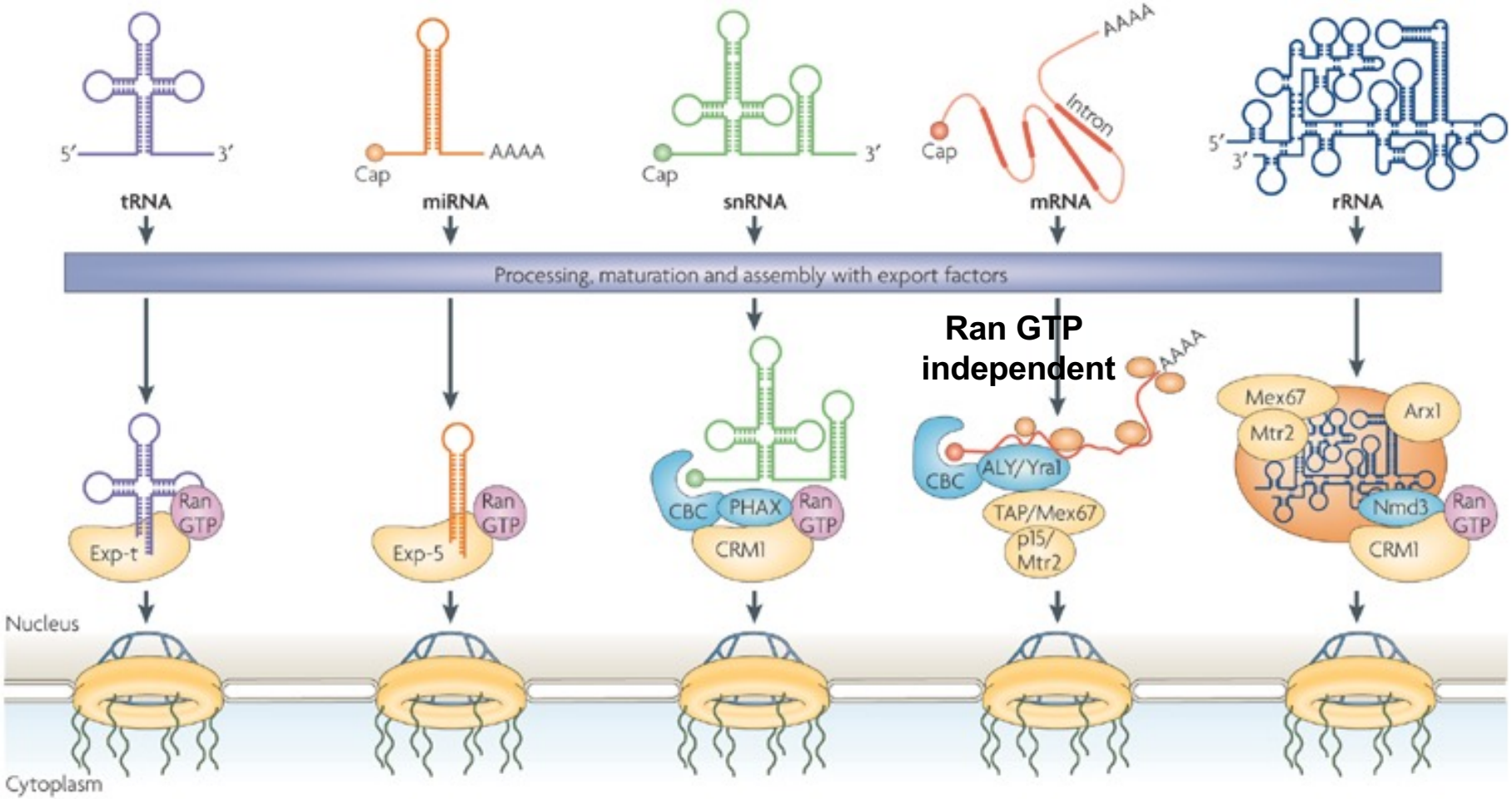
RNA Export

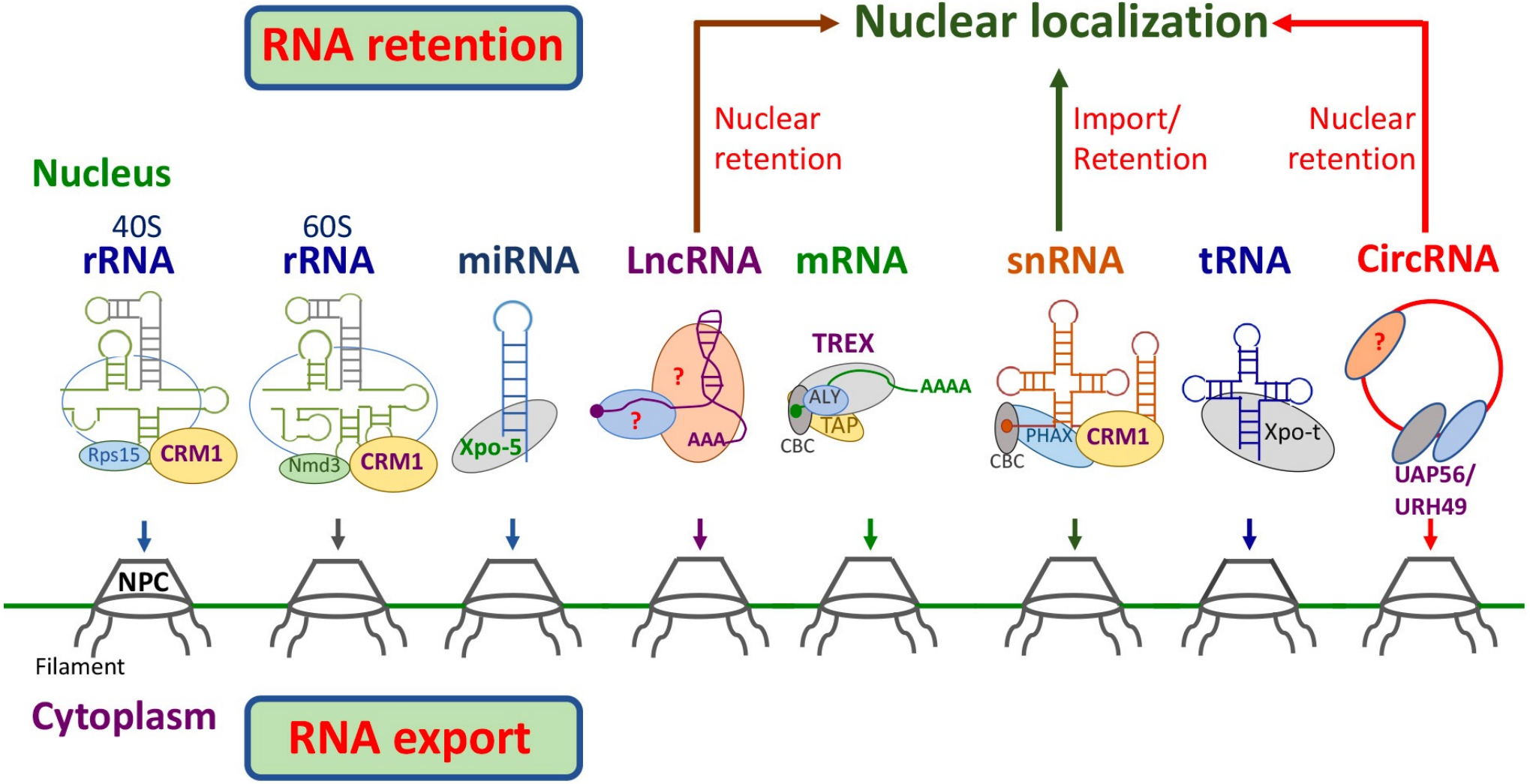
RNA Export

•**Small RNAs** (**tRNAs**, **microRNAs**) follow simple export routes by binding directly to **export receptors**

•**Large RNAs** (**rRNAs**, **mRNAs**) assemble into complicated ribonucleoprotein (RNP) particles and recruit their exporters via class-specific **adaptor proteins.**



Messenger RNAs, tRNAs, rRNAs, snRNAs, most of the lncRNAs, and circular RNAs are exported to the cytoplasm, but some lncRNAs and circular RNAs are retained in the nucleus and snRNAs are re-imported to the nucleus after processing in the cytoplasm

