Mechanism and effect of stress granule formation in cancer and its potential roles in breast cancer therapy
Taobo Hu a, Wei Hou b, Enhua Xiao c,**, Mengping Long b,c,*

a Department of Breast Surgery, Peking University People’s Hospital, Beijing 100044, PR China
b Department of Pathology, Peking University Cancer Hospital, Beijing 100142, PR China
c Department of Radiology, The Second Xiangya Hospital, Central South University, Changsha, Hunan 410011, PR China

Received 19 December 2020; received in revised form 4 February 2021; accepted 10 February 2021
Available online 23 February 2021

KEYWORDS
Apoptosis; Breast cancer; Drug resistance; Stress granules; Translation initiation

Abstract Stress granules are non-membranous cytoplasmic foci induced by various stress conditions. It is a protective strategy used by cells to suppress overall translation during stress. In cancer cells, it was thought that the formation of stress granules could protect them from apoptosis and induces resistance towards anti-cancer drugs or radiation treatment which makes the stress granules a potential target for cancer treatment. However, most of our understanding of stress granules are still in the stage of molecular and cell biology, and a transitional gap for its actual effect on clinical settings remains. In this review, we summarize the mechanism and effect of stress granules formation in cancer and try to illuminate its potential applications in cancer therapy, using breast cancer as an example.

Copyright © 2021, Chongqing Medical University. Production and hosting by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

Introduction
In animals, stress granules (SGs) were first named and morphologically characterized by Collier et al in 1988 as dense cytoplasmic bodies formed in restressed chicken embryonic fibroblast cells. Subsequent studies have revealed that SGs can be formed in many other eukaryotic cells ranging from yeast to human cells and induced by various stress conditions including heat, oxidative stress, and hypoxia, indicating that the formation of SGs is an evolutionally conservative strategy to protect cells from stress conditions. The detailed structure, composition, and function of SGs have been extensively studied during the past three decades, mainly using laboratory cancer cell lines. Noteworthily, SGs have also been recently detected in vivo, both in human pancreatic adenocarcinoma tissue...
and mouse xenograft of human osteosarcoma cell line.\textsuperscript{4,5} Specific oncogenic mutations including KRAS mutations in pancreatic adenocarcinoma and DDX3X mutations in medulloblastoma have been found to drive the spontaneous formation of SGs.\textsuperscript{4,6}

Messenger RNA (mRNA), RNA-binding proteins TIA-1 and TIAR was the first identified components of SGs.\textsuperscript{7} Later, an inactivated 48S initiation complex which is composed of translation initiation factors eIF, eIF4G, eIF3, and 40S ribosome subunit was detected in SGs.\textsuperscript{8,9} The compositions indicate SGs is a type of messenger ribonucleoprotein particle (mRNP) similar to other mRNPs like Cajal bodies and P bodies. After exposure to stress, two subsequent processes were necessary for SGs formation, the inhibition of translation initiation\textsuperscript{10,17} and RNA-protein aggregation nucleated by RNA-binding proteins.\textsuperscript{11} The interactions contributed to SGs formation include RNA–protein, RNA–RNA interactions and protein–protein interactions.\textsuperscript{12} Proteomic analysis of SGs proteins found that 15% of SGs proteins have Prion-like domain (PrLD) which is a type of intrinsically disordered region (IDR) and more than 50% of stress granules proteins have RNA-recognition motif.\textsuperscript{13,14} For many stress granule proteins, these two domains are found to be essential for their recruitment of SGs.\textsuperscript{15–17} Both domains and their combination are also important for the inducing of liquid–liquid phase separation (LLPS).\textsuperscript{3,5} Given the dynamic property of SGs, they are considered to be liquid droplets separated from cytoplasm through complex RNA and protein interactions.\textsuperscript{20} Meanwhile, super-resolution fluorescent microscopy and FRAP examination of SGs revealed that SGs harbored a biphasic structure, composed of multiple high-density solid cores and surrounding shell structure which is believed to be formed by LLPS. The cores harboring mRNA and proteins are relatively more condense and stable than the out shell structure which is more dynamically flexible with proteins transporting in and out rapidly.\textsuperscript{19,21} The composition of SGs formed in different conditions can be diverse. Different from the arsenite-induced SGs which are called canonical SGs, selenite induced SGs lacked many important classical proteins including RACK1 while chronic nutrition starvation-induced SGs lack 40S ribosome which is present in all of the other reported SGs.\textsuperscript{22,23}

Functionally, the formation of SGs is closely related to cancer, neurodegeneration diseases, and viral infection.\textsuperscript{24–26} In this review, we focus on the role of stress granules in cancer development and progress, aiming to summarize and provide more details about the association between SGs and cancer cell behavior.

### The effect of chemotherapy on the formation and disassembly of SGs

It is known that cancer cells are continuously confronted with environmental stresses including hypoxia, the toxicity of chemotherapy and radiation therapy and their responses to stresses make a critical aspect of their biological behavior.\textsuperscript{27–29} The formation of SGs is one type of protective strategy for cancer cells to survive from stresses. Numerous studies have reported the formation and function of SGs in cancers mostly using cancer cell lines, only one paper has reported the existence of SGs in human cancer tissues,\textsuperscript{30} probably due to the invisibility of SGs under normal optical microscopy and its tendency to disassemble when stress is relieved. Many anti-cancer chemicals can affect the formation and the disassembly of SGs.

