Review

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mRNA transport meets membrane traffic

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Active transport and local translation of mRNAs ensure the appropriate spatial organization of proteins within cells. Recent work has shown that this process is intricately connected to membrane trafficking. Here, we focus on new findings obtained in fungal model systems. Important highlights are that RNA-binding proteins recognize cargo mRNA synergistically and that mRNAs are co-transported with membranous compartments such as the endoplasmic reticulum (ER) and endosomes. We further discuss a novel concept of endosome-coupled translation that loads shuttling endosomes with septin cargo, a process important for correct septin filamentation. Interestingly, evidence is accumulating that RNA and membrane trafficking are also tightly interwoven in higher eukaryotes, suggesting that this phenomenon is a common theme and not an exception restricted to fungi.

Active transport of mRNPs and membranous compartments

Active cytoskeletal transport mediated by molecular motors is essential for cellular logistics in eukaryotes and transport defects often cause death or disease. An important class of molecular cargo is mRNA [1,2]. Actively transported mRNAs contain *cis*-acting regions, termed zip codes (see Glossary) or localization elements. These zip codes constitute binding sites for RNA-binding proteins of the transport complex, which themselves interact with adapter proteins and molecular motors such as kinesin, dynein, or myosin [2–4]. Furthermore, these complexes usually contain accessory factors such as helicases, translational repressors, RNA stability factors, or ribosomal proteins [5,6]. Therefore, mRNAs are never naked in the cell but form macromolecular complexes called mRNPs (messenger ribonucleoprotein particles). These mRNPs do not only contain the mRNA information for the encoded amino acid sequence but also determine the precise spatiotemporal regulation of translation and thereby guarantee the correct subcellular localization of the translated protein. Thus, 'the message is the mRNP' [7].

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Comprised in a second class of important molecular cargo are membranous compartments such as endosomes and small vesicles that transport lipids and proteins. The underlying transport events are known as membrane

Glossary

BAR domain (Bin–Amphiphysin–Rvs): this domain was first identified in BIN1, amphiphysins, and the yeast proteins Rvs167p and Rvs161p. It forms a banana-shaped dimer that interacts with curved membranes and thereby functions in membrane shaping.
Calmodulin: evolutionarily conserved regulatory protein that interacts with calcium via a characteristic binding motif called the EF hand. It functions in intracellular calcium signaling by interaction with other signaling proteins such as kinases.
Cortical endoplasmic reticulum (cER): specific type of ER found in plants, yeasts, and some metazoan cell types. cER forms tubular networks juxtaposed to the plasma membrane.
Early endosome: large vesicle-type membranous structure involved in endocytosis. Characteristic marker proteins for this endosomal compartment are the small G proteins Rab4 and Rab5. Early endosomes mature to late endosomes or are involved in rapid recycling towards the plasma membrane.
ELAV-type: specific family of RNA-binding proteins initially identified in *D. melanogaster*. Named after the mutant phenotype Embryonic Lethal Abnormal Vision.
ESCRT complex: a macromolecular machinery designated Endosomal Sorting Complexes Required for Transport. It consists of four subcomplexes called ESCRT-0, ESCRT-I, ESCRT-II, and ESCRT-III. The machinery assembles in an ordered manner and functions in membrane remodeling.
Golgi apparatus: a biological structure named after its discoverer, the Italian physician Camillo Golgi. The Golgi apparatus is a membranous compartment functioning in secretion of proteins. Transport vesicles from the ER deliver cargo to the Golgi complex and after maturation transport vesicles leave the Golgi for fusion with the plasma membrane.
Late endosome: large vesicle-type membranous structure involved in endocytosis. A characteristic marker protein for this compartment is the small G protein Rab7. Late endosomes mature to multivesicular bodies/vacuolar compartments.
Multivesicular body (MVB): specific endosomal compartment that is formed during maturation of late endosomes. MVBs are formed by pinching off vesicles inside the lumen of large endosomes. Similar to late endosomes they carry the small G protein Rab7 as a marker.
Pleckstrin homology domain (PH): domain found in the protein Pleckstrin, a major target of protein kinase C. The 120 amino acid PH domain interacts with phosphatidylinositols and therefore functions in lipid binding and recognition.
RNA-induced silencing complex (RISC): ribonucleoprotein complex that contains small RNAs and the Argonaute protein. It functions in the degradation or translational control of target mRNAs that are recognized by their complementarity to the bound small RNAs.
Rough endoplasmic reticulum (rER): specific type of ER that is covered with translating ribosomes for co-translational import of proteins into its lumen and therefore functioning in active protein secretion.
Signal recognition particle (SRP): ribonucleoprotein complex that recognizes specific signals in secreted and membrane proteins.
Unconventional secretion: conventional secretion in eukaryotes is mediated by the interaction of the signal recognition particles with specific signal sequences at the N terminus of secreted proteins. To differentiate from this classic export pathway the term 'unconventional secretion' is used.
Zip code: RNA sequence that functions as a recognition site for RBPs involved in subcellular mRNA localization.

