M.W. Kankel, F.-B. Gao, E.J. Bennett, E. Lécuyer, G.W. Yeo, Context-dependent and disease-specific diversity in protein interactions within stress granules, Cell 172 (2018) 590–604, e13.
[23] M. Chodasiewicz, E.M. Sokolowska, A.C. Nelson-Dittrich, A. Masiuk, J.C. M. Beltran, A.D.L. Nelson, A. Skirycz, Identification and characterization of the heat-induced plastidial stress granules reveal new insight into stress response, Front. Plant Sci. 11 (2020), 595792.
[24] T.W. Han, M. Kato, S. Xie, L.C. Wu, H. Mirzaei, J. Pei, M. Chen, Y. Xie, J. Allen, G. Xiao, S.L. McKnight, Cell-free formation of RNA granules: bound RNAs identify features and components of cellular assemblies, Cell 149 (2012) 768–779.
[25] Y. Lin, D.S.W. Protter, M.K. Rosen, R. Parker, Formation and maturation of phase-separated liquid droplets by RNA-binding proteins, Mol. Cell. 60 (2015) 208–219.
[26] S.F. Banani, H.O. Lee, A.A. Hyman, M.K. Rosen, Biomolecular condensates: organizers of cellular biochemistry, Nat. Rev. Mol. Cell Biol. 18 (2017) 285–298.
[27] C. Nunes, I. Mestre, A. Marcelo, R. Koppenol, C.A. Matos, C. Nóbrega, MSGP: the first database of the protein components of the mammalian stress granules, Database. 2019 (2019). https://doi.org/10.1093/database/baz031.
[28] J.-Y. Youn, W.H. Dunham, S.J. Hong, J.D.R. Knight, M. Bashkurov, G.I. Chen, H. Bagci, B. Rathod, G. MacLeod, S.W.M. Eng, S. Angers, Q. Morris, M. Fabian, J.-F. Côté, A.-C. Gingras, High-density proximity mapping reveals the subcellular organization of mRNA-associated granules and bodies, Mol. Cell. 69 (2018) 517–532, e11.
[29] A. Hubstenberger, M. Courel, M. Bénard, S. Souquere, M. Ernoult-Lange, R. Chouaib, Z. Yi, J.-B. Morlot, A. Munier, M. Fradet, M. Daunesse, E. Bertrand, G. Pierron, J. Mozziconacci, M. Kress, D. Weil, P-body purification reveals the condensation of repressed mRNA regulons, Mol. Cell. 68 (2017) 144–157, e5.

[30] J.R. Buchan, R. Parker, Eukaryotic stress granules: the ins and outs of translation, Mol. Cell. 36 (2009) 932–941.
[31] J.-Y. Youn, B.J.A. Dyakov, J. Zhang, J.D.R. Knight, R.M. Vernon, J.D. Forman-Kay, A.-C. Gingras, Properties of stress granule and P-body proteomes, Mol. Cell. 76 (2019) 286–294.
[32] N. Gilks, N. Kedersha, M. Ayodele, L. Shen, G. Stoecklin, L.M. Dember, P. Anderson, Stress granule assembly is mediated by prion-like aggregation of TIA-1, Mol. Biol. Cell. 15 (2004) 5383–5398.
[33] N. Kedersha, M.D. Panas, C.A. Achorn, S. Lyons, S. Tisdale, T. Hickman, M. Thomas, J. Lieberman, G.M. McInerney, P. Ivanov, P. Anderson, G3BP-Caprin1-USP10 complexes mediate stress granule condensation and associate with 40S subunits, J. Cell Biol. 212 (2016) 845–860.
[34] M. Kosmacz, M. Luzarowski, O. Kerber, E. Leniak, E. Gutiérrez-Beltrán, J. C. Moreno, M. Gorka, J. Szlachetko, D. Veyel, A. Graf, A. Skirycz, Interaction of 2′,3′-cAMP with Rbp47b Plays a Role in Stress Granule Formation, Plant Physiol. 177 (2018) 411–421.
[35] E. Gutiérrez-Beltran, P.V. Bozhkov, P.N. Moschou, Tudor Staphylococcal Nuclease plays two antagonistic roles in RNA metabolism under stress, Plant Signal. Behav. 10 (2015), e1071005.
[36] C.C. Nguyen, K. Nakaminami, A. Matsui, S. Kobayashi, Y. Kurihara, K. Toyooka, M. Tanaka, M. Seki, Oligouridylate binding protein 1b plays an integral role in plant heat stress tolerance, Front. Plant Sci. 7 (2016) 853.