### Formation of SGs

Many anti-cancer chemicals can induce SGs. They cover a wide range of anti-cancer working mechanisms from anti-metabolites like 5-fluorouracil (5-FU) to tyrosine kinase inhibitors like sorafenib. They also induce SGs through different mechanisms and signaling pathways which are summarized and presented in Table 1 and Fig. 1. Here, we focused on the current and potential therapeutics in breast cancer.

5-Fluorouracil (5-FU), one of the oldest and effective therapy for breast cancer, was reported to be SGs inducing.\textsuperscript{30} Its pro-drug capecitabine is widely used for metastatic breast cancer. Experiments have shown that the therapeutic dose of 5-FU can induce the formation of SGs in cultured HeLa cells. Among all the activities, its incorporation into RNA is necessary for promoting SGs formation while others are not.\textsuperscript{31} Noteworthy, other two FDA-approved RNA-incorporating anti-cancer drugs, 5-azacitidine and 6-thioguanine can also induce the formation of SGs although in much higher concentration than their clinical dosage.\textsuperscript{31} SGs formation induced by the RNA-

| Drug generic name | Anti-cancer mechanism | SGs inducing target | Ref. |
|-------------------|-----------------------|---------------------|-----|
| 5-FU*             | Anti-metabolic;       | eIF2α (PKR)         | 30,  |
|                   | DNA and RNA           |                     | 31  |
|                   | incorporation         |                     |     |
| Sorafenib*        | Tyrosine kinase       | eIF2α (PERK)        | 32,  |
|                   | inhibitor             |                     | 36  |
| Lapatinib*        | Tyrosine kinase       | eIF2α (PERK)        | 37  |
|                   | inhibitor             |                     |     |
| Selenite*         | ROS inducing;         | eIF4E               | 22,  |
|                   | Antophagy inhibiting  |                     | 67  |
| MG132*            | Proteasome inhibitor  | eIF2α (GCN2)        | 51  |
| Bortezomib        | 26S proteasome        | eIF2α (HRI)         | 51,  |
|                   | inhibitor             |                     | 90  |
| Arsenite          | ROS Inducing;         | eIF2α (HRI, PERK)   | 74,  |
|                   | Cell cycle arrest     |                     | 87  |
| Thapsigargin      | ER stress Inducing    | eIF2α (PERK)        | 40  |
| PateamineA        | Translation inhibition| eIF4A               | 42  |
|                   |                       |                     |     |
| Hippuristanol     | Translation inhibition| eIF4A               | 41  |
| Silvestrol        | Translation inhibition| eIF4A               | 41  |
| Oxaliplatin       | DNA crosslinking       | eIF2α               | 46  |
|                   | Immunogenic cell death|                     | 48  |

Ps. The # marked entries are physiologically relevant drugs that can possibly induce SGs in their therapeutic concentrations.
incorporating pathway was mediated by the phosphorylation of eIF2α. The detailed underlying mechanism, along with the question of whether all RNA-incorporating drugs are able to induce SGs formation remain to be revealed.

Sorafenib is a multikinase inhibitor targeting BRAF, CRAF, and VEGFR.32 Although it is currently used to treat advanced renal cell carcinoma and hepatocellular carcinoma, lots of trials have been launched to evaluate its effect as monotherapy or combined with others for advanced breast cancer.33–35 Sorafenib can induce SGs formation in various cancer cell lines via phosphorylation of eIF2α by PERK. Disruption of the SGs formation would make cells more sensitive to the sorafenib.36 Another tyrosine kinase inhibitor lapatinib was shown to be able to induce SGs formation in T-47D breast cancer cell line at therapeutic level.37 Lapatinib targeting HER2 and EGFR pathway and is used in combination therapy for HER2-overexpressed breast cancer. Same with sorafenib, it induces SGs formation via phosphorylation of eIF2α by PERK. Moreover, both sorafenib and lapatinib were found to activate PERK via the altered expression of GRP48/BiP, an ER chaperone binding to PERK to inhibit its dimerization.38,39 Thus a potential mechanism about the controlling role of GRP48/BiP in SGs formation was worth further exploration.

Other SGs inducing breast cancer therapeutics include selenite which induces SGs through generating ROS,23 thapsigargin is an extract from the plant and can induce SGs by generating ER stresses,40 both of them are currently in the clinical trial phase. Three eIF4A inhibitors, pateamine, hippuristanol, and silvestrol are also proved to promote SG formation and they all showed a certain degree of anti-cancer activity for both cell lines and xenograft tumors.41–44

Cisplatin, as common salvage chemotherapy and a potential first-line therapy in BRCA mutated patients, failed to induce SGs formation. While its analog oxaliplatin which was applied in ovarian and colorectal cancer was found to have the activity of SGs inducing via phosphorylation of eIF2α.45–48 It indicates the SGs inducing is not related to the shared DNA damage effect between them but is caused by the extra effect of oxaliplatin like the inducing of immunogenic cell death.49,50

It’s worth reminding readers that different cancer cell lines have different SG forming sensitivity towards specific stress conditions. For example, SGs can be successfully formed in many cell lines including Hela and Calu-1 by bortezomib, while its formation cannot be found in breast cancer cell line Hs578T,51 indicating that SGs formation is under sophisticated regulation. Thus, for the study of SGs, the experiments and conclusions should be processed in a drug- and cancer-specific manner.

