

Translational Control in Oocyte Development

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Translational control of specific mRNAs is a widespread mechanism of gene regulation, and it is especially important in pattern formation in the oocytes of organisms in which the embryonic axes are established maternally. *Drosophila* and *Xenopus* have been especially valuable in elucidating the relevant molecular mechanisms. Here, we comprehensively review what is known about translational control in these two systems, focusing on examples that illustrate key concepts that have emerged. We focus on protein-mediated translational control, rather than regulation mediated by small RNAs, as the former appears to be predominant in controlling these developmental events. Mechanisms that modulate the ability of the specific mRNAs to be recruited to the ribosome, that regulate polyadenylation of specific mRNAs, or that control the association of particular mRNAs into translationally inert ribonucleo-protein complexes will all be discussed.

Since it was first described in sea urchin and *Drosophila* oocytes more than five decades ago, masked or maternal mRNA has served as a preeminent model for the study of translational control. The key questions that have been addressed over the years in a variety of organisms, perhaps most vividly exemplified by *Drosophila* and *Xenopus*, are how do mRNAs come to be translationally repressed, how are they activated, and how is translational regulation regionally restricted? *Drosophila* genetics has identified a variety of mRNAs whose translational control is essential for embryonic development (e.g., *oskar*, *nanos*, and *gurken*). Moreover, genetics, as well as biochemistry, has revealed some of the factors that control the expression of these mRNAs, such as the

RNA binding proteins Cup and Pumilio. *Xenopus*, on the other hand, is not an organism particularly well suited to genetic analysis. However, *Xenopus* oocytes are an excellent biochemical system owing to their abundance, large size for the injection of molecules, and their synchronous entry into the meiotic divisions when cultured *ex vivo* in medium containing progesterone. Indeed, *Xenopus* oocytes have been instrumental in revealing how the translation of maternal mRNAs during oocyte maturation is regulated by changes in poly(A) tail length. Lessons learned in these systems have been important for revealing not only general principals of translational control, but also for serving as paradigms for translational control in other cells and tissues.

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J.D. Richter and P. Lasko

Here, we focus our attention on select examples of translational control in *Drosophila* and *Xenopus* oocytes. As the literature on this subject is now vast, we believe that the reader is best served by focusing on translational control in oocytes of these two organisms, where many of the key concepts in the field were first developed and have been most completely elaborated.

TRANSLATIONAL REGULATION UNDERLIES ESTABLISHMENT OF EMBRYONIC PATTERN IN *DROSOPHILA*

Molecular asymmetries underlying embryonic axis patterning and germ cell specification are established in *Drosophila* largely by position-dependent translational regulation of maternally transcribed mRNAs that are deposited into the oocyte. This translational regulation is often coupled with localization mechanisms that concentrate the mRNA in the spatial domain of the oocyte where the corresponding protein will be expressed and that reduce its concentration in areas where its expression would be deleterious to development. A detailed discussion of mRNA localization mechanisms is beyond the scope of this article, and the relevant work has been recently reviewed (Becalska and Gavis 2009). Translational repression in the developing oocyte is especially complex, and as will be discussed below, different mechanisms silence translation of specific mRNAs at different developmental stages, thus restricting their potential translation to particular spatial regions. Comparatively less is known about repression is alleviated and translation is activated for these regulated mRNAs, although increasing evidence implicates the DEAD-box helicase Vasa (Vas) as a positive regulator of specific target mRNAs.

TRANSLATIONAL REGULATION OF *oskar*

Localization of *oskar* (*osk*) mRNA and its targeted translation initiates assembly of polar granules and specification of the posterior soma and germ cells (Ephrussi and Lehmann 1992). Two different isoforms, termed Long

Osk and Short Osk, are expressed from different initiation codons (Markussen et al. 1995). Short Osk is sufficient to induce pole plasm assembly and rescue the developmental functions of *osk*, whereas Long Osk induces F-actin projections that are required for anchoring its mRNA in the posterior pole plasm (Markussen et al. 1995; Vanzo et al. 2007).

Drosophila oogenesis is conventionally divided into 14 developmental stages (Spradling 1993). Until stage 9, translation of *osk* is silenced throughout the nurse cells and oocyte, whereas afterward, *osk* translation is activated in the posterior pole plasm, while remaining repressed elsewhere. Repression of *osk* translation in early oogenesis involves RNA interference (RNAi) (Findley et al. 2003; Cook et al. 2004; Tomari et al. 2004; Chen et al. 2007; Lim and Kai 2007; Pane et al. 2007). There is, however, little specific information as to the mechanisms involved. miRNA-mediated translational repression is believed to act after initiation (Bushati and Cohen 2007).

As *osk* mRNA moves to the pole plasm, it is translationally repressed by Cup (Fig. 1). Cup, like Maskin (see below), competes with eIF4G for binding to eIF4E (Filardo and Ephrussi 2003; Wilhelm et al. 2003; Nakamura et al. 2004; Nelson et al. 2004; Zappavigna et al. 2004). Cup interacts with Bruno (Bru), an RNA binding protein that negatively regulates *osk* during its localization. Bru contains three RNA recognition motifs (RRMs) that interact directly with specific sequences (Bru-response elements, or BREs) in the *osk* 3' UTR, and repress its translation (Kim-Ha et al. 1995; Webster et al. 1997; Lie and Macdonald 1999; Snee et al. 2008), most likely by recruiting Cup. The third RRM of Bru has recently been shown to have a noncanonical structure, and 40 amino acids that extend it aminoterminaly significantly enhance RNA binding (Lyon et al. 2009).

Bru also packages *osk* RNA into large particles that render it inaccessible to the translational machinery (Chekulaeva et al. 2006). These silencing particles contain Cup and Maternal expression at 31B (Me31B) (Nakamura et al. 2001). Although Cup and Me31B

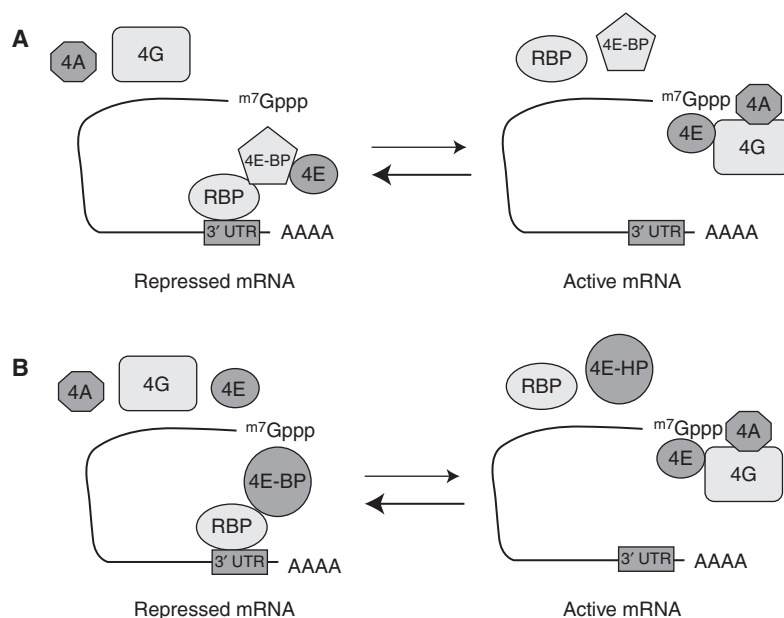


Figure 1. Translational repression by eIF4E binding proteins. (A) An eIF4E binding protein (4E-BP) competes with eIF4G (4G) for interaction with the cap-binding protein eIF4E (4E). In general eIF4G has a higher affinity for eIF4E than does the regulatory eIF4E binding protein and an active cap-binding complex including eIF4A (4A) can assemble, but that equilibrium is reversed if the regulatory protein is recruited to a target mRNA by an RNA binding protein (RBP) that binds to the 3' UTR. Then eIF4E is sequestered away from the cap-binding complex and translation is repressed. 4E-BPs that operate in this way include *Drosophila* Cup and *Xenopus* Maskin. RBPs that recruit these 4E-BPs to specific mRNAs include *Drosophila* Bru, *Drosophila* Smg, and *Xenopus* CPEB. (B) Translational repression by 4E homology proteins. 4E homology protein (4E-HP) can bind the 5' cap structure of the mRNA but not eIF4G. It competes with eIF4E for cap binding and represses translation of mRNAs with which it is associated. The affinity of 4E-HP for the 5' cap structure is less than that of eIF4E, but that equilibrium is reversed if 4E-HP is recruited to target mRNAs by RBPs that bind the 3' UTR. In *Drosophila*, 4E-HP can be recruited to target mRNAs by Bicoid or by the Pumilio/Nanos/Brain Tumor complex, whereas in mammalian cells 4E-HP can be recruited by Prep1.