[37] E. Gutierrez-Beltran, T.V. Denisenko, B. Zhivotovsky, P.V. Bozhkov, Tudor staphylococcal nuclease: biochemistry and functions, Cell Death Differ. 23 (2016) 1739–1748.
[38] C. Yan, Z. Yan, Y. Wang, X. Yan, Y. Han, Tudor-SN, a component of stress granules, regulates growth under salt stress by modulating GA20ox3 mRNA levels in Arabidopsis, J. Exp. Bot. 65 (2014) 5933–5944.
[39] X. Gao, X. Fu, J. Song, Y. Zhang, X. Cui, C. Su, L. Ge, J. Shao, L. Xin, J. Saarikettu, M. Mei, X. Yang, M. Wei, O. Silvennoinen, Z. Yao, J. He, J. Yang, Poly(A)(+) mRNA-binding protein Tudor-SN regulates stress granules aggregation dynamics, FEBS J. 282 (2015) 874–890.
[40] S. Zhu, J. Gu, J. Yao, Y. Li, Z. Zhang, W. Xia, Z. Wang, X. Gui, L. Li, D. Li, H. Zhang, C. Liu, Liquid-liquid phase separation of RBGD2/4 is required for heat stress resistance in Arabidopsis, Dev. Cell. 57 (2022) 583–597, e6.
[41] J. Tong, Z. Ren, L. Sun, S. Zhou, W. Yuan, Y. Hui, D. Ci, W. Wang, L.-M. Fan, Z. Wu, W. Qian, ALBA proteins confer thermotolerance through stabilizing HSF messenger RNAs in cytoplasmic granules, Nat. Plants 8 (2022) 778–791.
[42] X. Tian, Z. Qin, Y. Zhao, J. Wen, T. Lan, L. Zhang, F. Wang, D. Qin, K. Yu, A. Zhao, Z. Hu, Y. Yao, Z. Ni, Q. Sun, I. De Smet, H. Peng, M. Xin, Stress granule-associated TaMBF1c confers thermotolerance through regulating specific mRNA translation in wheat (Triticum aestivum), N. Phytol. 233 (2022) 1719–1731.
[43] C. Perea-Resa, C. Carrasco-López, R. Catalá, V. Turečková, O. Novak, W. Zhang, L. Sieburth, J.M. Jiménez-Gómez, J. Salinas, The LSM1-7 complex differentially regulates arabidopsis tolerance to abiotic stress conditions by promoting selective mRNA decapping, Plant Cell 28 (2016) 505–520.
[44] J. Xu, N.-H. Chua, Dehydration stress activates Arabidopsis MPK6 to signal DCP1 phosphorylation, EMBO J. 31 (2012) 1975–1984.
[45] M. Chodasiewicz, O. Kerber, M. Gorka, J.C. Moreno, I. Maruri-Lopez, R.I. Minen, A. Sampathkumar, A.D.L. Nelson, A. Skirycz, 2′,3′-cAMP treatment mimics the stress molecular response in Arabidopsis thaliana, Plant Physiol. 188 (2022) 1966–1978.
[46] M.C. Pomeranz, C. Hah, P.-C. Lin, S.G. Kang, J.J. Finer, P.J. Blackshear, J.-C. Jang, The Arabidopsis tandem zinc finger protein AtTZF1 traffics between the nucleus and cytoplasmic foci and binds both DNA and RNA, Plant Physiol. 152 (2010) 151–165.
[47] S. Bogamuwa, J.-C. Jang, The Arabidopsis tandem CCCH zinc finger proteins AtTZF4, 5 and 6 are involved in light-, abscisic acid- and gibberellic acid-mediated regulation of seed germination, Plant Cell Environ. 36 (2013) 1507–1519.
[48] A. Jan, K. Maruyama, D. Todaka, S. Kidokoro, M. Abo, E. Yoshimura, K. Shinozaki, K. Nakashima, K. Yamaguchi-Shinozaki, OsTZF1, a CCCH-tandem zinc finger protein, confers delayed senescence and stress tolerance in rice by regulating stress-related genes, Plant Physiol. 161 (2013) 1202–1216.
[49] G. Stoecklin, N. Kedersha, Relationship of GW/P-bodies with stress granules, Adv. Exp. Med. Biol. 768 (2013) 197–211.
[50] R. Parker, U. Sheth, P bodies and the control of mRNA translation and degradation, Mol. Cell. 25 (2007) 635–646.
[51] T. Chantarachot, J. Bailey-Serres, Polysomes, stress granules, and processing bodies: a dynamic triumvirate controlling cytoplasmic mRNA fate and function, Plant Physiol. 176 (2018) 254–269.