trafficking [8]. For outbound traffic, secreted proteins or those of the endomembrane system are directly translated at the rough endoplasmic reticulum (rER) where the translation products enter the secretory pathway. Subsequently, the respective proteins are transported by vesicles via the Golgi apparatus to the plasma membrane for secretion or for plasma membrane insertion [9]. During inbound traffic, distinct areas of the plasma membrane pinch off in the form of vesicles that fuse with specific endosomal compartments, called early endosomes. These can mature into late endosomes and fuse with the vacuole or lysosome for protein degradation. Alternatively, they can be redirected to the plasma membrane for receptor recycling [8]. To fulfill their function, compartments involved in membrane trafficking such as vesicles, endosomes, or ER substructures are actively distributed along the cytoskeleton.

In this review, we summarize how these two seemingly independent classes of intracellular transport are tightly linked. We focus on recent results obtained in fungal model systems describing mechanistic insights on the co-transport of mRNPs and ER as well as endosomes.

Assembly of mRNP cargo requires multiple RNA-binding proteins

Localizing mRNPs have been observed throughout all eukaryotic kingdoms. One of the best-studied model systems to investigate mRNP transport is *Saccharomyces cerevisiae*. Core factors of the mRNA trafficking machinery are the myosin motor Myo4p, the adaptor protein She3p, and the RNA-binding protein She2p (Figure 1) [10]. During cell division and polar growth, this so-called SHE machinery transports approximately 30 types of transcripts along actin tracks from the mother cell to the tip of the growing daughter cell, the bud (Box 1) [10,11]. Cargo mRNAs thereby accumulate at the growth pole of daughter cells, resulting in a specific subcellular localization in the bud. Currently, the best-studied example is the asymmetric localization of *ASH1*-containing mRNPs, which facilitates daughter cell specific expression of the transcriptional repressor Ash1p. The resulting selective distribution of Ash1p leads to inhibition of mating type switching specifically in the daughter cell nucleus [10].

The *ASH1* mRNA is already co-transcriptionally recognized by the RNA-binding protein She2p via its four zip code elements, forming an initial nuclear pre-complex [12]. Surprisingly, She2p binds to *ASH1* mRNA with low affinity and specificity *in vitro* [13,14], which is incompatible with processive transport of mRNA in the cell. This seeming paradox was recently resolved by the observation that joining of the nuclear ribosome biogenesis factor Loc1p dramatically stabilizes the complex and improves the specificity of She2p for *ASH1* mRNA [15]. The Loc1p-stabilized complex accumulates in the nucleolus [15], where it is most likely loaded with the translational regulators Puf6p and Khd1p (Figure 1) [10]. During nuclear export, the *ASH1* mRNP is remodeled and Loc1p is