can be copurified, *Me31B* mutations affect *osk* translation at an earlier stage of oogenesis than do *cup* mutations, suggesting that their functions are not mutually dependent. *Me31B* associates with Bru, implying it may be required for Cup-dependent *osk* silencing at stage 9, but as *Me31B*-null egg chambers degenerate around stage 6, it has not been possible to directly test this (Nakamura et al. 2001). Polypyrimidine tract binding protein (PTB) also promotes the formation of *osk*-containing silencing particles (Besse et al. 2009). To accomplish this, PTB binds directly to multiple sites in the *osk* 3' UTR, cross-linking many *osk* and PTB molecules into aggregates. It is unknown whether

PTB recognizes mRNA targets other than *osk*, but it seems likely that silencing particles containing PTB would include more than one species of mRNA. A detailed ultrastructural analysis of *osk* mRNA localization and its assembly into ribonucleoprotein complexes (RNPs) has recently been completed (Trucco et al. 2009). This study showed that the average size of *osk*-containing RNPs increases drastically from the anterior to the posterior of stage 8–10 oocytes, and that these particles form large aggregates in the pole plasm. This study also documented when and where proteins involved in *osk* localization become associated with these RNPs. An extension of this approach could

J.D. Richter and P. Lasko

reveal a great deal about the dynamics of the association of PTB, Me31B, and Bru with *osk* and potentially other mRNAs.

Hrp48, an abundant RNA-binding protein that interacts with elements in both the 5' and 3' UTRs of *osk*, also contributes to its translational repression (Huynh et al. 2004; Yano et al. 2004; Norvell et al. 2005). Hrp48 is recruited to *osk*-containing RNPs in the nurse cell nuclei and remains associated with these particles in the cytoplasm (Trucco et al. 2009). Hrp48 interacts with Squid (Sq) and Ovarian Tumor (Otu) (Norvell et al. 1999; Goodrich et al. 2004), and all three have been functionally implicated in *osk* mRNA localization and translational repression. Cup also copurifies with Hrp48 and Sq (Clouse et al. 2008), but this association requires RNA, making it unlikely that these proteins operate in an analogous manner to Bru. Glorund, an hnRNP F/H family member, may be another component of this complex (Kalifa et al. 2009). Another RNA binding protein, Bicaudal-C (Bic-C), has been implicated genetically as a negative regulator of *osk* translation (Saffman et al. 1998). Bic-C directly recruits the CCR4 deadenylase complex to target mRNAs through an association with its NOT3/5 subunit (Chicoine et al. 2007). These targets could potentially include *osk*.

Derepression and activation of *osk* translation in the pole plasm may be distinct processes (Gunkel et al. 1998). A key activator of *osk* translation is Orb, the *Drosophila* homolog of cytoplasmic polyadenylation element binding protein (CPEB) (see below). Orb interacts with the *osk* 3' UTR, and in *orb* mutants *osk* translation is reduced, suggesting that cytoplasmic polyadenylation might underlie activation of *osk* translation (Chang et al. 1999). Orb interacts physically with Bru, yet the addition of a long poly(A) tail to a chimeric *osk-lacZ* mRNA does not overcome Bru-mediated repression in vitro (Castagnetti and Ephrussi 2003). Orb directly associates with PAP and Wispy, two poly(A) polymerases that act at different developmental times: PAP is required during mid-oogenesis to promote Osk expression, whereas Wispy functions only during late oogenesis and in the early embryo (Juge

et al. 2002; Benoit et al. 2008). Bic-C interacts with Orb, PAP, and Wispy, and possibly inhibits their association with target mRNAs (Castagnetti and Ephrussi 2003; Chicoine et al. 2007; Benoit et al. 2008; Cui et al. 2008). Consistent with this, *Bic-C* and *orb* display antagonistic genetic interactions.

osk translation in the germ plasm also requires Staufen (Stau), a dsRBD type RNA binding protein essential as well for its localization (Micklem et al. 2000). Unlike Hrp48, Stau first becomes associated with *osk*-containing mRNPs in the nurse cell cytoplasm (Trucco et al. 2009). The mechanism through which Stau activates *osk* translation is unknown, but it involves more than simply relieving Bru-mediated repression, as its activity is BRE-independent (Kim-Ha et al. 1995). Also, Aub activates *osk* translation in the pole plasm, and, as for Stau, its function does not require the BRE (Wilson et al. 1996; Harris and Macdonald 2001). The basis for the contrasting roles for Aub with respect to unlocalized *osk* in early oogenesis and localized *osk* in the pole plasm remains unexplained.

TRANSLATIONAL REGULATION OF *nos*

Posterior targeting of Nos protein also involves RNA localization and translational regulation. A 90 nt region of the *nos* 3' UTR, termed the translational control element (TCE), forms a complex secondary structure, and mutations that disrupt any portion of it prevent the binding of repressors and render the entire element inactive (Crucs et al. 2000; Forrest et al. 2004). Different parts of the TCE interact with different *trans*-acting factors at different developmental stages to ensure translational repression of unlocalized *nos* mRNA. During late oogenesis, repression of *nos* is mediated by Glorund (Glo), which binds to the stem of stem-loop III of the TCE (Kalifa et al. 2006).

The loop of stem-loop II of the TCE contains a Smaug Recognition Element (SRE), the binding site for another translational repressor called Smaug (Smg) (Smibert et al. 1996). Smg interacts with Cup, and Cup binding is required for Smg-mediated repression of



SRE-containing mRNAs in embryo extracts (Nelson et al. 2004). Smg is therefore believed to function in a manner analogous to Bru. However, Smg has a second more widespread function in mRNA metabolism. It interacts directly with the POP2 subunit of the CCR4 deadenylase complex, and recruits it to a large set of maternal mRNAs in the early embryo, including *nos*, targeting them for decay (Semotok et al. 2005; Zaessinger et al. 2006; Tadros et al. 2007; Benoit et al. 2009). Thus, *nos* mRNA is repressed in two distinct ways by Smg: by cap-dependent translational repression and by deadenylation of the silenced transcript. Osk relieves Smg/CCR4-dependent deadenylation of *nos*, thus enabling its translation in the pole plasm (Zaessinger et al. 2006).

A recent study (Rangan et al. 2009) compares the translational regulation of 10 other mRNAs with similar localization kinetics to *nos* with the regulation of *nos* itself. In all cases, translational regulation elements resided in the 3' UTRs. Often, translational activation correlated with an increase in poly(A) tail length, but surprisingly for *pgc* and *gcl* mRNAs, reduction of *orb* activity had little effect on translation. As several translational regulators target the CCR4 deadenylase, it appears it may be more effective to regulate deadenylation rather than polyadenylation in this system.

TRANSLATIONAL REGULATION OF *grk*

As for *osk* and *nos*, *grk* expression is regulated through mRNA localization coupled with translational control. Bru, K10, and Sqd have all been implicated as translational repressors of *grk*, and Bru binds directly to the *grk* 3' UTR (Saunders and Cohen 1999; Filardo and Ephrussi 2003; Cáceres and Nilson 2009), although a link to Cup has not yet been established. *grk* translation is positively regulated by the DEAD-box helicase Vas. Severe *vas* mutations largely block Grk accumulation in early oocytes (Styhler et al. 1998; Tomancak et al. 1998). Vas interacts physically and functionally with eIF5B, a general translation factor required for recruitment of the 60S ribosomal subunit to the initiation complex (Carrera et al. 2000). Oocytes that contain only a form of Vas (Vas Δ 617) that is specifically compromised for eIF5B binding largely fail to accumulate Grk, implying that Vas positively regulates *grk* by recruiting eIF5B (Fig. 2) (Johnstone and Lasko 2004). Vas has recently been shown to bind specifically to a U-rich motif present in the 3' UTR of another mRNA, *mei-P26*, and to positively regulate its translation in stem cells and dividing cystocytes through that interaction (Liu et al. 2009). Activation of *grk* mRNA translation at the antero-dorsal cortex of the developing oocyte is also mediated by poly(A) binding

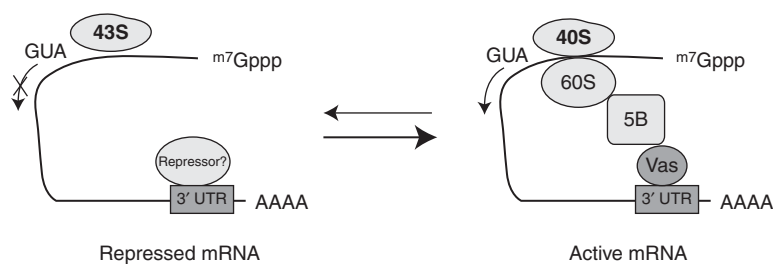


Figure 2. Proposed mechanism for Vas-mediated translational activation. eIF5B (5B) is required for recruitment of the large ribosomal subunit (60S) to the initiation complex, so that the elongation phase of translation can proceed. When this process is blocked, translation is stalled with the small ribosomal subunit and initiator tRNA-Met (43S) stalled at the start codon (AUG). Vas binds a sequence element in the 3' UTR and eIF5B (5B), thus promoting the recruitment of the 60S subunit and activating translation. Regulation of an mRNA expressed in erythroid cells, *r15-LOX*, at the level of subunit joining involves a 3' UTR binding repressor that inhibits eIF5B recruitment (Ostareck et al. 2003). It is unknown whether Vas-mediated activation involves displacement of such a repressor.