[52] A. Aizer, Y. Brody, L.W. Ler, N. Sonenberg, R.H. Singer, Y. Shav-Tal, The dynamics of mammalian P body transport, assembly, and disassembly in vivo, Mol. Biol. Cell. 19 (2008) 4154–4166.
[53] A. Patel, L. Malinovska, S. Saha, J. Wang, S. Alberti, Y. Krishnan, A.A. Hyman, ATP as a biological hydrotrope, Science 356 (2017) 753–756.
[54] R. Dandage, A. Bandyopadhyay, G.G. Jayaraj, K. Saxena, V. Dalal, A. Das, K. Chakraborty, Classification of chemical chaperones based on their effect on protein folding landscapes, ACS Chem. Biol. 10 (2015) 813–820.
[55] G. Cereghetti, C. Wilson-Zbinden, V.M. Kissling, M. Diether, A. Arm, H. Yoo, I. Piazza, S. Saad, P. Picotti, D.A. Drummond, U. Sauer, R. Dechant, M. Peter, Reversible amyloids of pyruvate kinase couple cell metabolism and stress granule disassembly, Nat. Cell Biol. 23 (2021) 1085–1094.
[56] E. Gutierrez-Beltran, P.H. Elander, K. Dalman, G.W. Dayhoff 2nd, P.N. Moschou, V. N. Uversky, J.L. Crespo, P.V. Bozhkov, Tudor staphylococcal nuclease is a docking platform for stress granule components and is essential for SnRK1 activation in Arabidopsis, EMBO J. 40 (2021), e105043.
[57] K. Begovich, A.Q. Vu, G. Yeo, J.E. Wilhelm, Conserved metabolite regulation of stress granule assembly via AdoMet, J. Cell Biol. 219 (2020), https://doi.org/10.1083/jcb.201904141.
[58] A. Eulalio, I. Behm-Ansmant, E. Izaurralde, P bodies: at the crossroads of post-transcriptional pathways, Nat. Rev. Mol. Cell Biol. 8 (2007) 9–22.
[59] N. Standart, D. Weil, P-bodies: cytosolic droplets for coordinated mRNA Storage, Trends Genet 34 (2018) 612–626.
[60] N. Kedersha, M.R. Cho, W. Li, P.W. Yacono, S. Chen, N. Gilks, D.E. Golan, P. Anderson, Dynamic shuttling of TIA-1 accompanies the recruitment of mRNA to mammalian stress granules, J. Cell Biol. 151 (2000) 1257–1268.
[61] A. Khong, T. Matheny, S. Jain, S.F. Mitchell, J.R. Wheeler, R. Parker, The stress granule transcriptome reveals principles of mRNA accumulation in stress granules, Mol. Cell. 68 (2017) 808–820, e5.
[62] S. Namkoong, A. Ho, Y.M. Woo, H. Kwak, J.H. Lee, Systematic characterization of stress-induced RNA granulation, Mol. Cell. 70 (2018) 175–187, e8.
[63] B. Van Treeck, D.S.W. Protter, T. Matheny, A. Khong, C.D. Link, R. Parker, RNA self-assembly contributes to stress granule formation and defining the stress granule transcriptome, Proc. Natl. Acad. Sci. U. S. A. 115 (2018) 2734–2739.
[64] A. Vandelli, F. Cid Samper, M. Torrent Burgas, N. Sanchez de Groot, G.G. Tartaglia, The interplay between disordered regions in RNAs and proteins modulates interactions within stress granules and processing bodies, J. Mol. Biol. 434 (2022), 167159.
[65] E.M. Langdon, Y. Qiu, A. Ghanbari Niaki, G.A. McLaughlin, C.A. Weidmann, T. M. Gerbich, J.A. Smith, J.M. Crutchley, C.M. Termini, K.M. Weeks, S. Myong, A. S. Gladfelter, mRNA structure determines specificity of a polyQ-driven phase separation, Science 360 (2018) 922–927.
[66] P.-C. Lin, M.C. Pomeranz, Y. Jikumaru, S.G. Kang, C. Hah, S. Fujioka, Y. Kamiya, J.-C. Jang, The Arabidopsis tandem zinc finger protein AtTZF1 affects ABA- and GA-mediated growth, stress and gene expression responses, Plant J. 65 (2011) 253–268.
[67] C. Guo, L. Chen, Y. Cui, M. Tang, Y. Guo, Y. Yi, Y. Li, L. Liu, L. Chen, RNA binding protein OsTZF7 traffics between the nucleus and processing bodies/stress granules and positively regulates drought stress in rice, Front. Plant Sci. 13 (2022), 802337.