SGs assembly pathways targeting by different anti-cancer chemicals

As mentioned above, translation initiation inhibition and macromolecular aggregation are two core steps for SGs forming. Although the above chemicals induce SGs through different initiating mechanisms, all of them targeted specific translation initiation factors in the process of SGs forming (see Fig. 1).

The eIF2α is a translation initiation factor whose phosphorylation at Ser51 serves as a classic SGs inducing step. Once eIF2 is phosphorylated at Ser51, it would bind strongly with eIF2B, a guanidine exchange factor (GEF) to form a
stable p-eIF2α-eIF2B complex. The GEF function of eIF2B promoting the conversion between eIF2α-GDP and eIF2α-GTP is disabled due to the tight and rigid interaction with p-eIF2α. Subsequently, the inactive eIF2α-GDP complex accumulates in cells with inadequate eIF2-GRP for translation initiation.52 eIF2α ser51 can be phosphorylated by four kinases PRK, PERK, GCN2 and HRI. For anti-cancer chemicals, MG132 promotes SGs formation through phosphorylation of eIF2α Ser51 by GCN2 while bortezomib enhances the eIF2α phosphorylation by HRI.31 PERK activation is targeted by sorafenib.51 Arsenite induced SG formation also involves eIF2α phosphorylation by HRI, PKR and PEKR. 5-FU can also induce the phosphorylation of eIF2α through PKR. In some SG forming conditions, eIF2α phosphorylation can be observed but are not necessary for SG formation, as in the case of selenite and H2O2.53,23 In a screening experiment observed but are not necessary for SG formation, as in the case of selenite and H2O2.53,23 In a screening experiment.

The eIF4F complex is another essential translation initiation complex that can capture the 5’ m7 cap of mRNA and activate mRNA for translation. eIF4F complex is comprised of three individual proteins eIF4A, eIF4E and eIF4G. Inactivation of any of them would cause translation initiation arrest and SGs formation.53 Small molecule compounds, pateamine, hippuristanol and silvestrol are proved to promote SG formation through inhibiting eIF4A.54-56 Two prostaglandins 15-d-PGJ2 and PGA1 can bind directly with eIF4A to inhibit the binding between eIF4A and eIF4G, thus suppressing the formation of eIF4F complex.57 The binding between eIF4E and eIF4G is also necessary for the assembly of eIF4F complex. eIF4E binding protein (eIF4EBP) binds to the dorsal surface of eIF4E which is the same binding site shared by eIF4G. Thus, eIF4EBP competes with eIF4G for the binding of eIF4E. As a regulation strategy, the binding affinity of eIF4EBP with eIF4E is affected by its phosphorylation status. In hypophosphorylation status, it binds strongly with eIF4E and thus inhibits the formation of eIF4F complex. SGs are subsequently formed due to translation initiation arrest. When it is phosphorylated by mTORC1, its affinity with eIF4E decreases and releases eIF4E for translation activation. Selenite induces SGs formation mainly by promoting eIF4EBP hypophosphorylation. eIF2α phosphorylation can also enhance selenite induced SGs but is not indispensable, unlike the case for arsenite-induced SGs. Meanwhile, malonate and H2O2 also induce SG forming via eIF4E-BP hypophosphorylation.53,60 Disassembly and persistence of SGs

SGs are transient structures whose formation is reversible. The dynamics of SGs disassembly is poorly understood, although polysomes and autophagy are known to be involved. Consistently, anti-cancer chemicals that regulate polysomes and autophagy are reported to affect the clearance and persistence of SGs.

Polysomes are complex consist of multiple ribosomes attaching to one mRNA chain for translating. Studies have shown that under stress conditions the majority of mRNA would disassociate from polysomes as a result of overall translation suppression.61 It is hypothesized that those depleted mRNA depleted would get sequestered into SGs. Indeed, research showed that arsenite induced SG formation got strongly inhibited and its disassembly was accelerated by stabilizing polysomes with well-established polysome stabilizing agents, cycloheximide or emetine, which indicated the existence of exchanging between SGs and polysomes. Moreover, as heat-induced SGs forming is inhibited by cycloheximide, cell apoptosis rate caused by heat shock would greatly increase. They both stabilize polysome by binding to ribosomal subunits and suppress translation elongation.62 The SGs inhibition effect makes them potential therapy for cancer or cancer chemoresistance. However, cycloheximide cannot be applied in the clinic due to its side effects including DNA damage and teratogenesis. Autophagy is another cellular response to various stresses responsible for the degradation of various cellular constituents.63 Moreover, SGs were found to be cleared by autophagy,64-65 and autophagy promoter agent rapamycin can accelerate the disassembly of SGs while autophagy inhibitor wortmannin attenuated the disassembly of SGs. Rapamycin, as the suppressor of mTOR pathway, has shown therapeutic potential for breast cancer,66 and its influence on SGs dynamics should be considered in the further study of pharmacodynamics. Selenite as a SGs inducing agent can also induce autophagy,67 however the mutual relations between autophagy and SGs under selenite were not revealed yet.