J.D. Richter and P. Lasko

protein 55D (PABP55D) in association with Encore (Clouse et al. 2008).

4EHP AND TRANSLATIONAL REPRESSION OF *caudal* AND *hunchback*

The homeodomain-containing protein Bicoid (Bcd) is a transcriptional regulator essential for anterior patterning in *Drosophila* (Berleth et al. 1988; Driever and Nüsslein-Volhard 1989). Bcd also associates with RNA and regulates translation, repressing *caudal* (*cad*) RNA, and producing a posterior-to-anterior gradient of Cad protein. Bcd binds to the 3' UTR of *cad* RNA, and *cad* repression depends on the presence of a 5' cap structure (Dubnau and Struhl 1996; Rivera-Pomar et al. 1996; Niessing et al. 1999, 2002).

4E homology protein (4EHP) is an eIF4E-related cap-binding protein that cannot bind eIF4G. Females homozygous for a hypomorphic allele of this gene produce embryos with anterior defects similar to embryos produced by *bcd* mutant mothers (Cho et al. 2005). These embryos also fail to repress *cad* translation in anterior regions. These phenotypes could be rescued by *4EHP* transgenic constructs, but not by constructs producing mutant forms of 4EHP that were abrogated for cap binding or for Bcd binding. Similarly, transgenically produced forms of Bcd that were abrogated for 4EHP binding could not repress *cad* translation. These results showed that simultaneous interactions of 4EHP with the cap structure and of Bcd with the *cad* 3' UTR renders *cad* mRNA translationally inactive. Recent evidence indicates that, like Bcd, a mammalian homeodomain-containing transcription factor, Prep1, also negatively regulates translation of a target gene, *Hoxb4*, through a mechanism that requires 4EHP and the *Hoxb4* 3' UTR (Villaescusa et al. 2009).

In *Drosophila*, 4EHP was also shown to be involved in *hb* translational repression (Cho et al. 2006). In wild-type embryos, translation of the uniformly distributed maternal *hunchback* (*hb*) mRNA is inhibited at the posterior to form an anterior-to-posterior protein concentration gradient. This inhibition requires assembly of an mRNP complex (the NRE-complex) that consists of Nanos (Nos), Pumilio (Pum) and

Brain tumor (Brat) proteins, and the Nos response element (NRE) present in the 3' UTR of *hb* mRNA (Wharton and Struhl 1991; Chagnovich and Lehmann 2001; Sonoda and Wharton 2001). The Hb gradient extends more posteriorly in embryos produced by *4EHP* mutant females, and 4EHP is recruited to the NRE-complex through binding to the dorsal surface of the NHL domain within Brat. Consistently, mutant forms of Brat abrogated for 4EHP binding do not fully support *hb* repression.

TRANSLATIONAL CONTROL BY CYTOPLASMIC POLYADENYLATION IN VERTEBRATES

Looking back, 1987–1989 were very good years for insights into posttranscriptional control of gene expression in vertebrates. Vassalli, Strickland, and their colleagues (Huarte et al. 1987) found that in maturing mouse oocytes, tissue plasminogen activator (tPA) mRNA underwent polyadenylation and translational activation. Similar observations were made in 1988 by the Dworkin laboratory (Smith et al. 1988a,b), which showed that a set of mRNAs underwent cytoplasmic polyadenylation and translational activation in maturing *Xenopus* oocytes. The year 1988 was also when Vassalli, Strickland and coworkers (Strickland et al. 1988) showed that “information” in the tPA 3' UTR promoted cytoplasmic polyadenylation and translation. Finally, in 1989 specific mRNAs in *Xenopus* oocytes were shown to contain specific *cis* elements that directed polyadenylation-induced translation (Fox and Wickens 1989; McGrew et al. 1989). Since these early observations got the ball rolling, many biochemical studies using *Xenopus* oocytes have revealed the details of how cytoplasmic polyadenylation works, how this process mediates translation, and how it controls oocyte development.

OOCYTE MATURATION

To place the translational control in a proper biological context, it is first necessary to consider the process of oocyte maturation particularly in *Xenopus laevis*, because most of what we



know about maternal mRNA expression in vertebrates comes from studies of that species (see Brook et al. 2009). Primordial germ cells (PGCs) divide mitotically while migrating to the developing gonad; once there, they enter meiosis, proceed through synapsis and recombination, and arrest at the end of prophase I, also known as diplotene, which resembles mitotic G2. The oocytes grow enormously by taking up vitellogenin from the bloodstream; during this time, they also synthesize and store large amounts of mRNA, tRNA, ribosomes, and all the factors necessary for translation. However, as little as 5% by mass of the mRNA is actively engaged in protein synthesis. The nontranslating or stored mRNAs are generally referred to as masked or maternal mRNA. Some of these mRNAs are translated when the oocytes reenter meiosis, a period known as oocyte maturation. The cells proceed through metaphase I (MI) and arrest at MII, where they await fertilization. Several other mRNAs are translated between MI and MII, whereas others are translated during fertilization or particular times of embryogenesis. Although no single mechanism regulates the translation of all mRNAs, one that is particularly important, and one that is understood in some biochemical detail, is cytoplasmic polyadenylation.

POLYADENYLATION-INDUCED TRANSLATION

The story of how poly(A) elongation controls translation during oocyte maturation begins, not surprisingly, in the nucleus. The synthesis of nuclear premRNAs proceeds far downstream from where the mature 3' end of the mRNA is located. A signal in the premRNA, AAUAAA, is usually found about 20 bases upstream of the mature 3' end (Richard and Manley 2009). This cleavage and polyadenylation signal, which is found on virtually all noncore histone mRNAs, is recognized by the cleavage and polyadenylation specificity factor CPSF. CPSF is composed of four subunits with sizes (in mammalian cells) of 150, 100, 73, and 30 kDa. Symplekin is another protein often intimately associated with CPSF (Shatkin and Manley

2000). Together with stimulatory factors, the CPSF 73-kDa subunit cleaves the RNA about 20 bases downstream from the AAUAAA (Mandel et al. 2006), whereupon a long poly(A) tail is added.

Some premRNAs have another key sequence that is often 20–100 bases upstream of AAUAAA, and that is the cytoplasmic polyadenylation element (CPE), which has the general structure of UUUUUU. Following splicing but still in the nucleus, the CPE is bound by the shuttling cytoplasmic polyadenylation element binding protein CPEB (Lin et al. 2010). Together with other proteins such as symplekin and the exon-junction protein eIF4AIII (see below), the ribonucleoprotein (RNP) complex is exported to the cytoplasm where it is joined by two new critical factors: Gld2 and PARN (Barnard et al. 2004; Kim and Richter 2006; Lin et al. 2010). Gld2 (germline development 2) is an unusual cytoplasmic poly(A) polymerase discovered in 2002 by three laboratories working in *Caenorhabditis elegans* and yeast (Read et al. 2002; Saitoh et al. 2002; Wang et al. 2002). PARN (poly(A) ribonuclease) is the second factor; as the name suggests, it is a deadenylating enzyme (Korner et al. 1998; Copeland and Wormington 2001). Both of these enzymes are active, but because PARN is the more active, the poly(A) tail originally acquired in the nucleus is now shortened, to about 20–40 bases. At this stage, Gld2 continues to catalyze polyadenylation but the adenylate residues are quickly removed by PARN. This “running in place” model for polyadenylation can theoretically take place for months on end (Kim and Richter 2006).