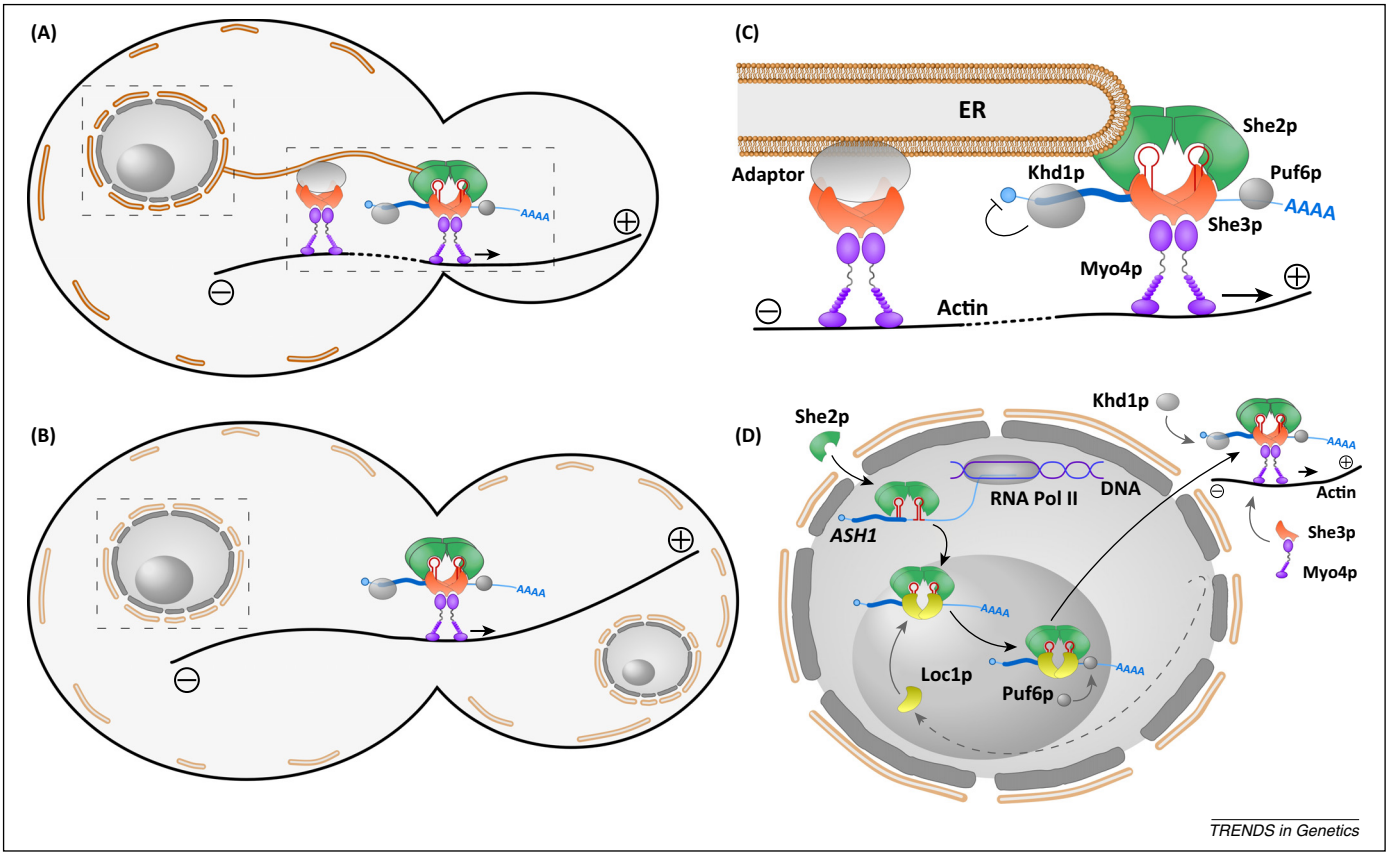

Figure 1. Messenger ribonucleoprotein particle (mRNP) transport in *Saccharomyces cerevisiae*. On the left two different stages of budding cells are depicted. Broken rectangles indicate subcellular regions enlarged on the right. **(A)** Early phase: co-transport of mRNA (blue line with 5′ cap structure as circle and 3′ poly[A] tail) with cortical endoplasmic reticulum (cER; gold) along actin (black line). Note that the membrane adaptor for cER transport by Myo4p/She3p is unknown. **(B)** Late phase: cER-independent transport of *ASH1* mRNA. **(C)** Detailed view of ER/mRNA co-transport. **(D)** Detailed view of nuclear events during She2p-dependent mRNP transport. She2p (green) is loaded co-transcriptionally on the mRNA elements (red hairpin). The nucleolar protein Loc1p (yellow) is replaced during cytoplasmic remodeling by She3p (red).