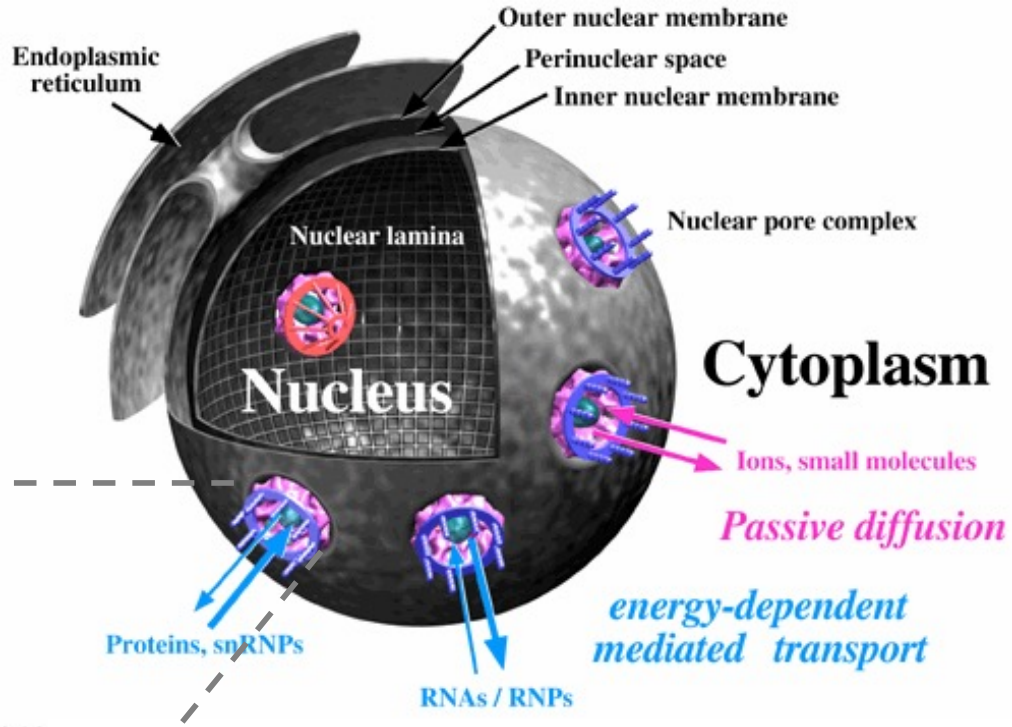


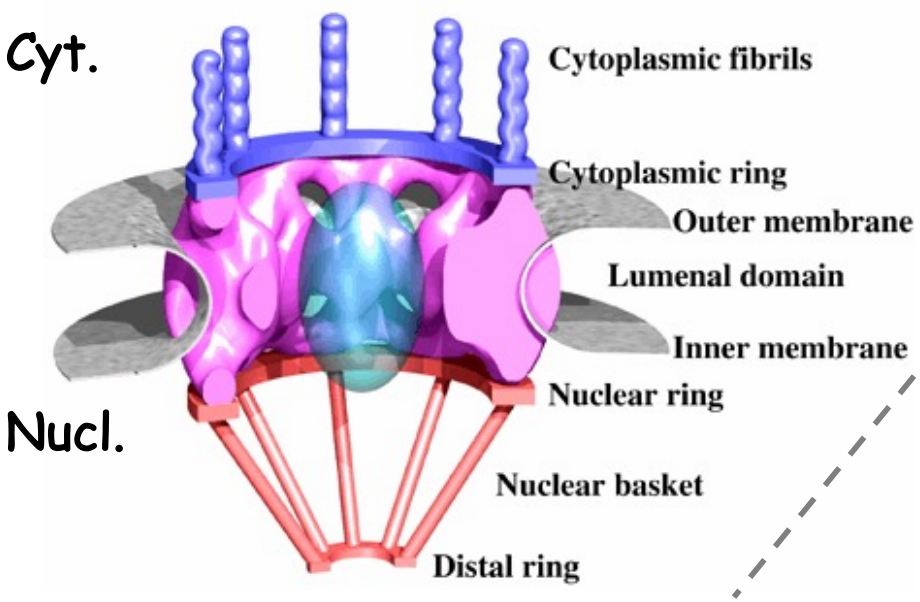
Nuclear Pore Complex (NPC)