Progesterone secretion from the follicle cells induces oocyte entry into the meiotic divisions. This steroid interacts with a surface-associated receptor, the result of which is a cascade of events that ultimately leads to another meiotic arrest at MII (Tunquist and Mallet 2003). More germane to this discussion is the observation that progesterone induces a decrease in GSK-3 β levels that in turn induces the activation of the kinase Aurora A (Mendez et al. 2000a; Sarkissian et al. 2004). Aurora A, and/or MAP kinase and a GTP exchange factor XGef

J.D. Richter and P. Lasko

(Reverte et al. 2003; Martinez et al. 2005; Keady et al. 2007), which is also activated by progesterone, phosphorylates CPEB on serine 174, an event that not only induces a close association between CPEB and CPSF (Mendez et al. 2000b), but also expels PARN from the RNP complex (Kim and Richter 2006). Because PARN is no longer present, Gld2 polyadenylates the RNA by default (Fig. 3). In other words, polyadenylation is controlled by the presence or absence of PARN, and not by the recruitment of Gld2 or another poly(A) polymerase as originally suggested (Richter 1999).

Is a short poly(A) tail in and of itself sufficient to keep mRNAs silent until the tails are elongated? Apparently, this is not the case because another factor, Maskin, is a key regulator of translation. Maskin, which probably shuttles to the nucleus with CPEB and is exported with it and mRNA into the cytoplasm (Lin et al. 2010), not only binds CPEB, but the cap-binding factor eIF4E as well. Like *Drosophila* Cup, the binding of Maskin to eIF4E precludes

the binding of eIF4G to eIF4E, thus Maskin inhibits translation because eIF4G, indirectly through eIF3, recruits the 40S ribosomal subunit to the 5' end of the mRNA (Stebbins-Boaz et al. 1999; Cao and Richter 2006).

How is Maskin-mediated repression alleviated by polyadenylation? The answer to this question lies with another translationally dormant mRNA encoding the protein RINGO (Ferby et al. 1999). Oocytes contain no detectable RINGO protein but instead contain inactive RINGO mRNA that is translated on progesterone stimulation. RINGO mRNA translation is not controlled by CPEB and/or Maskin, but instead it is regulated by Pum2 (Pumilio 2), DAZL (deleted in azoospermia-like), and ePAB (embryonic poly(A) binding protein) (Padmanabhan and Richter 2006). Pum2 binds both a Pumilio binding element (PBE) in the RINGO 3' UTR as well as the 5' cap, thereby prevent eIF4E from binding the cap and repressing translation (Cao et al. 2010). On progesterone stimulation, Pum2

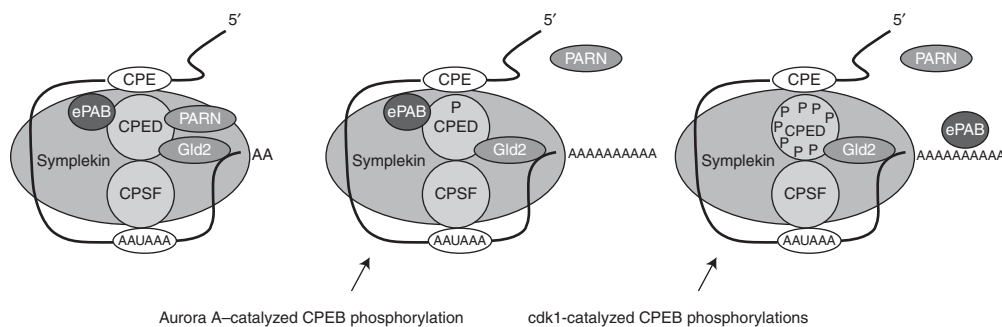


Figure 3. Cytoplasmic polyadenylation. In immature oocytes, certain maternal mRNAs contain a CPE (general structure of UUUUUAU) that is bound by CPEB, and the polyadenylation hexanucleotide AAUAAA that is bound by CPSF (for simplicity, only the 3' UTR is depicted in the figure). CPEB is also bound by Gld2, a poly(A) polymerase; PARN, a deadenylating enzyme; and ePAB, a poly(A) binding protein. The entire complex is assembled on symplekin, a scaffold protein. When exported from the nucleus, the CPE-containing RNA has a long poly(A) tail. Once assembled with CPEB and the other factors noted above, PARN shortens the poly(A) tail to ~20–40 nucleotides. Gld2 is also activated at this time and catalyzes poly(A) addition, but because PARN is more active, the poly(A) tail is maintained in a shortened state. Progesterone secretion from follicle cells sets off a signaling cascade in the oocyte that results in activation of the kinase Aurora A, which phosphorylates CPEB on a single site (serine 174). This event causes the expulsion of PARN from the ribonucleoprotein (RNP) complex, resulting in default Gld2-catalyzed polyadenylation. Next, the kinase cdk1 is activated, which phosphorylates CPEB on six additional sites; these events cause ePAB to dissociate from CPEB and bind the newly elongated poly(A) tail. ePAB not only protects the poly(A) tail from hydrolysis by exonucleases, but it binds the initiation factor eIF4G and helps stimulate translation.

dissociates from the mRNA and RINGO mRNA is translated (Padmanabhan and Richter 2006). RINGO protein, which is functionally similar to cyclin B1 (Nebreda and Ferby 2000; Karaïskou et al. 2001; Gutierrez et al. 2006), binds to and activates cdk1. The now active kinase cdk1 phosphorylates CPEB on six sites (excluding the Aurora A/MAP kinase site) (Mendez and Richter 2002; Kim and Richter 2007). These phosphorylation events cause the dissociation of another CPEB-bound factor, ePAB, which in turn now binds the newly elongated poly(A) tail (Fig. 3). ePAB bound to the poly(A) tail performs two tasks: It prevents deadenylation of PARN as well as other deadenylating enzymes such as CCR4/Not1 that are present in the oocyte cytoplasm, and it serves to dock eIF4G. At this point, PABP may potentiate the interaction of eIF4G with eIF4E, thus displacing Maskin (Cao et al. 2002; Kim and Richter 2007). Maskin is also phosphorylated, which also induces its dissociation from eIF4E (Barnard et al. 2005; Cao et al. 2006). Thus, the loop is now closed: eIF4E-bound eIF4G positions the 40S subunit on the 5' end of the mRNA and translation is initiated.

Although these events are probably necessary to enter MI, additional events are important for progression to MII. For example, it has been known for some time that polyadenylation does not occur en masse, but instead there is sequence specificity as to when it occurs (Sheets et al. 1994; Stebbins-Boaz and Richter 1994, 1996; Mendez et al. 2002). Pique et al. (2008) have found that the number of CPEs, and their proximity to PBEs, determines, at least in part, the timing of when the polyadenylation occurs (see also Nakahata et al. 2001, 2003). Moreover, this polyadenylation may also be controlled by the amount of CPEB that is present because the majority of this protein undergoes ubiquitin-mediated destruction early during maturation (Mendez et al. 2002; Ververt et al. 2001).

The oocyte's arrest at MII also requires changes in poly(A) tail length, particularly of the mRNA encoding Emi2, an inhibitor of the anaphase promoting complex/cyclosome (Tunquist and Maller 2003). Emi2 is required

specifically at MII to arrest meiotic progression, and this is controlled by CPEB-mediated cytoplasmic polyadenylation of Emi2 mRNA (Tung et al. 2007). This process of meiotic arrest is more complex, for it involves negative feedback loops that mediate not only the expression of CPE-containing RNAs, but ARE (AU-rich element)-containing mRNAs as well (Belloc and Mendez 2008).

Several other proteins have been found to be associated with the CPEB complex, in particular p54/DDX6/RCK1 (Minshall et al. 2001). This protein, a DEAD box helicase that is the ortholog of *Drosophila* Me31B, is a component of p-bodies. It resides in a complex with CPEB, and in RNA tethering experiments, represses translation (Minshall et al. 2009). 4E-T, a protein with Maskin-like functions, also binds CPEB as well as eIF4E (Minshall et al. 2007). Because 4E-T appears to bind CPEB specifically in small, early-stage oocytes, there may be a developmental switch between 4E-T and Maskin as the oocytes grow.

