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## REVIEW

# From Birth to Death: The Complex Lives of Eukaryotic mRNAs

Melissa J. Moore

Recent work indicates that the posttranscriptional control of eukaryotic gene expression is much more elaborate and extensive than previously thought, with essentially every step of messenger RNA (mRNA) metabolism being subject to regulation in an mRNA-specific manner. Thus, a comprehensive understanding of eukaryotic gene expression requires an appreciation for how the lives of mRNAs are influenced by a wide array of diverse regulatory mechanisms.

Many written accounts of eukaryotic gene expression might start something like this: "Messenger RNAs (mRNAs) are the central conduits in the flow of information from DNA to protein. In eukaryotes, mRNAs are first synthesized in the nucleus as pre-mRNAs that are subject to 5'-end capping, splicing, 3'-end cleavage, and polyadenylation. Once pre-mRNA processing is complete, mature mRNAs are exported to the cytoplasm, where they serve as the blueprints for protein synthesis by ribosomes and then are degraded." Like a short obituary, however, this dry and simplistic description captures nothing of the intricacies, intrigues, and vicissitudes defining the life history of even the most mundane mRNA. In

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addition, of course, some mRNAs lead lives that, if not quite meriting an unauthorized biography, certainly have enough twists and turns to warrant a more detailed nucleic acid interest story. It is these intricacies, and our recent progress in understanding them, that are the subject of this review. We will follow the lives of eukaryotic mRNAs from the point at which they are birthed from the nucleus until they are done in by gangs of exonucleases lying in wait in dark recesses of the cytoplasm. Along the way, mRNAs may be shuffled to and from or anchored at specific subcellular locations, be temporarily withheld from the translation apparatus, have their 3' ends trimmed and extended, fraternize with like-minded mRNAs encoding proteins of related function, and be scrutinized by the quality-control police. Although some of these processes were originally thought to affect only select mRNA popula-

tions or be largely limited to highly specialized cell types like germ cells and neurons, recent work suggests that the majority of mRNAs in multiple cell types are subject to a diverse array of regulatory activities affecting essentially every aspect of their lives.

## The mRNP as a Posttranscriptional Operon

Throughout their lifetimes, mRNAs are escorted by a host of associated factors, some of which remain stably bound while others are subject to dynamic exchange (Table 1). Together with mRNA, this complement of proteins and small noncoding RNAs [e.g., microRNAs (miRNAs)] constitute the messenger ribonucleoprotein particle (mRNP). It is the unique combination of factors accompanying any particular mRNA, as well as their relative positions along the transcript, that dictates almost everything that happens to that mRNA in the cytoplasm. In budding yeast, it is estimated that ~570 different proteins have the capacity to bind RNA (1). This number is no doubt considerably larger in humans, because a single type of RNA binding domain, the RNA recognition motif (RRM), is represented in

**Table 1.** mRNP cheat sheet.

CBC20/80	The nuclear cap binding complex. A heterodimer of 20 and 80 kD subunits. Joins the mRNP coincident with cap formation during transcription and facilitates pre-mRNA splicing. In the cytoplasm, can serve as a translation initiation factor through interactions with eIF4G but is ultimately replaced by eIF4E.
eIF4E	Eukaryotic translation initiation factor 4E. The major cytoplasmic cap binding protein. Target of many translational regulators [eIF4E binding proteins (4E-BPs)] that disrupt its interaction with eIF4G.
eIF4G	Eukaryotic translation initiation factor 4G. A large scaffolding protein that can simultaneously interact with cap binding proteins, PABPCs, and eIF3 bound to the small ribosomal subunit.
PABPN1	The nuclear poly(A) binding protein. Binds poly(A) by a single RNA recognition motif (RRM) and an arginine-rich C-terminal domain. In budding yeast, the evolutionarily unrelated Nab2 protein serves this role.
PABPCs	Cytoplasmic poly(A) binding proteins. Single-celled eukaryotes contain a single PABPC, whereas human cells contain four. All PABPCs bind poly(A) RNA through four RRM.
HnRNP proteins	A diverse set of factors loosely defined as all proteins associating with heterogeneous nuclear RNA (hnRNA, made up of pre-mRNA and nuclear mRNA) that are not stable components of other RNP complexes, such as small nuclear RNPs (snRNPs). Some hnRNP proteins accompany mRNAs to the cytoplasm; others are confined to the nucleus.
EJC	The exon junction complex. A set of proteins loaded onto mRNAs upstream of exon-exon junctions as a consequence of pre-mRNA splicing and which accompanies the spliced mRNA to the cytoplasm.
SR proteins	A family of structurally related, nuclear RNA binding proteins containing an RRM and a domain rich in serines and arginines (RS domain). The serines in the RS domain serve as sites of dynamic phosphorylation. Some SR proteins accompany mRNAs to the cytoplasm; others are confined to the nucleus. Many SR proteins play key roles in pre-mRNA splicing.
Y-box proteins	A family of multifunctional nucleic acid binding proteins containing a "cold-shock" domain. Along with PABPCs, Y-box proteins constitute the major mRNP structural components in somatic cells. They are thought to bind along the body of the message and have a packaging role that modulates translational activity. In <i>Xenopus</i> oocytes, Y-box proteins FRGY2 and mRNP3 are major components of stored mRNPs.
TIA-1/TIAR	Structurally related RNA binding proteins consisting of three RRM and a C-terminal prionlike domain. The prionlike domain is thought to self-oligomerize in vivo and drive the formation of stress granules.
miRNAs	MicroRNAs. Small noncoding RNAs that imperfectly base-pair with recognition sites in 3' UTRs. In combination with RISC (RNA-induced silencing complex), miRNAs negatively regulate protein synthesis by the cognate mRNA.