•It is a complex (**60 MDa** in yeast and **125 MDa** in metazoa) formed by 30 different **nucleoporins** that exist in 8 or 16 copies per NPC

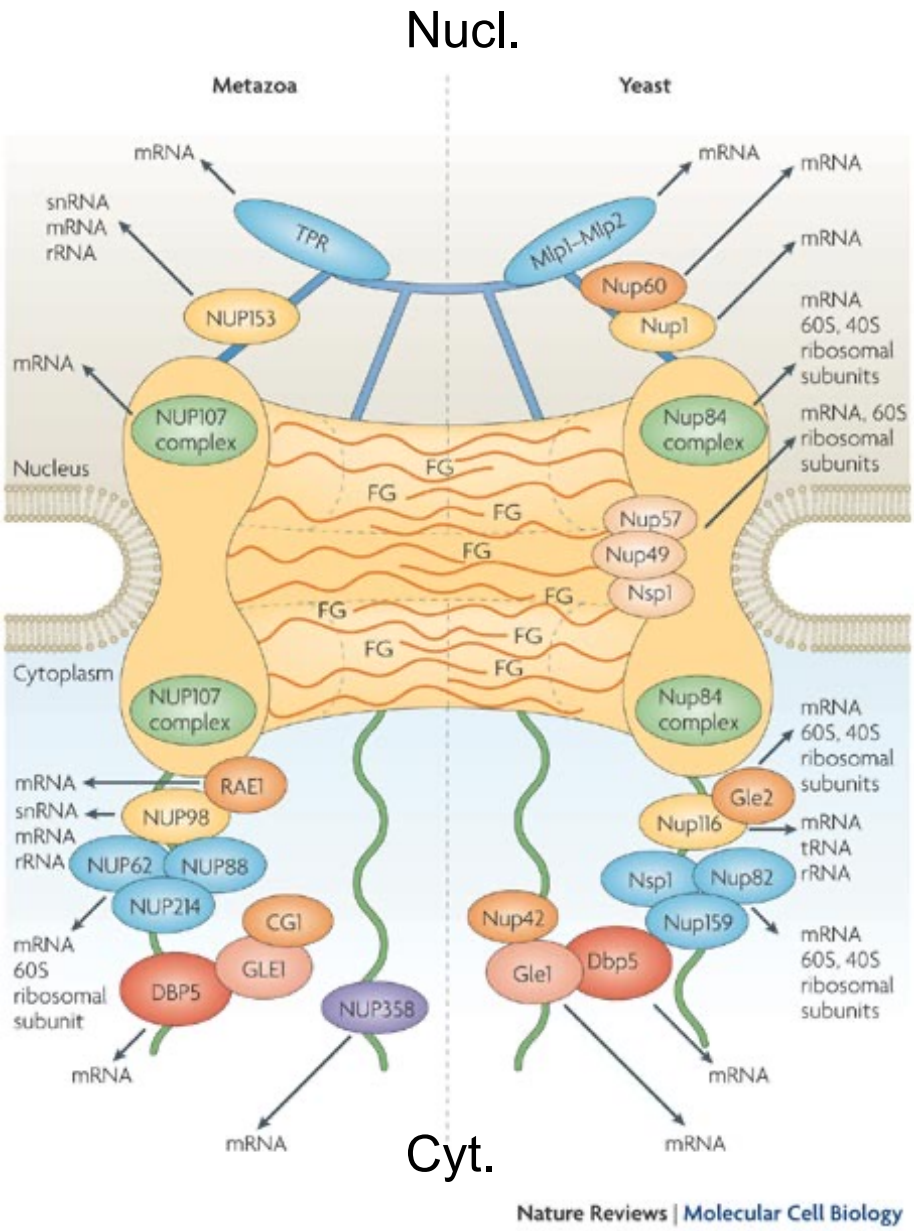
•Ions and small molecules use passive diffusion (<20-40KDa/5 nm diameter)

•Proteins and RNPs use <u>energy-dependent mediated transport</u>





Nucleoporins



•**Nucleoporins** are grouped in three major classes:

1. **FG nucleoporins**: contain Phe-Gly-rich repeat. They are present in the transport channel and mediate the passage of soluble transport receptors, small molecule diffusion, but blocks large molecules.