Additional factors are also involved in a developmental switch during oogenesis. Musashi is an RNA binding protein that was initially shown to control neural stem-cell fate, be involved in certain cancers, and influence oocyte meiotic progression (MacNicol et al. 2008). Musashi, together with CPEB and Pumilio, may help control the timing of when particular mRNAs are polyadenylated and translated during maturation (Charlesworth et al. 2006; Arumugam et al. 2009). The biochemical mechanism by which Musashi accomplishes these tasks is unclear.

CONCLUDING REMARKS

Drosophila and *Xenopus* oocytes have served as model systems not only for gamete development, but also for general principles of translational control. Moreover, when these evolutionarily distant organisms have conserved mechanisms of translational control, one might imagine that such mechanisms occur not only in gametes, but in other species, other tissues, or in other developmental events. Consider, for example, cytoplasmic polyadenylation-induced

J.D. Richter and P. Lasko

translation. This process controls translation in *Xenopus* and *Drosophila* oocytes (reviewed above; see also Dworkin and Hershey 1981; Salles et al. 1994; for early studies), in sea urchin eggs (Wilt 1973; Duncan and Humphreys 1984), in mouse oocytes (Tay and Richter 2001), and probably in every other metazoan as well. Cytoplasmic polyadenylation-induced translation is not restricted to oocytes; it occurs in the brain where it mediates neuronal synaptic plasticity, and learning and memory (Wu et al. 1998; Alarcon et al. 2004; Berger-Sweeney et al. 2006), and in mitotic mammalian somatic cells where it controls cellular senescence (Groisman and Richter 2006; Burns and Richter 2008). Thus, cytoplasmic polyadenylation as a regulatory process for translation is conserved both phylogenetically as well as ontogenically.

When discussing conservation of regulatory proteins among animal groups, one usually thinks of primary sequence conservation. However, in the case of *Drosophila* and *Xenopus* oocytes, there is also functional conservation of a key factor that controls translation. Maskin was identified in *Xenopus* oocytes as the protein that bridges the CPEB-controlled cytoplasmic polyadenylation machinery with the translational regulatory apparatus (i.e., the cap-binding complex of eIF4E and eIF4G). However, a clear ortholog of Maskin is not present in *Drosophila*. Its closest fly counterpart is TACC (transforming acidic coiled coil protein), but as TACC does not contain the eIF4E binding domain it is unlikely to influence translation through a mechanism similar to that used by Maskin. However, *Drosophila* contains Cup, a protein that is functionally equivalent to Maskin in that on the one hand, it interacts with a protein that binds a specific 3' UTR element while on the other hand, it binds eIF4E. Another *Drosophila* protein related to Cup, CG32016, has not yet been well studied but is likely to have a similar function. Recently, two other proteins functionally equivalent to Maskin/Cup have been identified in the nervous system of vertebrates, neuroguidin (Jung et al. 2006) and CYFIP1 (Napoli et al. 2008). It seems unlikely that the activities of neuroguidin or CYFIP1 would not have been revealed in a

timely manner without the precedential observations of Maskin and Cup. Once again, it is easy to see why oocyte molecular biology is not just for developmental biologists anymore.

REFERENCES

- Alarcon JM, Hodgman R, Theis M, Huang YS, Kandel ER, Richter JD. 2004. Selective modulation of some forms of schaffer collateral-CA1 synaptic plasticity in mice with a disruption of the *CPEB-1* gene. *Learn Mem* **11**: 318–327.
- Arumugam K, Wang Y, Hardy LL, Macnicol MC, Macnicol AM. 2009. Enforcing temporal control of maternal mRNA translation during oocyte cell-cycle progression. *EMBO* **29**: 387–397.
- Barnard DC, Ryan K, Manley JL, Richter JD. 2004. Symplekin and xGLD-2 are required for CPEB-mediated cytoplasmic polyadenylation. *Cell* **119**: 641–651.
- Barnard DC, Cao Q, Richter JD. 2005. Differential phosphorylation controls Maskin association with eukaryotic translation initiation factor 4E and localization on the mitotic apparatus. *Mol Cell Biol* **25**: 7605–7615.
- Becalska AN, Gavis ER. 2009. Lighting up mRNA localization in *Drosophila* oogenesis. *Development* **136**: 2493–2503.
- Belloc E, Méndez R. 2008. A deadenylation negative feedback mechanism governs meiotic metaphase arrest. *Nature* **452**: 1017–1021.
- Benoit P, Papin C, Kwak JE, Wickens M, Simonelig M. 2008. PAP- and GLD-2-type poly(A) polymerases are required sequentially in cytoplasmic polyadenylation and oogenesis in *Drosophila*. *Development* **135**: 1969–1979.
- Benoit B, He CH, Zhang F, Votruba SM, Tadros W, Westwood JT, Smibert CA, Lipshitz HD, Theurkauf WE. 2009. An essential role for the RNA-binding protein Smaug during the *Drosophila* maternal-to-zygotic transition. *Development* **136**: 923–932.
- Berger-Sweeney J, Zearfoss NR, Richter JD. 2006. Reduced extinction of hippocampal-dependent memories in CPEB knockout mice. *Learn Mem* **13**: 4–7.
- Berleth T, Burri M, Thoma G, Bopp D, Riechstein S, Frigerio G, Noll M, Nüsslein-Volhard C. 1988. The role of localization of *bicoid* RNA in organizing the anterior pattern of the *Drosophila* embryo. *EMBO J* **7**: 1749–1756.
- Besse F, López de Quinto S, Marchand V, Trucco A, Ephrussi A. 2009. *Drosophila* PTB promotes formation of high-order RNP particles and represses *oskar* translation. *Genes Dev* **23**: 195–207.
- Brook M, Smith JW, Gray NK. 2009. The DAZL and PABP families: RNA-binding proteins with interrelated roles in translational control in oocytes. *Reproduction* **137**: 595–617.
- Burns DM, Richter JD. 2008. CPEB regulation of human cellular senescence, energy metabolism, and p53 mRNA translation. *Genes Dev* **22**: 3449–3460.
- Bushati N, Cohen SM. 2007. microRNA functions. *Annu Rev Cell Dev Biol* **23**: 175–205.