Regarding the persistence of SGs, studies have indicated that mTOR pathway might be intimidatedly involved although the underlying mechanisms remain to be addressed. The manipulation of both kinases in the mTOR pathway, DYRK3 and S6K2, has been reported to promote the persistence of SGs. While inhibition of DYRK3 kinase by a small compound GSK-626616 can promote the persistence of arsenite induced SGs,68 overexpression of S6K2 can prolong the presence of SGs.59 When there is almost no SGs retained in the control group 2 h after recovery, SGs retained more than 60% in S6K2 overexpressing cells. A recent study has identified the role of raloxifene in preventing the dissolution of hypoxia induced SGs possibly by inhibiting the re-activation of mTOR pathway.70 Conversely, a small molecule called ISRIB has been shown to induce rapid disassembly of formed SGs and inhibit eIF2α phosphorylation dependent SGs formation, making it a potential drug for the treatment of chemoresistance through SGs.71 Radiation therapy and SGs

Radiation therapy was used in almost every stage of breast cancer to reduce the recurring risk. It works by generating DNA damage with X-ray and causing cell apoptosis. Even though radiation itself can not directly induce SGs, research found that SGs can help induce radiation resistance of cancer cells.72 Cancers are commonly in hypoxia status which is an
SGs inducing stressor. However, shortly after the application of radiation, the cancer tissue would be reoxygenated and gradually recover to its original hypoxia status later. In the reoxygenation process, SGs are destabilized due to the elimination of hypoxia stress and the sequestered mRNAs get released. Among them are many hypoxia-inducible factor (HIF)-regulated transcripts and the release of them makes the HIF1 pathway signaling enhanced. It is known that activated HIF1 pathway would elevate the expression of VEGF and bFGF in vascular endothelial cells, promote their survival and thus cause radiation resistance.73

Protective role of SGs in cancer cells

Indeed, numerous researches have provided evidence for the anti-apoptosis and drug resistance inducing effect of SGs. A recent study has directly proven that by adding SGs inhibiting drug compounds EPS, the hypoxia-induced drug resistance of HeLa cells can be relieved.54 Moreover, as SGs have been detected in human cancer tissues, more attention should be paid to the potential role of SGs as a new cancer biomarker and a treatment target. Here, we aimed to summarize the drugs, proteins and pathways that induce SGs as well as the effect of SGs on cancer cells.

SGs formation triggers overall translation suppression as a protective strategy of cells. In cancer cells, it is known that the formation of SGs can protect cells by inhibiting apoptosis and promoting drug resistance. The related mechanisms have been summarized below.

Anti-apoptosis effect of SGs

The anti-apoptosis effect of SGs has been observed in many studies. Zou et al showed that arsenite treated intestinal epithelium cells with SGs forming are more resistant to apoptosis when exposed to TNF-α/CHX than non-arsenite-treated cells.74 In the treatment of bortezomib, non-SGs treated cells are treated with arsenite or in hypoxia, the formed SG essential for stress induced pathway activation. When the cells are treated with arsenite or in hypoxia, the formed SG will effectively chelate RACK1 protein instead of MTK1, resulting in the inability of MTK1 and SAPK/JNK pathways to activate and preventing the subsequent pro-apoptosis effect.81 Another protein essential for the stress induced JNK pathway activation is ROCK1. It is responsible for the phosphorylation of JIP-3 which is necessary for the activation of JNK pathway. Under heat shock stress, the activated form of ROCK1 is recruited into SGs while the inactive form remains free in cytoplasm, making the JNK pathway inactivated and apoptosis inhibited.82 It is well known that ROS is an apoptosis inducer.83,84 However, the level of ROS is found to be reduced in cells when G3BP1 is overexpressed and SGs are formed. This effect is mediated by a G3BP1 interacting protein USP10 which has antioxidant activity. In a steady-state, G3BP1 inhibits the antioxidant activity of USP10. When SGs are formed, they are both recruited to SGs and this inhibition effect was inactivated, subsequently, the unrecruited free USP10 then decreases the ROS level and apoptosis is prevented. When USP10 is knockout, cells would have increased arsenite-induced apoptosis rate which can be rescued by adding ROS scavenger, revealing the close relationship between SGs-regulated ROS level and apoptosis.85

Drug resistance induced by SGs

Besides anti-apoptosis, SGs can also protect cancer cells by inducing drug-resistance. It is known that cancer cells are commonly in a hypoxia status due to fast growth and insufficient blood supply. Meanwhile cells adapt to hypoxia through making metabolic changes which subsequently cause drug resistance.86 Shikshya et al reported that, when the hypoxia-induced SGs gets inhibited by EPS compounds, drug sensitivity of HeLa cells can be recovered to normoxia.54 Moreover, drug resistance can also be restored through overexpressing G3BP1 and new SGs formation indicating the direct relations between drug resistance and SGs.54

Glass et al showed that repeated sodium arsenite exposure would cause the development of cells resistant to SGs-inducing chemotherapeutic agents including arsenite itself and diclofenac sodium, although the resistance would disappear when arsenite is removed for 10 cell passage time.87 This resistance phenomenon is probably mediated by the altered cytokine secretion (the elevation of serpin E1 and decrease of MCP1 by resistant cells) and functions in a paracrine way. More works are needed to elucidate the mechanism behind.87,88
Looking into the future: the potential of SGs in anticancer therapy

Translation and stress response has attracted lots of attention in the research field of anticancer therapy. Formation of SGs under stresses suppresses overall translation, protects mRNA from degradation and stimulates stress-adaptive protein synthesis. Its protective role and its potential as a novel anticancer target have been proved both in vitro and in vivo. In breast cancer research field, the related SGs inducing anticancer drugs including 5-FU, sorafenib, lapatinib, selenite and rapamycin. Further experiments to test whether strategies that combine suppressors of SGs with anticancer drugs may be effectives in preventing resistance are worth investigating.