2. **Nucleoporins devoid of FG-repeat**. These are structural costituent of the NPC that interact with transport receptors.

3. **Nups**. These are integral membrane proteins that anchor the NPC to the membrane

Transport Receptors

•Transport through NPCs requires a family of conserved **transport receptors** (also known as **karyopherins**).

• **karyopherins** recognize a short peptide signal on a cargo protein, either a **nuclear localization signal** (NLS) or a **nuclear export signal** (NES)

•Typically, karyopherins that import cargo are called **importins** and karyopherins that export cargo are called **exportins**

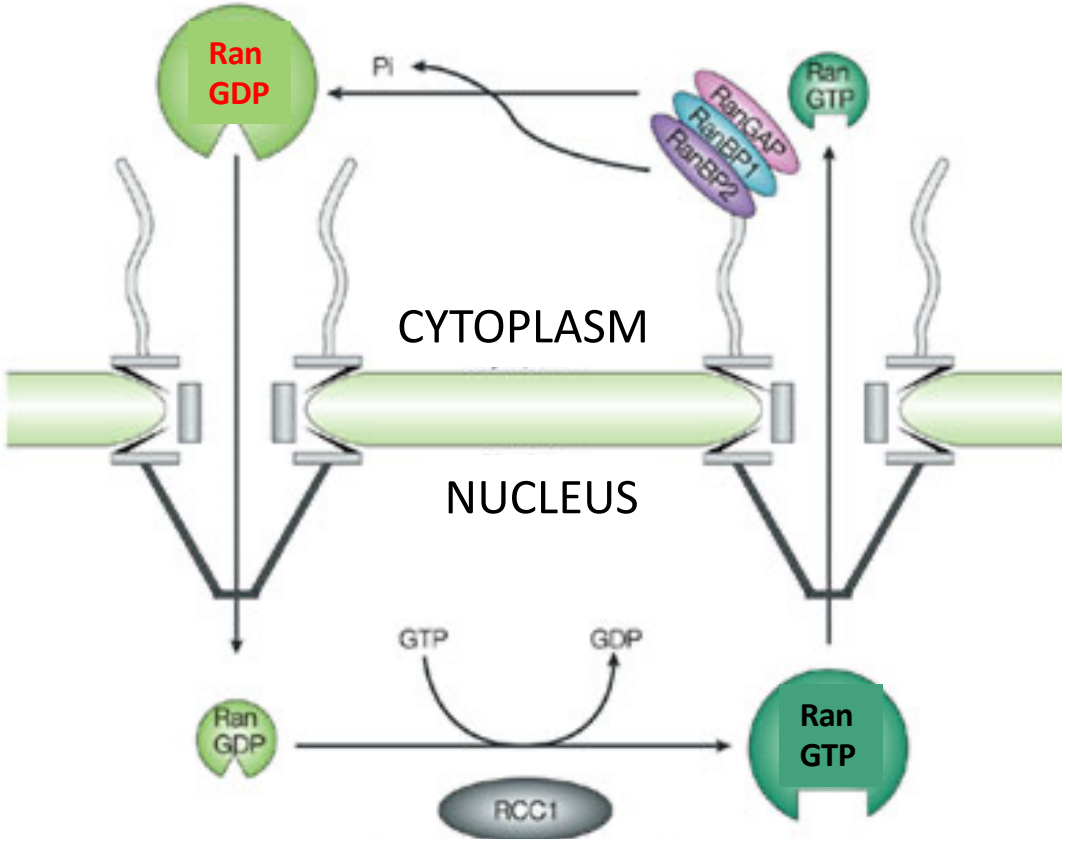
•karyopherins can recognize nucleotide motifs in RNA cargoes, which also enables them to export RNAs.

•A feature of karyopherins is their regulation by the **small GTPase Ran**

Small GTPase RAN

Ran exists in a **GTP-bound** state in the nucleus and a **GDP-bound** state in the cytoplasm.