- Cáceres L, Nilson LA. 2009. Translational repression of *gurken* mRNA in the *Drosophila* oocyte requires the hnRNP Squid in the nurse cells. *Dev Biol* **326**: 327–334.
- Cao Q, Richter JD. 2002. Dissolution of the maskin-eIF4E complex by cytoplasmic polyadenylation and poly(A)-binding protein controls cyclin B1 mRNA translation and oocyte maturation. *EMBO J* **21**: 3852–3862.
- Cao Q, Kim JH, Richter JD. 2006. CDK1 and calcineurin regulate Maskin association with eIF4E and translational control of cell cycle progression. *Nat Struct Mol Biol* **13**: 1128–1134.
- Cao Q, Padmanabhan K, Richter JD. 2010. Pumilio 2 controls translation by competing with eIF4E for 7-methyl guanosine cap recognition. *RNA* **16**: 221–227.
- Carrera P, Johnstone O, Nakamura A, Casanova J, Jäckle H, Lasko P. 2000. VASA mediates translation through interaction with a *Drosophila* yIF2 homolog. *Mol Cell* **5**: 181–187.
- Castagnetti S, Ephrussi A. 2003. Orb and a long poly(A) tail are required for efficient *oskar* translation at the posterior pole of the *Drosophila* oocyte. *Development* **130**: 835–843.
- Chagnovich D, Lehmann R. 2001. Poly(A)-independent regulation of maternal *hunchback* translation in the *Drosophila* embryo. *Proc Natl Acad Sci* **98**: 11359–11364.
- Chang JS, Tan L, Schedl P. 1999. The *Drosophila* CPEB homolog, Orb, is required for Oskar protein expression in oocytes. *Dev Biol* **215**: 91–106.
- Charlesworth A, Wilczynska A, Thampi P, Cox LL, MacNicol AM. 2006. Musashi regulates the temporal order of mRNA translation during *Xenopus* oocyte maturation. *EMBO J* **25**: 2792–2801.
- Chekulaeva M, Hentze MW, Ephrussi A. 2006. Bruno acts as a dual repressor of *oskar* translation, promoting mRNA oligomerization and formation of silencing particles. *Cell* **124**: 521–533.
- Chen Y, Pane A, Schüpbach T. 2007. *cutoff* and *aubergine* mutations result in retrotransposon upregulation and checkpoint activation in *Drosophila*. *Curr Biol* **17**: 637–642.
- Chicoine J, Benoit P, Gamberi C, Paliouras M, Simonelig M, Lasko P. 2007. Bicaudal-C recruits CCR4-NOT deadenylase to target mRNAs and regulates oogenesis, cytoskeletal organization, and its own expression. *Dev Cell* **13**: 691–704.
- Cho P, Poulin F, Cho-Park YA, Cho-Park IB, Chicoine JD, Lasko P, Sonenberg N. 2005. A new paradigm for translational control: Inhibition of *caudal* via 5′-3′ tethering by Bicoid and the eIF4E cognate 4EHP. *Cell* **121**: 411–423.
- Cho P, Gamberi C, Cho-Park YA, Cho-Park IB, Lasko P, Sonenberg N. 2006. Cap-dependent translational inhibition establishes two opposing morphogen gradients in the *Drosophila* embryo. *Curr Biol* **16**: 2035–2041.
- Clouse KN, Ferguson SB, Schüpbach T. 2008. Squid, Cup, and PABP55B function together to regulate *gurken* translation in *Drosophila*. *Dev Biol* **313**: 713–724.
- Cook HA, Koppetsch BS, Wu J, Theurkauf WE. 2004. The *Drosophila* SDE3 homolog Armitage is required for *oskar* mRNA silencing and embryonic axis specification. *Cell* **116**: 817–829.
- Copeland PR, Wormington M. 2001. The mechanism and regulation of deadenylation: Identification and characterization of *Xenopus* PARN. *RNA* **7**: 875–886.
- Crucs S, Chatterjee S, Gavis ER. 2000. Overlapping but distinct RNA elements control repression and activation of *nanos* translation. *Mol Cell* **5**: 457–467.
- Cui J, Sackton KL, Horner VL, Kumar KE, Wolfner MF. 2008. Wispy, the *Drosophila* homolog of GLD-2, is required during oogenesis and egg activation. *Genetics* **178**: 2017–2029.
- Driever W, Nüsslein-Volhard C. 1989. The Bicoid protein is a positive regulator of *hunchback* transcription in the early *Drosophila* embryo. *Nature* **337**: 138–143.
- Dubnau J, Struhl G. 1996. RNA recognition and translational regulation by a homeodomain protein. *Nature* **379**: 694–699.
- Duncan R, Humphreys T. 1984. The poly(A)⁺RNA sequence complexity is also represented in poly(A)[−]RNA in sea-urchin embryos. *Differentiation* **28**: 24–29.
- Dworkin MB, Hershey JW. 1981. Cellular titers and subcellular distributions of abundant polyadenylate-containing ribonucleic acid species during early development in the frog *Xenopus laevis*. *Mol Cell Biol* **1**: 983–993.
- Ephrussi A, Lehmann R. 1992. Induction of germ cell formation by *oskar*. *Nature* **358**: 387–392.
- Ferby I, Blazquez M, Palmer A, Eritja R, Nebreda AR. 1999. A novel p34^{cdc2}-binding and activating protein that is necessary and sufficient to trigger G₂/M progression in *Xenopus* oocytes. *Genes Dev* **13**: 2177–2189.
- Filardo P, Ephrussi A. 2003. Bruno regulates *gurken* during *Drosophila* oogenesis. *Mech Dev* **120**: 289–297.
- Findley SD, Tamanaha M, Clegg NJ, Ruohola-Baker H. 2003. *maelstrom*, a *Drosophila* spindle-class gene, encodes a protein that colocalizes with Vasa and RDE1/AGO1 homolog, Aubergine, in nuage. *Development* **130**: 859–871.
- Forrest KM, Clark IE, Jain RA, Gavis ER. 2004. Temporal complexity within a translational control element in the *nanos* mRNA. *Development* **131**: 5849–5857.
- Fox CA, Sheets MD, Wickens MP. 1989. Poly(A) addition during maturation of frog oocytes: distinct nuclear and cytoplasmic activities and regulation by the sequence UUUUUAU. *Genes Dev* **3**: 2151–2162.
- Goodrich JS, Clouse KN, Schüpbach T. 2004. Hrb27C, Sqd, and Otu cooperatively regulate *gurken* RNA localization and mediate nurse cell chromosome dispersion in *Drosophila* oogenesis. *Development* **131**: 1949–1958.
- Groisman I, Ivshina M, Marin V, Kennedy NJ, Davis RJ, Richter JD. 2006. Control of cellular senescence by CPEB. *Genes Dev* **20**: 2701–2712.
- Gunkel N, Yano T, Markussen F-H., Olsen LC, Ephrussi A. 1998. Localization-dependent translation requires a functional interaction between the 5′ and 3′ ends of *oskar* mRNA. *Genes Dev* **12**: 1652–1664.
- Gutierrez GJ, Vögtlin A, Castro A, Ferby I, Salvagiotto G, Ronai Z, Lorca T, Nebreda AR. 2006. Meiotic regulation of the CDK activator RINGO/Speedy by ubiquitin-proteasome-mediated processing and degradation. *Nat Cell Biol* **8**: 1084–1094.
- Harris AN, Macdonald PM. 2001. *aubergine* encodes a *Drosophila* polar granule component required for pole cell