[68] J. Qu, S.G. Kang, W. Wang, K. Musier-Forsyth, J.-C. Jang, The Arabidopsis thaliana tandem zinc finger 1 (AtTZF1) protein in RNA binding and decay, Plant J. 78 (2014) 452–467.
[69] V. Iadevaia, S. Caldarola, E. Tino, F. Amaldi, F. Loreni, All translation elongation factors and the e, f, and h subunits of translation initiation factor 3 are encoded by 5′-terminal oligopyrimidine (TOP) mRNAs, RNA 14 (2008) 1730–1736.
[70] J.H. Wilbertz, F. Voigt, I. Horvathova, G. Roth, Y. Zhan, J.A. Chao, Single-Molecule Imaging of mRNA Localization and Regulation during the Integrated Stress Response, Mol. Cell. 73 (2019) 946–958, e7.
[71] M.R. Scarpin, S. Leiboff, J.O. Brunkard, Parallel global profiling of plant TOR dynamics reveals a conserved role for LARP1 in translation, Elife 9 (2020), https://doi.org/10.7554/eLife.58795.
[72] R. Merret, J. Descombin, Y.-T. Juan, J.-J. Favory, M.-C. Carpentier, C. Chaparro, Y.-Y. Charng, J.-M. Deragon, C. Bousquet-Antonelli, XRN4 and LARP1 are required for a heat-triggered mRNA decay pathway involved in plant acclimation and survival during thermal stress, Cell Rep. 5 (2013) 1279–1293.
[73] M. Anders, I. Chelysheva, I. Goebel, T. Trenkner, J. Zhou, Y. Mao, S. Verzini, S.-B. Qian, Z. Ignatova, Dynamic mA methylation facilitates mRNA triaging to stress granules, Life Sci. Alliance 1 (2018) e201800113.
[74] Y. Fu, X. Zhuang, mA-binding YTHDF proteins promote stress granule formation, Nat. Chem. Biol. 16 (2020) 955–963.
[75] J. Zhou, J. Wan, X. Gao, X. Zhang, S.R. Jaffrey, S.-B. Qian, Dynamic m(6)A mRNA methylation directs translational control of heat shock response, Nature 526 (2015) 591–594.
[76] K.D. Meyer, D.P. Patil, J. Zhou, A. Zinoviev, M.A. Skabkin, O. Elemento, T. V. Pestova, S.-B. Qian, S.R. Jaffrey, 5′ UTR m(6)A Promotes Cap-Independent Translation, Cell 163 (2015) 999–1010.
[77] R.J. Ries, S. Zaccara, P. Klein, A. Olarerin-George, S. Namkoong, B.F. Pickering, D. P. Patil, H. Kwak, J.H. Lee, S.R. Jaffrey, m6A enhances the phase separation potential of mRNA, Nature 571 (2019) 424–428.
[78] J. Scutenaire, J.-M. Deragon, V. Jean, M. Benhamed, C. Raynaud, J.-J. Favory, R. Merret, C. Bousquet-Antonelli, The YTH domain protein ECT2 Is an mA reader required for normal trichome branching in arabidopsis, Plant Cell 30 (2018) 986–1005.
[79] A. Khong, T. Matheny, T.N. Huynh, V. Babl, R. Parker, Limited effects of mA modification on mRNA partitioning into stress granules, Nat. Commun. 13 (2022) 3735.
[80] M. Alriquet, G. Calloni, A. Martínez-Limón, R. Delli Ponti, G. Hanspach, M. Hengesbach, G.G. Tartaglia, R.M. Vabulas, The protective role of m1A during stress-induced granulation, J. Mol. Cell Biol. 12 (2020) 870–880.
[81] D. Dominissini, S. Nachtergaele, S. Moshitch-Moshkovitz, E. Peer, N. Kol, M.S. Ben-Haim, Q. Dai, A. Di Segni, M. Salmon-Divon, W.C. Clark, G. Zheng, T. Pan, O. Solomon, E. Eyal, V. Hershkovitz, D. Han, L.C. Doré, N. Amariglio, G. Rechavi, C. He, The dynamic N(1)-methyladenosine methylome in eukaryotic messenger RNA, Nature 530 (2016) 441–446.
[82] R. Merret, M.-C. Carpentier, J.-J. Favory, C. Picart, J. Descombin, C. Bousquet-Antonelli, P. Tillard, L. Lejay, J.-M. Deragon, Y.-Y. Charng, Heat shock protein HSP101 affects the release of ribosomal protein mRNAs for recovery after heat shock, Plant Physiol. 174 (2017) 1216–1225.
[83] P. Juntawong, J. Bailey-Serres, Dynamic light regulation of translation status in arabidopsis thaliana, Front. Plant Sci. 3 (2012) 66.