However, since the formation of SGs is regulated in a sophisticated manner and is involved with various pathways including ER stress, ROS, ubiquitin proteasome and autophagy, how to target SGs for anticancer or antichemoresistance therapy is still a great challenge. Taken bortezomib treatment as an example, researches showed that maintaining eIF2α hyperphosphorylation can enhance the efficiency of bortezomib treatment by eradicating the survival quiescent cells. Whereas previous studies revealed that hyperphosphorylation of eIF2α can induce the formation of SGs and contribute to cell survival. Thus, the balance or cross-talk between SGs and ER stress should further be elucidated to have a better understanding of the mechanism.

Meanwhile, the reported components and functions of SGs are still expanding, possibly revealing more potential treatment targets. A recently conducted comprehensive transcriptome analysis of SGs identified 215 SGs enriched ncRNA compared to cytoplasmic level. Also, Anthony et al reported that the small RNA binding protein argonaute-2 which plays an essential role in RNA silencing process is recruited into SGs in a miRNA-dependent way, indicating the existence of correlation and possible regulation pathway between SGs and ncRNA. A recent study showed dynamics of stress granule are also affected by circadian clock via oscillating eIF2α expression. More studies, especially the in vivo studies involving SGs and systematic studies including deep transcriptomic and proteomic data is need for better understanding.

Last but not the least, the identified SGs inducing anticancer drugs are just a small part of all the currently available anti-cancer drugs which may be due to the lack of research on large-scale screening of drugs and molecules that affect SG. Though, a recent drug screening study has identified the effect of raloxifene on the persistence of SGs in a pool of 1120 drugs. More similar efforts should be paid to achieve a more comprehensive understanding of SGs.

Conflict of interests

Authors declare no conflict of interests.
Stress granule in breast cancer

Funding

This work was supported by the National Natural Science Foundation of China (No. 82002979 and 81702839), and the Scientific Research and Development Funds of Peking University People’s Hospital, China (No. R DY2020-16).