J.D. Richter and P. Lasko

- formation and related to eIF2C. *Development* **128**: 2823–2832.
- Huarte J, Belin D, Vassalli A, Strickland S, Vassalli JD. 1987. Meiotic maturation of mouse oocytes triggers the translation and polyadenylation of dormant tissue-type plasminogen activator mRNA. *Genes Dev* **1**: 1201–1211.
- Huynh J-R., Munro TP, Smith-Litière K, Lepesant J-A, St Johnston D. 2004. The *Drosophila* hnRNP A/B homolog, Hrp48, is specifically required for a distinct step in *osk* mRNA localization. *Dev Cell* **6**: 625–635.
- Johnstone O, Lasko P. 2004. Interaction with eIF5B is essential for Vasa function during development. *Development* **131**: 4167–4178.
- Juge F, Zaessinger S, Temme C, Wahle E, Simonelig M. 2002. Control of poly(A) polymerase level is essential to cytoplasmic polyadenylation and early development in *Drosophila*. *EMBO J* **21**: 6603–6613.
- Jung MY, Lorenz L, Richter JD. 2006. Translational control by neuroguidin, a eukaryotic initiation factor 4E and CPEB binding protein. *Mol Cell Biol* **26**: 4277–4287.
- Kalifa Y, Huang T, Rosen LN, Chatterjee S, Gavis ER. 2006. Glorund, a *Drosophila* hnRNP F/H homolog, is an ovarian repressor of *nanos* translation. *Dev Cell* **10**: 291–301.
- Kalifa Y, Armenti ST, Gavis ER. 2009. Glorund interactions in the regulation of *gurken* and *oskar* mRNAs. *Dev Biol* **326**: 68–74.
- Karaiskou A, Perez LH, Ferby I, Ozon R, Jessus C, Nebreda AR. 2001. Differential regulation of Cdc2 and Cdk2 by RINGO and cyclins. *J Biol Chem* **276**: 36028–36034.
- Keady BT, Kuo P, Martínez SE, Yuan L, Hake LE. 2007. MAPK interacts with XGef and is required for CPEB activation during meiosis in *Xenopus* oocytes. *J Cell Sci* **120**: 1093–1103.
- Kim JH, Richter JD. 2006. Opposing polymerase-deadenylase activities regulate cytoplasmic polyadenylation. *Mol Cell* **24**: 173–183.
- Kim JH, Richter JD. 2007. RINGO/cdk1 and CPEB mediate poly(A) tail stabilization and translational regulation by ePAB. *Genes Dev* **21**: 2571–2579.
- Kim-Ha J, Kerr K, Macdonald PM. 1995. Translational regulation of *oskar* mRNA by Bruno, an ovarian RNA-binding protein, is essential. *Cell* **81**: 403–412.
- Körner CG, Wormington M, Muckenthaler M, Schneider S, Dehlin E, Wahle E. 1998. The deadenylating nuclease (DAN) is involved in poly(A) tail removal during the meiotic maturation of *Xenopus* oocytes. *EMBO J* **17**: 5427–5437.
- Lie YS, Macdonald PM. 1999. Translational regulation of *oskar* mRNA occurs independent of the cap and poly(A) tail in *Drosophila* ovarian extracts. *Development* **126**: 4989–4996.
- Lim AK, Kai T. 2007. Unique germ-line organelle, nuage, functions to repress selfish genetic elements in *Drosophila melanogaster*. *Proc Natl Acad Sci* **104**: 6714–6719.
- Lin CL, Evans V, Shen S, Xing Y, Richter JD. 2010. The nuclear experience of CPEB: implications for RNA processing and translational control. *RNA* in press.
- Liu N, Han H, Lasko P. 2009. Vasa promotes *Drosophila* germ line stem cell differentiation by activating *mei-P26* translation through a direct interaction with a U-rich motif in its 3' UTR. *Genes Dev* **23**: 2742–2752.
- Lyon AM, Reveal BS, Macdonald PM, Hoffman DW. 2009. Bruno protein contains an expanded RNA recognition motif. *Biochemistry* **48**: 12202–12212.
- MacNicol AM, Wilczynska A, MacNicol MC. 2008. Function and regulation of the mammalian Musashi mRNA translational regulator. *Biochem Soc Trans* **36**: 528–530.
- Mandel CR, Kaneko S, Zhang H, Gebauer D, Vethantham V, Manley JL, Tong L. 2006. Polyadenylation factor CPSF-73 is the pre-mRNA 3'-end-processing endonuclease. *Nature* **444**: 953–956.
- Markussen F-H, Michon AM, Breitwieser W, Ephrussi A. 1995. Translational control of *oskar* generates Short OSK, the isoform that induces pole plasma assembly. *Development* **121**: 3723–3732.
- Martínez SE, Yuan L, Lacza C, Ransom H, Mahon GM, Whitehead IP, Hake LE. 2005. XGef mediates early CPEB phosphorylation during *Xenopus* oocyte meiotic maturation. *Mol Biol Cell* **16**: 1152–1164.
- McGrew LL, Dworkin-Rastl E, Dworkin MB, Richter JD. 1989. Poly(A) elongation during *Xenopus* oocyte maturation is required for translational recruitment and is mediated by a short sequence element. *Genes Dev* **3**: 803–815.
- Mendez R, Hake LE, Andresson T, Littlepage LE, Ruderman JV, Richter JD. 2000a. Phosphorylation of CPE binding factor by Eg2 regulates translation of *c-mos* mRNA. *Nature* **404**: 302–307.
- Mendez R, Murthy KG, Ryan K, Manley JL, Richter JD. 2000b. Phosphorylation of CPEB by Eg2 mediates the recruitment of CPSF into an active cytoplasmic polyadenylation complex. *Mol Cell* **6**: 1253–1259.
- Mendez R, Barnard D, Richter JD. 2002. Differential mRNA translation and meiotic progression require Cdc2-mediated CPEB destruction. *EMBO J* **21**: 1833–1844.
- Micklem DR, Adams J, Grünert S, St Johnston D. 2000. Distinct roles of two conserved Staufen domains in *oskar* mRNA localization and translation. *EMBO J* **19**: 1366–1377.
- Minshall N, Thom G, Standart N. 2001. A conserved role of a DEAD box helicase in mRNA masking. *RNA* **7**: 1728–1742.
- Minshall N, Reiter MH, Weil D, Standart N. 2007. CPEB interacts with an ovary-specific eIF4E and 4E-T in early *Xenopus* oocytes. *J Biol Chem* **282**: 37389–37401.
- Minshall N, Kress M, Weil D, Standart N. 2009. Role of p54 RNA helicase activity and its C-terminal domain in translational repression, P-body localization and assembly. *Mol Biol Cell* **20**: 2464–2472.
- Nakahata S, Katsu Y, Mita K, Inoue K, Nagahama Y, Yamashita M. 2001. Biochemical identification of *Xenopus* Pumilio as a sequence-specific cyclin B1 mRNA-binding protein that physically interacts with a Nanos homolog, Xcat-2, and a cytoplasmic polyadenylation element-binding protein. *J Biol Chem* **276**: 20945–20953.
- Nakahata S, Kotani T, Mita K, Kawasaki T, Katsu Y, Nagahama Y, Yamashita M. 2003. Involvement of *Xenopus* Pumilio in the translational regulation that is specific to cyclin B1 mRNA during oocyte maturation. *Mech Dev* **120**: 865–880.
- Nakamura A, Amikura R, Hanyu K, Kobayashi S. 2001. Me31B silences translation of oocyte-localizing RNAs



- through the formation of cytoplasmic RNP complex during *Drosophila* oogenesis. *Development* **128**: 3233–3242.
- Nakamura A, Sato K, Hanyu-Nakamura K. 2004. *Drosophila* Cup is an eIF4E binding protein that associates with Bruno and regulates *oskar* mRNA translation in oogenesis. *Dev Cell* **6**: 69–78.
- Napoli I, Mercaldo V, Boyl P, Eleuteri B, Zalfa F, De Rubeis S, Di Marino D, Mohr E, Massimi M, Falconi M, et al. 2008. The fragile X syndrome protein represses activity-dependent translation through CYFIP1, a new 4E-BP. *Cell* **134**: 1042–1054.
- Nebreda AR, Ferby I. 2000. Regulation of the meiotic cell cycle in oocytes. *Curr Opin Cell Biol* **12**: 666–675.
- Nelson MR, Leidal AM, Smibert CA. 2004. *Drosophila* Cup is an eIF4E-binding protein that functions in Smaug-mediated translational repression. *EMBO J* **23**: 150–159.
- Niessing D, Dostatni N, Jäckle H, Rivera-Pomar R. 1999. Sequence interval within the PEST motif of Bicoid is important for translational repression of *caudal* mRNA in the anterior region of the *Drosophila* embryo. *EMBO J* **18**: 1966–1973.
- Niessing D, Blanke S, Jäckle H. 2002. Bicoid associates with the 5' cap-bound complex of *caudal* mRNA and represses translation. *Genes Dev* **16**: 2576–2582.
- Norvell AKR, Wehr K, Schüpbach T. 1999. Specific isoforms of Squid, a *Drosophila* hnRNP, perform distinct roles in Gurken localization during oogenesis. *Genes Dev* **13**: 864–876.
- Norvell A, Debec A, Finch D, Gibson L, Thoma B. 2005. Squid is required for efficient posterior localization of *oskar* mRNA during *Drosophila* oogenesis. *Dev Genes Evol* **215**: 340–349.
- Ostareck DH, Ostareck-Lederer A, Shatsky IN, Hentze MW. 2003. Lipoygenase mRNA silencing in erythroid differentiation: The 3' UTR regulatory complex controls 60S ribosomal subunit joining. *Cell* **104**: 281–290.
- Padmanabhan K, Richter JD. 2006. Regulated Pumilio-2 binding controls RINGO/Spy mRNA translation and CPEB activation. *Genes Dev* **20**: 199–209.
- Pane A, Wehr K, Schüpbach T. 2007. *zucchini* and *squash* encode two putative nucleases required for rasiRNA production in the *Drosophila* germline. *Dev Cell* **12**: 851–862.
- Piqué M, López JM, Foissac S, Guigó R, Méndez R. 2008. A combinatorial code for CPE-mediated translational control. *Cell* **132**: 434–448.
- Rangan P, DeGennaro M, Jamie-Bustamante K, Coux RX, Martinho RG, Lehmann R. 2009. Temporal and spatial control of germ-plasm RNAs. *Curr Biol* **19**: 72–77.
- Read RL, Martinho RG, Wang SW, Carr AM, Norbury CJ. 2002. Cytoplasmic poly(A) polymerases mediate cellular responses to S phase arrest. *Proc Natl Acad Sci* **99**: 12079–12084.
- Reverte CG, Ahearn MD, Hake LE. 2001. CPEB degradation during *Xenopus* oocyte maturation requires a PEST domain and the 26S proteasome. *Dev Biol* **231**: 447–458.
- Reverte CG, Yuan L, Keady BT, Lacza C, Attfield KR, Mahon GM, Freeman B, Whitehead IP, Hake LE. 2003. XGef is a CPEB-interacting protein involved in *Xenopus* oocyte maturation. *Dev Biol* **255**: 383–398.
- Richard P, Manley JL. 2009. Transcription termination by nuclear RNA polymerases. *Genes Dev* **23**: 1247–1269.
- Richter JD. 1999. Cytoplasmic polyadenylation in development and beyond. *Microbiol Mol Biol Rev* **63**: 446–456.
- Rivera-Pomar R, Niessing D, Schmidt-Ott U, Gehring WJ, Jäckle H. 1996. RNA binding and translational suppression by Bicoid. *Nature* **379**: 746–749.
- Saffman EE, Styhler S, Rother K, Li W, Richard S, Lasko P. 1998. Premature translation of *oskar* in oocytes lacking the RNA-binding protein Bicaudal-C. *Mol Cell Biol* **18**: 4855–4862.
- Saitoh S, Chabes A, McDonald WH, Thelander L, Yates JR, Russell P. 2002. Cid13 is a cytoplasmic poly(A) polymerase that regulates ribonucleotide reductase mRNA. *Cell* **109**: 563–573.
- Sallés FJ, Lieberfarb ME, Wreden C, Gergen JP, Strickland S. 1994. Coordinate initiation of *Drosophila* development by regulated polyadenylation of maternal messenger RNAs. *Science* **266**: 1996–1999.
- Sarkissian M, Mendez R, Richter JD. 2004. Progesterone and insulin stimulation of CPEB-dependent polyadenylation is regulated by Aurora A and glycogen synthase kinase-3. *Genes Dev* **18**: 48–61.
- Saunders C, Cohen RS. 1999. The role of oocyte transcription, the 5' UTR, and translation repression and derepression in *Drosophila gurken* mRNA and protein localization. *Mol Cell* **3**: 43–54.
- Semotok JL, Cooperstock RL, Pinder BD, Vari HK, Lipshitz HD, Smibert CA. 2005. Smaug recruits the CCR4/POP2/NOT deadenylase complex to trigger maternal transcript localization in the early *Drosophila* embryo. *Curr Biol* **15**: 284–294.
- Shatkin AJ, Manley JL. 2000. The ends of the affair: Capping and polyadenylation. *Nature Struct Mol Biol* **7**: 838–842.
- Sheets MD, Fox CA, Hunt T, Vande Woude G, Wickens M. 1994. The 3'-untranslated regions of c-mos and cyclin mRNAs stimulate translation by regulating cytoplasmic polyadenylation. *Genes Dev* **8**: 926–938.
- Smibert CA, Wilson JE, Kerr R, Macdonald PM. 1996. Smaug protein represses translation of unlocalized *nanos* mRNA in the *Drosophila* embryo. *Genes Dev* **10**: 2600–2609.
- Smith RC, Dworkin-Rastl E, Dworkin MB. 1988a. Expression of a histone H1-like protein is restricted to early *Xenopus* development. *Genes Dev* **2**: 1284–1295.
- Smith RC, Dworkin MB, Dworkin-Rastl E. 1988b. Destruction of a translationally controlled mRNA in *Xenopus* oocytes delays progesterone-induced maturation. *Genes Dev* **2**: 1296–1306.
- Snee M, Benz D, Jen J, Macdonald PM. 2008. Two distinct domains of Bruno bind specifically to the *oskar* mRNA. *RNA Biol* **5**: 1–9.
- Sonoda J, Wharton RP. 2001. *Drosophila* Brain Tumor is a translational repressor. *Genes Dev* **15**: 762–773.
- Spradling AC. 1993. Developmental genetics of oogenesis. In *The development of Drosophila melanogaster* (ed. Bate M, Martinez-Arias A), pp. 1–70. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.
- Stebbins-Boaz B, Richter JD. 1994. Multiple sequence elements and a maternal mRNA product control cdk2 RNA