The RanGTP–RanGDP gradient across the nuclear membrane is generated by the action of two regulators, **RanGEF/RCC1** (Ran-GDP-exchange factor) in the nucleus and **RanGAP** (Ran-GTPase-activating protein) in the cytoplasm, and creates a driving force for directional nucleocytoplasmic transport processes

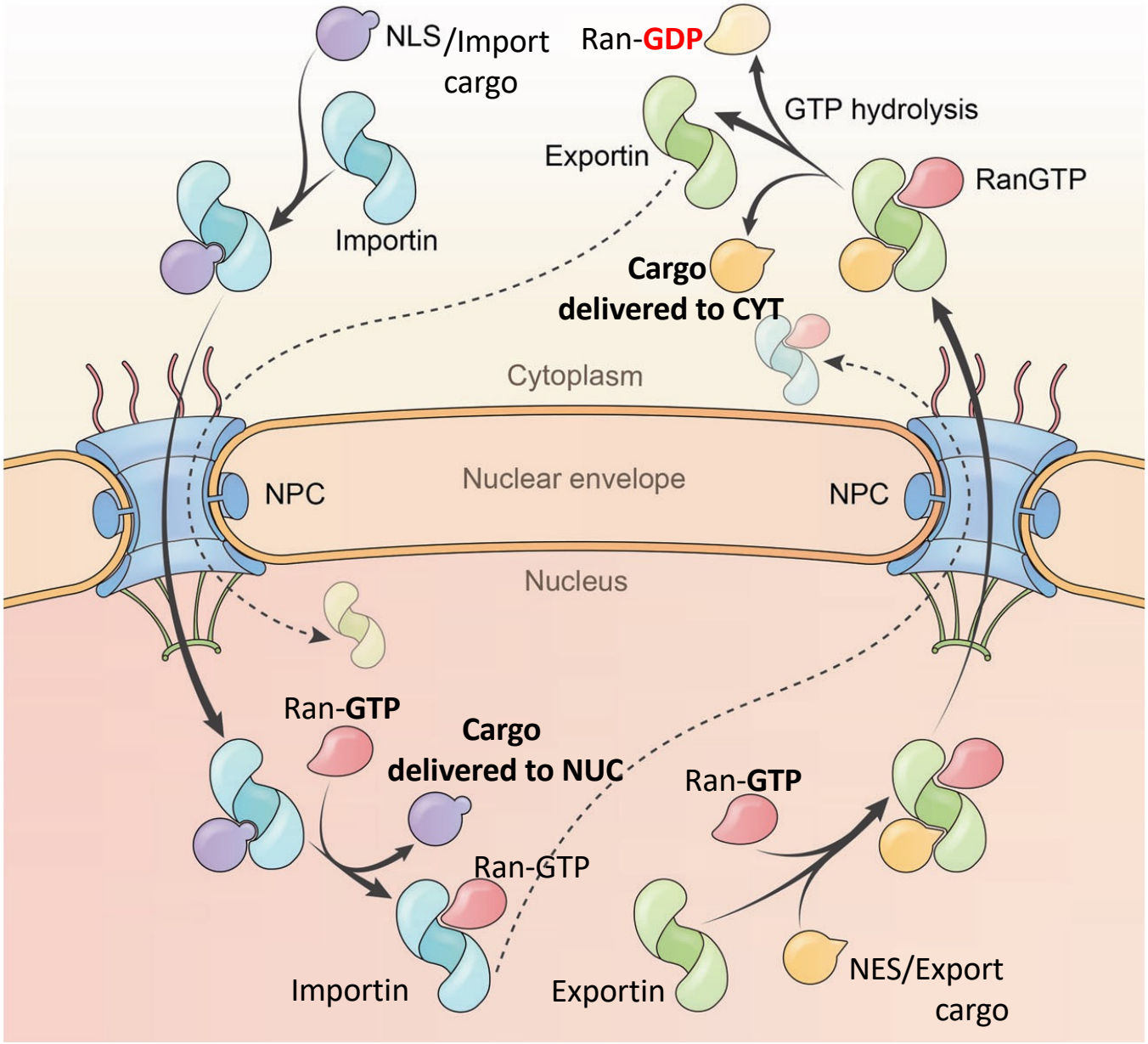


https://www.nature.com/articles/nrm2255

Nuclear import and export cycles through the NPC

•**Importins** bind cargo in the cytoplasm and release it after transport into the nucleus upon binding of RanGTP