References

1. Collier NC, Heuser J, Levy MA, Schlesinger MJ. Ultrastructural and biochemical analysis of the stress granule in chicken embry fibroblasts. J Cell Biol. 1988;106(4):1131–1139.
2. Jevto I, Zacharogianni M, van Oorschot MM, et al. TORC2 mediates the heat stress response in Drosophila by promoting the formation of stress granules. J Cell Sci. 2015;128(14):2497–2508.
3. Buchan JR, Muhlrad D, Parker R. P bodies promote stress granule assembly in Saccharomyces cerevisiae. J Cell Biol. 2008;183(3):441–455.
4. Grabocka E, Bar-Sagi D. Mutant KRAS enhances tumor cell fitness by upregulating stress granules. Cell. 2016;167(7):1803–1813.
5. Somasekharan SP, El-Naggar A, Leprivier G, et al. YB-1 regulates stalled translation initiation complexes. Cell. 2008;138(3):547–558.
6. Kedersha NL, Gupta M, Li W, Miller I, Anderson P. RNA-binding protein-1 (eIF4E-GTP-tRNA(i)(Met))-deficient preinitiation complexes are core constituents of mammalian stress granules. J Cell Biol. 1999;147(7):1431–1442.
7. Kimball SR, Horetsky RL, Ron D, Jefferson LS, Harding HP. Mammalian stress granules represent sites of accumulation of stalled translation initiation complexes. Am J Physiol Cell Physiol. 2003;284(2):C273–C284.
8. Kedersha N, Chen S, Gilks N, et al. Evidence that ternary complex (eIF2-GTP-tRNA(i)(Met))-deficient preinitiation complexes are core constituents of mammalian stress granules. Mol Cell Biol. 2002;13(1):195–210.
9. Makas S, Mills JR, Garreau C, et al. Uncoupling stress granule assembly and translation initiation inhibition. Mol Biol Cell. 2009;20(11):2673–2683.
10. Tourni`ere H, Chebli K, Zekri L, et al. The RasGAP-associated endoribonuclease G3BP assembles stress granules. J Cell Biol. 2003;160(6):823–831.
11. Proter DSW, Barker R. Principles and properties of stress granules. Trends Cell Biol. 2016;26(9):668–679.
12. Jain S, Wheeler JR, Walters RW, Agrawal A, Barsic A, Parker R. ATPase-modulated stress granules contain a diverse proteome and substructure. Cell. 2016;164(3):487–498.
13. Martin S, Tazi J. Visualization of G3BP stress granules dynamics in live primary cells. J Vis Exp. 2014;87:51197.
14. Wall ML, Lewis SM. Methylarginines within the RGG-motif region of hnRNP A1 affect its IRES trans-acting factor Activity and are required for hnRNP A1 stress granule localization and formation. J Mol Biol. 2017;429(2):295–307.
15. Baron DM, Kaushansky LJ, Ward CL, et al. Amytropic lateral sclerosis-linked FUS/TLS alters stress granule assembly and dynamics. Mol Neurodegener. 2013;8:30.
16. Gilks N, Kedersha N, Ayodele M, et al. Stress granule assembly is mediated by prion-like aggregation of TIA-1. Mol Biol Cell. 2004;15(12):5383–5398.
17. Aguzzi A, Altmeyer M. Phase separation: linking cellular compartmentalization to disease. Trends Cell Biol. 2016;26(7):547–558.
18. Lin Y, Proter DSW, Rosen MK, Parker R. Formation and maturation of phase-separated liquid droplets by RNA-binding proteins. Mol Cell. 2015;60(2):208–219.
19. Panas MD, Ivanov P, Anderson P. Mechanistic insights into mammalian stress granule dynamics. J Cell Biol. 2016;215(3):313–323.
20. Buchan JR, Parker R. Eukaryotic stress granules: the ins and outs of translation. Mol Cell. 2009;36(6):932–941.
21. Reineke LC, Cheema SA, Dubulle J, Neilson JR. Chronic starvation induces noncanonical pro-death stress granules. J Cell Sci. 2018;131(19):jcs220444.
22. Fujimura K, Sasaki AT, Anderson P. Selenite targets eIF4E-binding protein-1 to inhibit translation initiation and induce the assembly of non-canonical stress granules. Nucleic Acids Res. 2012;40(16):8099–8110.
23. Visser LJ, Medina GN, Rabouw HH, et al. Foot-and-Mouth disease virus leader protease cleaves G3BP1 and G3BP2 and inhibits stress granule formation. J Virol. 2019;93(2):e00922-18.
24. Gordon D, Dafinca R, Scaber J, et al. Single-copy expression of an amyotrophic lateral sclerosis-linked TDP-43 mutation (M337V) in BAC transgenic mice leads to altered stress granule dynamics and progressive motor dysfunction. Neurobiol Dis. 2019;121:148–162.
25. Anderson P, Kedersha N, Ivanov P. Stress granules, P-bodies and cancer. Biochim Biophys Acta. 2015;1849(7):861–870.
26. Pelicano H, Carney D, Huang P. ROS stress in cancer cells and therapeutic implications. Drug Resist Update. 2004;7(2):97–110.
27. Conklin KA. Chemotherapy-associated oxidative stress: impact on chemotherapy effectiveness. Integr Canc Ther. 2004;3(4):294–300.
28. Moeller BJ, Richardson RA, Dewhirst MW. Hypoxia and radiotherapy: opportunities for improved outcomes in cancer treatment. Cancer Metastasis Rev. 2007;26(2):241–248.
29. Longley DB, Harkin DP, Johnston PG. 5-fluorouracil: mechanisms of action and clinical strategies. Nat Rev Cancer. 2003;3(5):330–338.
30. Kaehler C, Ilsesee J, Huc ho T, Lehrhac H, Krobitsch S. 5-Fluorouracil affects assembly of stress granules based on RNA incorporation. Nucleic Acids Res. 2014;42(10):6436–6447.
31. Wilhelm S, Carter C, Lynch M, et al. Discovery and development of sorafenib: a multitarget inhibitor for treating cancer. Nat Rev Drug Discov. 2006;5(10):835–844.
32. Escudier B, Eisen T, Stadler WM, et al. Sorafenib in advanced clear-cell renal-cell carcinoma. N Engl J Med. 2007;356(2):125–134.
33. Spinz P, Pagli S. Sorafenib in advanced hepatocellular carcinoma. N Engl J Med. 2008;359(23):2497–2498.
34. Zafarakas M, Papasozomenou P, Emmanouilides C. Sorafenib in breast cancer treatment: a systematic review and overview of clinical trials. World J Clin Oncol. 2016;7(4):331–336.
35. Adajide B, St-Sanver VG, Quevillon Huberdeau M, et al. Sorafenib, a multitarget inhibitor, induces formation of stress granules in hepatocarcinoma cells. Oncotarget. 2015;6(41):43927–43943.
36. Adajide B, Simoune B, Leducu N, et al. Treatment of cancer cells with Lapatinib negatively regulates general translation and induces stress granules formation. PLoS One. 2020;15(5):e0231894.
37. Cruickshanks N, Tang Y, Booth L, Hamed H, Grant S, Dent P. Lapatinib and obatoclax kill breast cancer cells through reactive oxygen species-dependent endoplasmic reticulum stress. Mol Pharmacol. 2012;82(6):1217–1229.
38. Rahmani M, Davis EM, Crabtree TR, et al. The kinase inhibitor sorafenib induces cell death through a process involving induction of endoplasmic reticulum stress. Mol Cell Biol. 2007;27(15):5499–5513.
39. Doan NTQ, Paulsen ES, Sehgal P, et al. Targeting thapsigargin towards tumors. Steroids. 2015;97:2–7.
41. Macor JE. Annual Reports in Medicinal Chemistry. vol. 46. San Diego, California: Academic Press; 2011. Annual reports in medicinal chemistry.

42. Hood KA, West LM, Northcote PT, Miller JH. Induction of apoptosis by the marine sponge (Mycale) metabolites, mycalamide A and pateamine. Apoptosis. 2001;6(3): 207–219.