J.D. Richter and P. Lasko

- polyadenylation and translation during early *Xenopus* development. *Mol Cell Biol* **14**: 5870–5880.
- Stebbins-Boaz B, Hake LE, Richter JD. 1996. CPEB controls the cytoplasmic polyadenylation of cyclin, Cdk2 and c-mos mRNAs and is necessary for oocyte maturation in *Xenopus*. *EMBO J* **15**: 2582–2592.
- Stebbins-Boaz B, Cao Q, de Moor CH, Mendez R, Richter JD. 1999. Maskin is a CPEB-associated factor that transiently interacts with eIF-4E. *Mol Cell* **4**: 1017–1027.
- Strickland S, Huarte J, Belin D, Vassalli A, Rickles RJ, Vassalli JD. 1988. Antisense RNA directed against the 3' noncoding region prevents dormant mRNA activation in mouse oocytes. *Science* **241**: 680–684.
- Styler S, Nakamura A, Swan A, Suter B, Lasko P. 1998. *vasa* is required for GURKEN accumulation in the oocyte, and is involved in oocyte differentiation and germline cyst development. *Development* **125**: 1569–1578.
- Tadros W, Goldman AL, Babak T, Menzies F, Vardy L, Orr-Weaver T, Hughes TR, Westwood JT, Smibert CA, Lipshitz HD. 2007. SMAUG is a major regulator of maternal mRNA destabilization in *Drosophila* and its translation is activated by the PAN GU kinase. *Dev Cell* **12**: 143–155.
- Tay J, Richter JD. 2001. Germ cell differentiation and synaptonemal complex formation are disrupted in CPEB knockout mice. *Dev Cell* **1**: 201–213.
- Tomancak P, Guichet A, Zavorszky P, Ephrussi A. 1998. Oocyte polarity depends on regulation of *gurken* by *Vasa*. *Development* **125**: 1723–1732.
- Tomari Y, Du T, Haley B, Schwarz DS, Bennett R, Cook HA, Koppetsch BS, Theurkauf WE, Zamore PD. 2004. RISC assembly defects in the *Drosophila* RNAi mutant *armi-tag*. *Cell* **119**: 831–841.
- Trucco A, Gaspar I, Ephrussi A. 2009. Assembly of endogenous *oskar* mRNA particles for motor-dependent transport in the *Drosophila* oocyte. *Cell* **139**: 983–998.
- Tung JJ, Padmanabhan K, Hansen DV, Richter JD, Jackson PK. 2007. Translational unmasking of Emi2 directs cytostatic factor arrest in meiosis II. *Cell Cycle* **6**: 725–731.
- Tunquist BJ, Maller JL. 2003. Under arrest: Cytostatic factor (CSF)-mediated metaphase arrest in vertebrate eggs. *Genes Dev* **17**: 683–710.
- Vanzo N, Oprins A, Xanthakis D, Ephrussi A, Rabouille C. 2007. Stimulation of endocytosis and actin dynamics by Oskar polarizes the *Drosophila* oocyte. *Dev Cell* **12**: 543–555.
- Villaescusa JC, Buratti C, Penkov D, Mathiasen L, Pianagumà J, Ferretti E, Blasi F. 2009. Cytoplasmic Prep1 interacts with 4EHP inhibiting *Hoxb4* translation. *PLoS ONE* **4**: e5213.
- Wang L, Eckmann CR, Kadyk LC, Wickens M, Kimble J. 2002. A regulatory cytoplasmic poly(A) polymerase in *Caenorhabditis elegans*. *Nature* **419**: 312–316.
- Webster PJ, Liang L, Berg CA, Lasko P, Macdonald PM. 1997. Translational repressor Bruno plays multiple roles in development and is widely conserved. *Genes Dev* **11**: 2510–2521.
- Wharton RP, Struhl G. 1991. RNA regulatory elements mediate control of *Drosophila* body pattern by the posterior morphogen *nanos*. *Cell* **67**: 955–967.
- Wilhelm JE, Hilton M, Amos Q, Henzel WJ. 2003. Cup is an eIF4E binding protein required for both the translational repression of *oskar* and the recruitment of Barentsz. *J Cell Biol* **163**: 1197–1204.
- Wilson JE, Connell JE, Macdonald PM. 1996. *Aubergine* enhances *oskar* translation in the *Drosophila* ovary. *Development* **122**: 1631–1639.
- Wilt FH. 1973. Polyadenylation of maternal RNA of sea urchin eggs after fertilization. *Proc Natl Acad Sci* **70**: 2345–2349.
- Wu L, Wells D, Tay J, Mendis D, Abbott MA, Barnitt A, Quinlan E, Heynen A, Fallon JR, Richter JD. 1998. CPEB-mediated cytoplasmic polyadenylation and the regulation of experience-dependent translation of α -CaMKII mRNA at synapses. *Neuron* **21**: 1129–1139.
- Yano T, Lopez de Quinto S, Matsui Y, Shevchenko A, Shevchenko A, Ephrussi A. 2004. Hrp48, a *Drosophila* hnRNP A/B homolog, binds and regulates translation of *oskar* mRNA. *Dev Cell* **6**: 637–648.
- Zaessinger S, Busseau I, Simonelig M. 2006. Oskar allows *nanos* mRNA translation in *Drosophila* embryos by preventing its deadenylation by Smaug/CCR4. *Development* **133**: 4573–4583.
- Zappavigna V, Piccioni F, Villaescusa JC, Verrotti AC. 2004. Cup is a nucleocytoplasmic shuttling protein that interacts with the eukaryotic translation initiation factor 4E to modulate *Drosophila* ovary development. *Proc Natl Acad Sci* **101**: 14800–14805.



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