nearly 500 different human genes (2). Other common RNA binding motifs include the KH domain, the double-stranded RNA binding domain (dsRBD), zinc fingers, RGG boxes, and the Pumilio homology domain found in PUF proteins (3, 4). The human genome has also been estimated to encode more than 400 different miRNAs targeting transcripts from ~5,000 different genes, or ~20% of the genome (5–7).

A few mRNP components target the two elements common to almost every message: the 7-methylguanosine cap found at the 5' end of all RNA polymerase II transcripts and the poly(A) tail comprising most mRNA 3' ends (8, 9). Others, such as the abundant mRNA-packaging Y-box proteins, appear to associate along the length of transcripts in a largely sequence-independent manner (10). Yet another set, exemplified by the exon junction complex (EJC), is loaded at specific positions independent of sequence (11). The majority of mRNA binding factors, however, target particular structures or sequences present in some mRNAs but not others. Such specific recognition elements most commonly occur in the untranslated regions (UTRs) at the 5' and 3' ends of the message.

Individual mRNP components can be thought of as adaptors that allow mRNAs to interface with the numerous intracellular machineries mediating their subcellular localization, translation, and decay, as well as the various signal transduction systems. Some adaptors make positive interactions and thereby serve as activators of a particular process, whereas others disrupt the positive interactions and act as repressors. By containing binding

sites for diverse adaptors, individual mRNAs can respond to myriad inputs, allowing their expression to be exquisitely fine-tuned to changing conditions. These changing conditions can also alter the levels and RNA binding properties of the adaptors, transforming the subpopulations of mRNAs to which they bind. The result is an elaborate web of regulatory networks of equal, if not greater, complexity to those controlling initial mRNA synthesis (12, 13). Indeed, eukaryotic mRNPs have been likened to "posttranscriptional operons" that serve to markedly expand the regulatory plasticity of our unexpectedly small genomes (12). The importance of such posttranscriptional regulatory mechanisms in the control of eukaryotic gene expression is highlighted by the wide variability in the degree to which mRNA and protein abundances correlate in vivo (14, 15). Thus, changes in mRNA levels, as measured by microarrays, for example, cannot be presumed to reflect proportionate changes in protein abundance or activity.

A key assertion of the posttranscriptional operon model is that mRNAs encoding functionally related proteins should be coordinately regulated by specific mRNP components recognizing sequence elements common to that set of mRNAs (12). Evidence that this may be the case on a genome-wide scale was recently provided by a study identifying the complement of mRNAs bound to each of the five individual Puf proteins in *Saccharomyces cerevisiae* (16). The Puf proteins are a family of structurally related cytoplasmic mRNP proteins that have been implicated in the control of mRNA translation and stability through binding sites in the 3' UTR.