[84] P. Kanodia, P. Vijayapalani, R. Srivastava, R. Bi, P. Liu, W.A. Miller, S.H. Howell, Control of translation during the unfolded protein response in maize seedlings: Life without PERKs, Plant Direct 4 (2020), e00241.
[85] M.N. Cabili, C. Trapnell, L. Goff, M. Koziol, B. Tazon-Vega, A. Regev, J.L. Rinn, Integrative annotation of human large intergenic noncoding RNAs reveals global properties and specific subclasses, Genes Dev. 25 (2011) 1915–1927.
[86] K. Palos, A.C. Nelson Dittrich, L. ang Yu, J.R. Brock, C.E. Railey, H.-Y.L. Wu, E. Sokolowska, A. Skirycz, P.Y. Hsu, B.D. Gregory, E. Lyons, M.A. Beilstein, A.D. L. Nelson, Identification and functional annotation of long intergenic non-coding RNAs in Brassicaceae, Plant Cell (2022), https://doi.org/10.1093/plcell/koac166.
[87] S. Pitchiaya, M.D.A. Mourao, A.P. Jalihal, L. Xiao, X. Jiang, A.M. Chinnaiyan, S. Schnell, N.G. Walter, Dynamic recruitment of single RNAs to processing bodies depends on RNA functionality, Mol. Cell. 74 (2019) 521–533, e6.
[88] S. Lee, F. Kopp, T.-C. Chang, A. Sataluri, B. Chen, S. Sivakumar, H. Yu, Y. Xie, J. T. Mendell, Noncoding RNA NORAD regulates genomic stability by sequestering PUMILIO Proteins, Cell 164 (2016) 69–80.
[89] A. Tichon, N. Gil, Y. Lubelsky, T. Havkin Solomon, D. Lemze, S. Itzkovitz, N. Stern-Ginossar, I. Ulitsky, A conserved abundant cytoplasmic long noncoding RNA modulates repression by Pumilio proteins in human cells, Nat. Commun. 7 (2016) 12209.
[90] J.P. Vessey, A. Vaccani, Y. Xie, R. Dahm, D. Karra, M.A. Kiebler, P. Macchi, Dendritic localization of the translational repressor Pumilio 2 and its contribution to dendritic stress granules, J. Neurosci. 26 (2006) 6496–6508.
[91] R. Wang, L. Cao, R.F. Thorne, X.D. Zhang, J. Li, F. Shao, L. Zhang, M. Wu, LncRNA GIRGL drives CAPRIN1-mediated phase separation to suppress glutaminase-1 translation under glutamine deprivation, Sci. Adv. 7 (2021), https://doi.org/10.1126/sciadv.abe5708.
[92] N. Kedersha, S. Chen, N. Gilks, W. Li, I.J. Miller, J. Stahl, P. Anderson, Evidence that ternary complex (eIF2-GTP-tRNA(i)(Met))-deficient preinitiation complexes are core constituents of mammalian stress granules, Mol. Biol. Cell. 13 (2002) 195–210.
[93] S. Mollet, N. Cougot, A. Wilczynska, F. Dautry, M. Kress, E. Bertrand, D. Weil, Translationally repressed mRNA transiently cycles through stress granules during stress, Mol. Biol. Cell. 19 (2008) 4469–4479.
[94] S.L. Moon, T. Morisaki, A. Khong, K. Lyon, R. Parker, T.J. Stasevich, Multicolour single-molecule tracking of mRNA interactions with RNP granules, Nat. Cell Biol. 21 (2019) 162–168.
[95] D. Mateju, B. Eichenberger, F. Voigt, J. Eglinger, G. Roth, J.A. Chao, Single-molecule imaging reveals translation of mRNAs localized to stress granules, Cell 183 (2020) 1801–1812, e13.
[96] S.R. Kimball, R.L. Horetsky, D. Ron, L.S. Jefferson, H.P. Harding, Mammalian stress granules represent sites of accumulation of stalled translation initiation complexes, Am. J. Physiol. Cell Physiol. 284 (2003) C273–C284.
[97] S.J. Seguin, F.F. Morelli, J. Vinet, D. Amore, S. De Biasi, A. Poletti, D. C. Rubinsztein, S. Carra, Inhibition of autophagy, lysosome and VCP function impairs stress granule assembly, Cell Death Differ. 21 (2014) 1838–1851.
[98] S. Das, L. Santos, A.V. Failla, Z. Ignatova, mRNAs sequestered in stress granules recover nearly completely for translation, RNA Biol. 19 (2022) 877–884.