43. Cencic R, Carrier M, Galicia-Vázquez G, et al. Antitumor activity and mechanism of action of the cyclopentabenzofuran, silvestrol. PLoS One. 2009;4(4): e5223.

44. Tsumuraya T, Ishikawa C, Machijima Y, et al. Effects of hupiristanol, an inhibitor of eIF4A, on adult T-cell leukemia. Biochem Pharmacol. 2011;81(6):713–722.

45. Martins I, Kepp O, Schlemmer F, et al. Restoration of the immunogenicity of cisplatin-induced cancer cell death by endoplasmic reticulum stress. Oncogene. 2011;30(10):1147–1158.

46. Raymond E, Chaney SG, Taamma A, Cvitkovic E. Oxaliplatin: a review of preclinical and clinical studies. Ann Oncol. 1998; 9(10):1053–1071.

47. Noordhuis P, Laan AC, van de Born K, Losekoot N, Kathmann I, van der Voort GJ. Oxaliplatin activity in selected and unselected human ovarian and colorectal cancer cell lines. Biochem Pharmacol. 2008;76(1):53–61.

48. Damtröder C, Solass W, Zieren J, Strumburg D, Giger-Pabst U, Aebersold R, Pelkmans L. Dual specificity kinase DYRK3 couples stress granule condensation/dissolution to mTORC1 signaling. Cell. 2013;152(4):791–805.

49. Moeller BJ, Cao Y, Li CY, Dewhirst MW. Radiation activates HIF-1. 2011;81(6):713–722.

50. Noordhuis P, Laan AC, van de Born K, Losekoot N, Kathmann I, van der Voort GJ. Oxaliplatin activity in selected and unselected human ovarian and colorectal cancer cell lines. Biochem Pharmacol. 2008;76(1):53–61.

51. Fournier MJ, Gareau C, Mazroui R. The Chemotherapeutic Agent Bortezomib Induces the Formation of Stress Granules. BioMed Central Ltd; 2010.

52. Kashiwagi K, Ito T, Yokoyama S. Crystal structure of elf2B and insights into elf2B-elf2B interactions. FEBS J. 2017;284(6): 868–874.

53. Emara MM, Fujimura K, Sciaranghella D, Ivanov P, Anderson P. Hydrogen peroxide induces stress granule formation independent of elf2B phosphorylation. Biochem Biophys Res Commun. 2012;423(4):763–769.

54. Timalinsa S, Arimoto-Matsuzaki K, Kitamura M, et al. Chemical compounds that suppress hypoxia-induced stress granule formation enhance cancer drug sensitivity of human cervical cancer HeLa cells. J Biochem. 2018;164(5):381–391.

55. Bordeleau ME, Mari A, Oberer M, et al. Functional characterization of IRESes by an inhibitor of the RNA helicase elf4A4. Nat Chem Biol. 2006;2(4):213–220.

56. Bordeleau ME, Robert F, Gerard B, et al. Therapeutic suppression of translation initiation modulates chemosensitivity in a mouse lymphoma model. J Clin Invest. 2008;118(7): 2651–2660.

57. Cramer Z, Sadek J, Vazquez GG, et al. elf4A inhibition prevents the onset of cytokine-induced muscle wasting by blocking the STAT3 and INOS pathways. Sci Rep. 2018;8(1): 8414.

58. Mazroui R, Sukarieh R, Bordeleau ME, et al. Inhibition of ribosome recruitment induces stress granule formation independently of eukaryotic initiation factor 2A phosphorylation. Mol Biol Cell. 2006;17(10):4212–4219.

59. Kim WJ, Kim JH, Jang SK. Anti-inflammatory lipid mediator 15d-PGJ2 inhibits translation through inactivation of elf4A. EMBO J. 2007;26(24):5020–5032.

60. Fu X, Gao X, Ge L, et al. Malonate induces the assembly of cytoplasmic stress granules. FEBS Lett. 2016;590(1):22–33.

61. Thomas JD, Johannes GJ. Identification of mRNAs that continue to associate with polysomes during hypoxia. RNA. 2007;13(7): 1116–1131.

62. Baglioni C. Inhibition of protein synthesis in reticulocytes by antibiotics. 3. Mechanism of action of sparsomycin. Biochim Biophys Acta. 1966;129(3):642–645.

63. Klionsky DJ, Emr SD. Autophagy as a regulated pathway of cellular degradation. Science. 2000;290(5497):1717–1721.

64. Seguin SJ, Morelli FF, Vinet J, et al. Inhibition of autophagy, lysosome and VCP function impairs stress granule assembly. Cell Death Differ. 2014;21(12):1838–1851.

65. Buscani MR, Taylor JP, Parker R. Eukaryotic stress granules are cleared by autophagy and Cdc48/VCP function. Cell. 2013;153(7):1461–1474.

66. Benjamin D, Colombi M, Moroni C, Hall MN. Rapamycin passes the torch: a new generation of mTOR inhibitors. Nat Rev Drug Discov. 2011;10(11):868–880.

67. Kim EH, Sohn S, Kwon HJ, et al. Sodium selenite induces superoxide-mediated mitochondrial damage and subsequent autophagic cell death in malignant glioma cells. Cancer Res. 2007;67(13):6314–6324.