In all, 12% of known or predicted yeast mRNAs were found to stably associate with one or more of these proteins, although the vast majority (645 out of 735) bound only one. Notably, each Puf protein exhibited a highly skewed distribution of bound mRNAs: Puf1p and Puf2p bound mostly mRNAs encoding membrane-associated proteins, Puf3p almost exclusively targeted messages for nuclear-encoded mitochondrial proteins, and Puf4p and Puf5p associated primarily with transcripts encoding proteins bound for the nucleus. In several cases, a majority of the subunits comprising a particular multiprotein machine, such as the mitochondrial ribosome and a number of nuclear chromatin modification complexes, were encoded by mRNAs "tagged" by a single Puf protein. Together with earlier data (12), these new results (16) strongly support the idea that the expression of proteins with common functional themes or subcellular distributions is coordinated by large-scale regulatory networks operating at the mRNP level.

### Nuclear mRNP Embryology and Export

Many components of the cytoplasmic mRNP are first recruited in the nucleus, coincident with transcription and pre-mRNA processing. Such factors include the nucleocytoplasmic shuttling hnRNP (heterogeneous nuclear RNP) and SR (serine/arginine rich) proteins as well as the EJC (11, 17, 18) (Table 1). Both hnRNP and SR proteins recognize short consensus sequences through their RNA binding domains (17); the SR proteins additionally contain a domain rich in Arg-Ser dipeptides that can variously interact with proteins or RNA and is subject to dynamic phosphorylation (18). The

EJC is a set of proteins deposited onto spliced mRNAs by the process of pre-mRNA splicing. Unlike other known mRNP components, the EJC is loaded in a position-dependent, but sequence-independent, manner. EJC deposition sites are about 20 to 25 nucleotides upstream of exon-exon junctions, the sites where introns resided in the pre-mRNA. Intriguingly, the downstream consequences of EJC deposition are highly dependent on EJC location along the mRNA—EJCs inside the open reading frame (ORF) can positively influence translation, whereas EJCs in the 3' UTR can target the bound mRNA for rapid destruction via nonsense-mediated mRNA decay (NMD) (11). It was recently found that certain SR proteins can recapitulate both of these effects, although the extent to which this is dependent on SR protein position along the mRNA remains to be elucidated (18).

A key issue regarding mRNP composition is how the complement of bound factors evolves as an mRNA proceeds through the various stages of its life. The first major change in mRNP composition occurs as mRNAs are birthed from the nucleus through the nuclear pore complex (NPC) (Fig. 1). The NPC is a mammoth, eight-fold symmetric supramolecular assembly (50 to 125 MD) that serves as the molecular gatekeeper for movement of proteins and protein-RNA complexes between the nucleus and cytoplasm (19). Some nuclear-acquired mRNP proteins, such as the mRNA export adaptors and receptors responsible for targeting the nuclear mRNP to the NPC, are shed as a consequence of the birthing process. In general, export adaptors are mRNA binding proteins that serve to bridge the mRNA to one or more receptor proteins, which in turn contact components of the NPC. Like other nuclear-

acquired mRNP proteins, these adaptors and receptors are recruited cotranscriptionally, but they dissociate from the mRNA either as it is transiting the pore or soon after reaching the cytoplasm (20). In the case of the yeast adaptor and SR-like protein Npl3p, this dissociation is triggered by its cytoplasmic phosphorylation, which serves to destabilize its interaction with both the mRNA and the export receptor NXF1/Mex67p. Reimport and nuclear dephosphorylation of Npl3p creates a regulated RNA binding-and-release cycle capable of imparting overall directionality to the mRNA export process (18, 21).

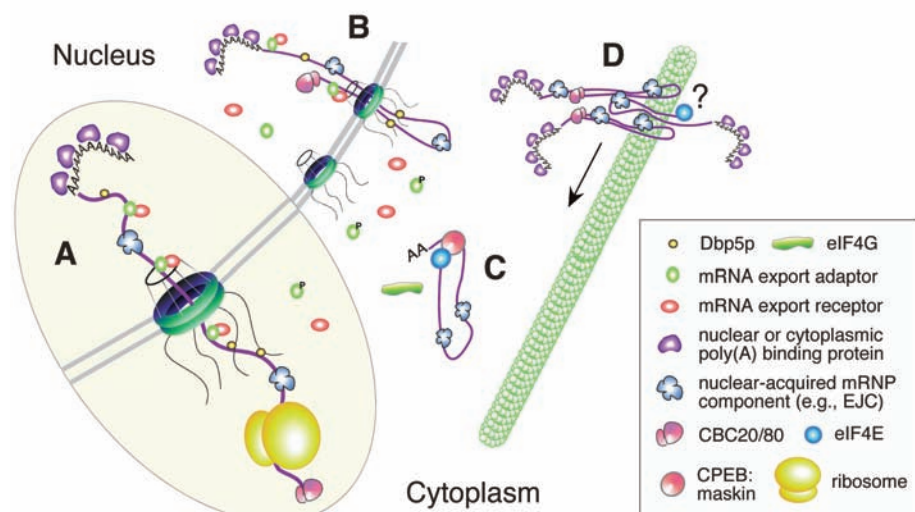
Other nuclear-restricted mRNP components might be removed by DEXH/D-box proteins, a family of RNA binding nucleotide triphosphatases, some of which can remove secondary structures and/or bound proteins from RNA (22). One such protein is the essential mRNA export factor Dbp5p, which is recruited to mRNPs both cotranscriptionally and as they transit the pore (23). It has been suggested that Dbp5p assists in "remodeling" the mRNP during nuclear export, possibly by facilitating binding of new cytoplasmic mRNP factors as it bumps off other proteins that return to the nucleus. If this is the case, however, it is unclear how Dbp5p would be prevented from indiscriminately removing the many nuclear-acquired proteins known to remain associated with the cytoplasmic mRNP. An alternate role for Dbp5p is suggested by its strong interactions with the long fibrils extending away from the cytoplasmic face of the NPC. By simultaneously binding the mRNA and these fibrils, Dbp5p might instead serve to prevent the mRNP from backsliding into the nucleus as it exits the pore and thereby contribute to export directionality.

To date, the only mRNPs that have been caught in the act of transiting the pore are the

gigantic Balbiani ring mRNPs (24). Balbiani ring mRNAs 1 and 2 of the dipteran *Chironomus tentans* are each >30,000 nucleotides long. This immense size, coupled with their extremely high expression levels in larval salivary glands, has enabled direct electron microscopic visualization of Balbiani mRNP docking and translocation through the NPC. In the nucleoplasm, Balbiani ring mRNPs exist as tightly packed ringlike structures. Upon docking with the NPC, these ring structures partially unfold, allowing the mRNA to enter the pore 5' end first. As soon as their 5' ends begin to protrude into the cytoplasm, Balbiani ring mRNAs are engaged by the translation machinery, with multiple ribosomes often visible attached to mRNAs still transiting the pore. It should be noted, however, that this one-at-a-time, 5'-end-first birthing order of Balbiani ring mRNPs does not necessitate that this is how all mRNPs emerge from the nucleus; lesser mRNPs could well be born as multiples or even in a breach position. Many mRNAs destined for particular subcellular locations appear to travel in multi-mRNA packets or particles. Currently it is unknown whether these particles first form in the cytoplasm after mRNP export, or whether they are initially assembled in the nucleus and are then exported to the cytoplasm en masse. Other data support the idea that mRNAs might not always emerge 5' end first. For example, neither the 7-methylguanosine 5' cap structure nor the nuclear 5' cap binding complex (CBC20/80) is essential for mRNA export in budding yeast, and injection of large amounts of cap analog only minimally affected mRNA export in *Xenopus* oocytes (25). Further, mRNA export adaptors are apparently recruited along the length of nascent transcripts rather than being concentrated near 5' ends (26). Finally, consistent with a crucial role for the poly(A) tail in mRNA export, the nuclear poly(A) tail-binding proteins in both metazoans and budding yeast have known interactions with export receptors and NPC components (9). Indeed, a provocative possibility is that simply because of their gigantic size and their need to be efficiently recruited to the endoplasmic reticulum (which constitutes the cytoplasmic face of the nuclear envelope and into which proteins bound for secretion are extruded), Balbiani ring mRNPs may have evolved specific mechanisms ensuring 5'-end-first delivery that are not employed by the bulk of cellular mRNPs.