68. Wippich F, Bodenmüller B, Trajkovska MG, Wanka S, Andersson R, Peikema L. Dual specificity kinase DYRK3 couples stress granule condensation/dissolution to mTORC1 signaling. Cell. 2013;152(4):791–805.

69. Sfakianos AP, Mellor LE, Pang YF, et al. The mTOR-S6 kinase pathway promotes stress granule assembly. Cell Death Differ. 2018;25(10):1766–1780.

70. Attwood KM, Robichaud A, Westhaver LP, et al. Raploxfene prevents stress granule dissolution, impairs translational control and promotes cell death during hypoxia in glioblastoma cells. Cell Death Dis. 2020;11(11):989.

71. Sidrauskis C, McGeachy AM, Ingolia NT, Walter P. The small molecule ISRIB reverses the effects of elf2 phosphorylation on translation and stress granule assembly. Elife. 2015;4:e05033.

72. Moeller BJ, Cao Y, Li CY, Dewhirst MW. Radiation activates HIF-1 to regulate vascular radiosensitivity in tumors: role of reoxygenation, free radicals, and stress granules. Cancer Cell. 2004;5(5):429–441.

73. Semenza GL. Intratumoral hypoxia, radiation resistance, and HIF-1. Cancer Cell. 2004;5(5):405–406.

74. Zou T, Rao JN, Liu L, et al. Polyamines inhibit the assembly of stress granules in normal intestinal epithelial cells regulating apoptosis. Am J Physiol Cell Physiol. 2012;303(1):C102–C111.

75. Shah OJ, Wang Z, Hunter T. Inappropriate activation of the TSC/Rheb/mTOR/S6K cassette induces IRS1/2 depletion, insulin resistance, and cell survival deficiencies. Curr Biol. 2004;14(18): 1650–1656.

76. Takahara T, Maeda T. Transient sequestration of TORC1 into stress granules during heat stress. Mol Cell. 2012;47(2): 242–252.

77. Thedieck K, Holzwarth B, Pretzell MT, et al. Inhibition of mTORC1 by astrin and stress granules prevents apoptosis in cancer cells. Cell. 2013;154(4):859–874.

78. Zanke BW, Boudreau K, Rubie E, et al. The stress-activated protein kinase pathway mediates cell death following injury induced by cis-platinum, UV irradiation or heat. Curr Biol. 1996;6(5):606–613.

79. Davis RJ. Signal transduction by the JNK group of MAP kinases. Cell. 2000;103(2):239–252.

80. Sui X, Kong N, Ye L, et al. p38 and JNK MAPK pathways control the balance of apoptosis and autophagy in response to chemotherapeutic agents. Cancer Lett. 2014;344(2):174–179.

81. Arimoto K, Fukuda H, Imajoh-Ohmi S, Saltare H, Takekawa M. Formation of stress granules inhibits apoptosis by suppressing...
stress-responsive MAPK pathways. Nat Cell Biol. 2008;10(11):1324–1332.

82. Tsai NP, Wei LN. RhoA/ROCK1 signaling regulates stress granule formation and apoptosis. Cell Signal. 2010;22(4):668–675.

83. Simon HU, Haj-Yehia A, Levi-Schaffer F. Role of reactive oxygen species (ROS) in apoptosis induction. Apoptosis. 2000;5(5):415–418.

84. Khong A, Matheny T, Jain S, Mitchell SF, Wheeler JR, Parker R. The stress granule transcriptome reveals principles of mRNA accumulation in stress granules. Mol Cell. 2017;68(4):808–820.

85. Takahashi M, Higuchi M, Matsuki H, et al. Stress granules inhibit apoptosis by reducing reactive oxygen species production. Mol Cell Biol. 2013;33(4):815–829.

86. Rohwer N, Cramer T. Hypoxia-mediated drug resistance: novel insights on the functional interaction of HIFs and cell death pathways. Drug Resist Updat. 2011;14(3):191–201.

87. Glass L, Wente SR. Gle1 mediates stress granule-dependent survival during chemotoxic stress. Adv Biol Regul. 2019;71:156–171.

88. Aditi, Folkmann AW, Wente SR. Cytoplasmic hGle1A regulates stress granules by modulation of translation. Mol Biol Cell. 2015;26(8):1476–1490.

89. El-Naggar AM, Sorensen PH. Translational control of aberrant stress responses as a hallmark of cancer. J Pathol. 2018;244(5):650–666.

90. Schewe DM, Aguirre-Ghiso JA. Inhibition of elf2alpha dephosphorylation maximizes bortezomib efficiency and eliminates quiescent multiple myeloma cells surviving proteasome inhibitor therapy. Cancer Res. 2009;69(4):1545–1552.

91. Leung AKL, Calabrese JM, Sharp PA. Quantitative analysis of Argonaute protein reveals microRNA-dependent localization to stress granules. Proc Natl Acad Sci U S A. 2006;103(48):18125–18130.

92. Pare JM, Tahbaz N, López-Orozco J, LaPointe P, Lasko P, Hobman TC. Hsp90 regulates the function of argonaute 2 and its recruitment to stress granules and P-bodies. Mol Biol Cell. 2009;20(14):3273–3284.

93. Wang R, Jiang X, Bao P, Qin M, Xu J. Circadian control of stress granules by oscillating elf2a. Cell Death Dis. 2019;10(3):215.