### From Birth to Baptism: Engaging the Translation Apparatus

Although CBC20/80 is not essential for mRNA export, it can serve as an initiation factor for protein synthesis. Like the Balbiani ring mRNAs, many mRNAs enter the translationally active pool immediately upon export to the cytoplasm. At this stage, the 5' cap is still largely bound by the nuclear CBC20/80 com-



**Fig. 1.** Schematic of mRNA export and alternate mRNA fates in the cytoplasm. Export through the NPC requires export adaptors and receptors as well as Dbp5p. Some mRNAs are exported 5' end first and are immediately engaged by ribosomes (A), whereas others may be exported by a non-5'-end-first mechanism (B). Once in the cytoplasm, some mRNPs are stored in a translationally silent state (C), and others are transported to specific subcellular locations along the cytoskeleton (D).

plex, whereas the poly(A) tail carries a mixture of nuclear and cytoplasmic poly(A) binding proteins PABPN1 and PABPCs (Table 1). In this newly exported mRNP, CBC20/80 can functionally interact with translation initiation factor 4G (eIF4G), which serves to recruit the small ribosomal subunit and initiate 5'→3' scanning along the 5' UTR for an AUG start codon (27). Once the start codon is identified, the large ribosomal subunit is engaged to form an 80S complex competent for protein synthesis.

Another major change in mRNP composition necessarily occurs upon the first passage of the 80S ribosome along the mRNA—the so-called “pioneering round” of translation (28). Threading of the mRNA through the narrow space between the two ribosomal subunits strips away any remaining nuclear-acquired mRNP proteins, such as EJCs, residing inside the ORF. At some point, CBC20/80 and PABPN1 are also replaced by eIF4E (the major cytoplasmic cap-binding protein) and PABPCs, respectively. Whether these exchanges require any special mechanisms, such as the phosphorylation that promotes dissociation of Npl3p from newly exported mRNPs, or whether they occur simply as a consequence of mass action, is unknown. Regarding the second possibility, the low cytoplasmic concentrations of CBC20/80 and PABPN1 coupled with the high concentrations of eIF4E and PABPCs could naturally lead to the latter set replacing the former, given reasonable dissociation rates. In any event, once the transition is complete, a network of simultaneous interactions between the 5' cap, eIF4E, eIF4G, PABPCs, and the poly(A) tail results in functional circularization of the message (Fig. 2), an arrangement thought to facilitate translational control by regulatory elements in the 3' UTR, promote efficient ribosome reinitiation during active translation, and protect both ends of the transcript from the mRNA degradation machinery (9).

Upon export, not all mRNAs immediately enter the translationally active pool. Many are held instead in a translationally quiescent state awaiting either proper subcellular localization or some signal that the timing is now right to make protein. In early metazoan embryos, for example, no new transcription occurs until after several cell divisions. Therefore, the oocyte must accumulate and store all the mRNAs required for early development. In immature frog oocytes, a number of these maternal mRNAs are translationally silenced through a mechanism involving substantial shortening of their poly(A) tails from their initial nuclear length of 200 to 250 adenosines to a mere 20 to 40 bases. This shortening is modulated by CPEB, a protein that recognizes the so-called cytoplasmic polyadenylation element (CPE) in the 3' UTR. CPEB also interacts with Maskin, a protein that competes with eIF4G for binding to eIF4E. In the context of a short poly(A) tail,

which cannot effectively recruit PABPCs or eIF4G, the Maskin-eIF4E interaction inhibits translation. When the oocytes are induced to complete meiosis, CPEB becomes phosphorylated; in this phosphorylated form, CPEB stimulates readdition of the poly(A) tail by cytoplasmic poly(A) polymerases. The longer poly(A) tails rebind PABPCs, which in turn recruit eIF4G to initiate translation (29).

The CPEB-Maskin-eIF4E interaction is just one example of translational regulation by so-called “4E inhibitory proteins,” which target the eIF4E-eIF4G interface. Some 4E inhibitory proteins like Maskin are tethered to a cis element in the 3' UTR and therefore act only on mRNAs containing that element. Another class, the “4E binding proteins” (4E-BPs), are not tethered and therefore act more globally by sequestering any available eIF4E; this results in preferential translational inhibition of mRNAs that normally require high eIF4E levels. In addition to the control of development and cell growth, variants of this general translational regulatory scheme have been implicated in tumor suppression as well as the control of localized protein synthesis at neuronal synapses, which is believed to be essential for long-term potentiation (LTP) and memory consolidation (29).

A currently open question about translationally quiescent mRNPs has been whether they undergo a “pioneering round” of translation driven by CBC20/80 before entering their translationally silent phase. At least for one mRNA, this appears not to be the case. Proper localization and regulated translation of oskar mRNA at the posterior pole of *Drosophila* oocytes is essential for germline and abdomen formation in the future embryo. During transport from its sites of production in nurse cells to the posterior pole of the oocyte, oskar mRNA is translationally silenced by a 3'-UTR-tethered 4E inhibitory protein, Cup (29). In addition to sequences in the 3' UTR, oskar mRNA localization requires deposition of an EJC within the ORF, and the bound EJC proteins accumulate along with oskar mRNA at the posterior pole (30). If oskar mRNA were subject to a pioneering round of translation before translational silencing and transport, then the EJC would be expected to be removed in the nurse cells and be unable to participate in mRNP localization or colocalize with oskar mRNA at the posterior pole. Further, the observation that translational silencing of oskar during transport involves a 4E inhibitory protein supports the idea that exchange of CBC20/80 for eIF4E at the cap can occur independent of any pioneering round of translation.

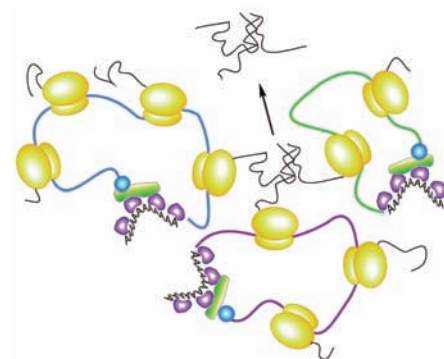
### Location, Location, Location

Oskar is but one example of a plethora of localized mRNPs. Such localization, usually coupled with regulated translation, serves to restrict synthesis of the encoded protein to a

specific subcellular compartment. For example, repression of mating-type switching by *S. cerevisiae* daughter cells is facilitated by localizing the mRNA encoding Ash1p, a transcriptional repressor, to the developing bud tip. In all, 24 transcripts have been shown to localize to the bud tip and 8 to the vicinity of yeast mitochondria (31). In metazoans, regulated translation of localized mRNAs is particularly rife in highly polarized cells such as oocytes and neurons. Fully one-tenth of randomly selected *Drosophila* ovarian mRNAs localize to the anterior pole of the oocyte, and ~400 different mRNAs have been identified in mammalian neuronal dendrites (32).

Mechanisms for mRNA localization include active transport along the cytoskeleton, diffusion and anchoring, local protection from degradation, and local synthesis by subsets of nuclei in syncytial cells. In many instances, a combination of mechanisms work on a single transcript. For example, oskar mRNA is transported along microtubules by kinesin and then becomes anchored at the posterior pole by its own gene product. Another posterior pole mRNA, nanos, achieves its localization pattern by diffusion and anchoring, along with regional stabilization. Some localized mRNAs travel as individual mRNPs, whereas others appear to migrate as higher order RNP structures or particles. In neurons, such particles have been estimated to contain ~30 mRNAs and have diameters up to 1  $\mu$ m (32).

Although mRNA localization and regulated translation have been most intensively studied in specialized cells such as oocytes and neurons, it now appears that many mRNAs may exhibit asymmetric localization even in somatic cells. One particularly well-characterized example is  $\beta$ -actin mRNA, which localizes to sites of actin polymerization at the leading edges of crawling cells (33). Local  $\beta$ -actin



**Fig. 2.** Schematic showing cotranslational assembly of a protein complex encoded by a family of colocalized mRNAs. Functional circularization of mRNAs by a network of interactions between eIF4E, eIF4G, and PABPs promotes efficient translation by polyribosomes. Physical juxtaposition of mRNAs encoding individual components of the complex may facilitate cotranslational polypeptide interaction and complex assembly.

translation likely contributes to overall cell motility by supplying new actin monomers precisely at the sites where they are needed. Another set of proteins found at leading edges is the Arp2/3 complex, a stable assemblage of seven polypeptides responsible for nucleating branched actin filaments. New data indicate that upon serum induction, all seven Arp2/3 complex mRNAs are recruited to the leading protrusions of polarized fibroblasts by a mechanism requiring both actin filaments and microtubules (34). Such colocalization of mRNAs encoding all the components of a single macromolecular complex has numerous potential advantages. Not only is translation and degradation of colocalized mRNAs amenable to coordinate regulation, synthesis of the component parts in close physical proximity very likely facilitates assembly of the complex. The high local concentration of nascent polypeptides might even promote their cotranslational association (Fig. 2), an arranged marriage having added advantages of preventing alternate folding pathways and excluding unwanted interactions with competing cellular components.

### The End of the Line (or Is It?): P-bodies and Stress Granules

Because of their key position as transient intermediates in the flow of genetic information, mRNAs have limited lifetimes. As with all other aspects of mRNA metabolism, these half-lives are subject to modulation by changing intra- and extracellular conditions. How long an mRNA lives depends on how efficiently the mRNA degradation machinery is recruited to that mRNP. In general, the core degradation machinery attacks mRNA from its ends—the 3' poly(A) tail is removed by a host of deadenylases, while the 5' cap is removed by specific decapping enzymes. The body of the message is then degraded by 5'→3' and 3'→5' exonucleases. Whether a particular mRNA is destroyed primarily in one direction or the other is a function of which set of enzymes is most active in that particular cell type and which set is recruited most efficiently to that mRNP (35). Of course, endonucleolytic degradation mechanisms also exist, most notably sequence-specific mRNA cleavage by the RNA-induced silencing complex (RISC) in association with endogenous small interfering RNA (siRNA) (5–7).

The general mRNA decay machinery is also required for the elimination of aberrant mRNAs containing a premature translational stop signal (nonsense mRNA) or lacking a translational signal altogether (nonstop mRNA) (28, 36). Such defective mRNAs can arise through a variety of mechanisms, including genetic mutation, missplicing, and premature polyadenylation. Their efficient elimination is thought to protect cells from the potentially deleterious consequences of inappropriately

terminated proteins. Recognition of nonsense and nonstop mRNAs as abnormal requires their functional engagement by ribosomes, which fail to terminate properly on both nonsense and nonstop mRNAs (36, 37). This improper termination leads to recruitment of the decay machinery, presumably through interactions with ribosome release factors and/or the empty A site tRNA binding pocket on the ribosome. In mammalian cells, decay of some nonsense mRNAs is quite efficient, occurring soon after they emerge from the nucleus and are still associated with CBC20/80 (28). However, it remains to be determined whether this timing is true of all mRNAs or is limited to those that immediately engage the translation apparatus upon export.

Consistent with the emerging idea that many mRNPs spend their productive lives at specific subcellular addresses or in working groups with other mRNPs, recent data also suggest that mRNPs go to specific places to die. In both yeast and mammalian cells, much of the mRNA decay machinery is concentrated in discrete cytoplasmic foci. These so-called cytoplasmic processing bodies, or “P-bodies” (PBs), appear to form around aggregates of mRNPs not actively involved in translation (38). Targeting of mRNPs to these structures requires their removal from the translationally active pool, one mechanism for which appears to be interaction with miRNAs and the RISC complex (39). Proof that mRNA decay occurs within PBs came with the demonstration that mRNA degradation intermediates accumulate there upon either general or mRNA-specific inhibition of decay (40, 41).

Whereas PBs may represent the end of the line for mRNPs, “stress granules” (SGs), related but distinct structures in mammalian cells, serve as temporary retirement homes. When mammalian cells are exposed to an assortment of environmental stresses, global translational arrest of “housekeeping” transcripts is accompanied by the formation of distinct cytoplasmic structures containing translationally inactive mRNPs, 40S ribosomal subunits, and the mRNA binding proteins TIA-1 and TIAR. Prionlike domains in TIA-1/TIAR are thought to self-oligomerize and promote SG assembly (42). Although translational arrest upon application of stress is widespread, selective translation of heat shock proteins, as well as some transcription factors, under these conditions allows the cell to repair the stress-induced damage while conserving anabolic energy. When the stress is relieved, SGs disassemble and the sequestered mRNAs either return to the translationally active pool or are targeted for degradation in PBs (43, 44). So far, SGs have not been observed in budding yeast. Instead, it has been suggested that *S. cerevisiae* PBs serve dual roles as way stations for translationally inactive mRNPs and sites of mRNA degradation (38).

In summary, recent advances have greatly heightened our appreciation of the extent to which eukaryotic cells regulate gene expression at the mRNP level. In some areas, such as the control of translation by 4E interacting proteins, underlying themes have begun to emerge. In other areas, such as the spatial localization of protein synthesis and the existence of genome-wide posttranscriptional regulatory networks, we have only begun to scratch the surface. No doubt further surprises await discovery along the path from birth to death of eukaryotic mRNAs.

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## From Birth to Death: The Complex Lives of Eukaryotic mRNAs

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