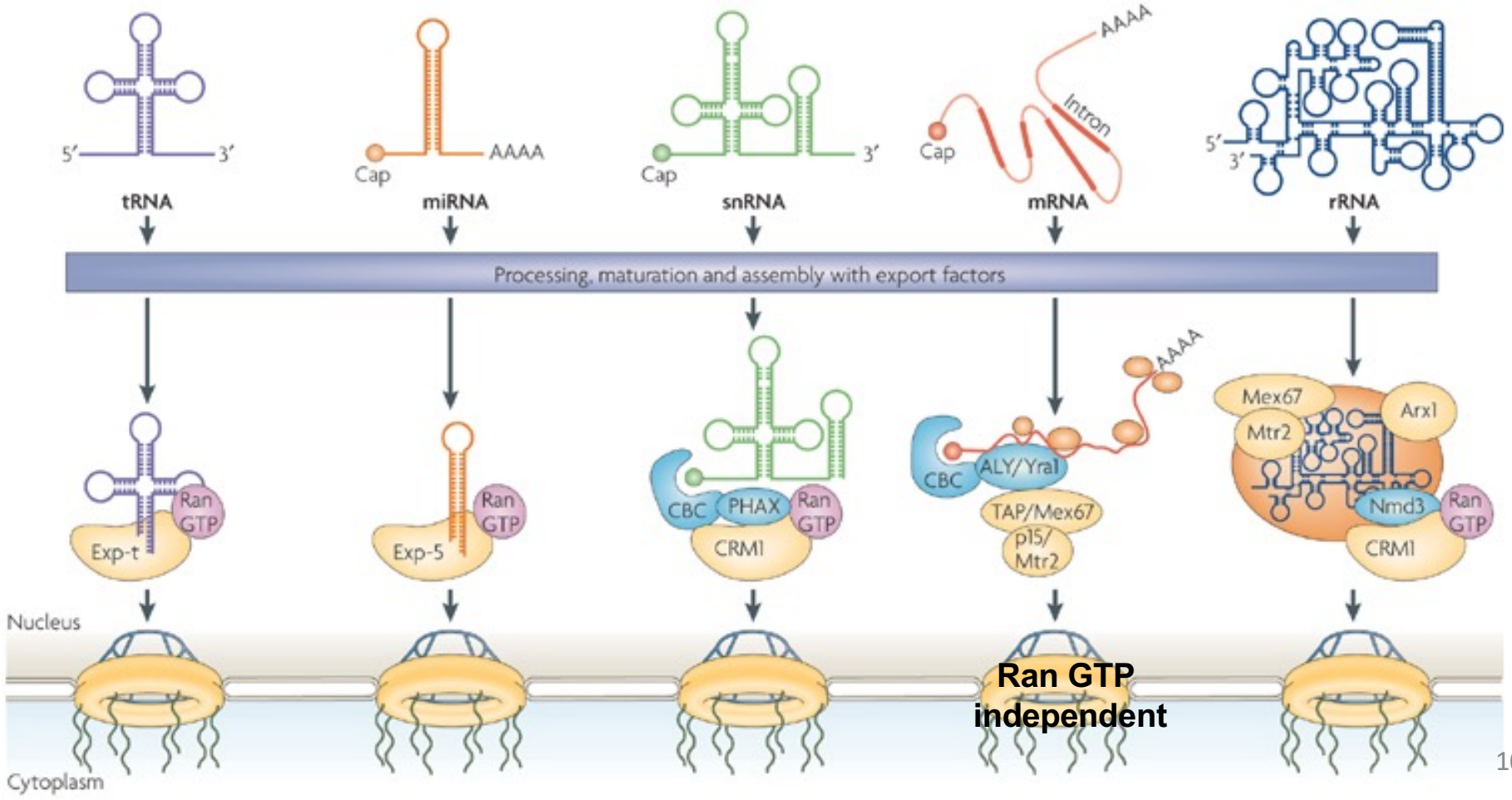
•**Exportins** bind nuclear cargo only together with RanGTP, and this ternary complex is translocated to the cytoplasm, where it dissociates upon hydrolysis of RanGTP by RanGAP.

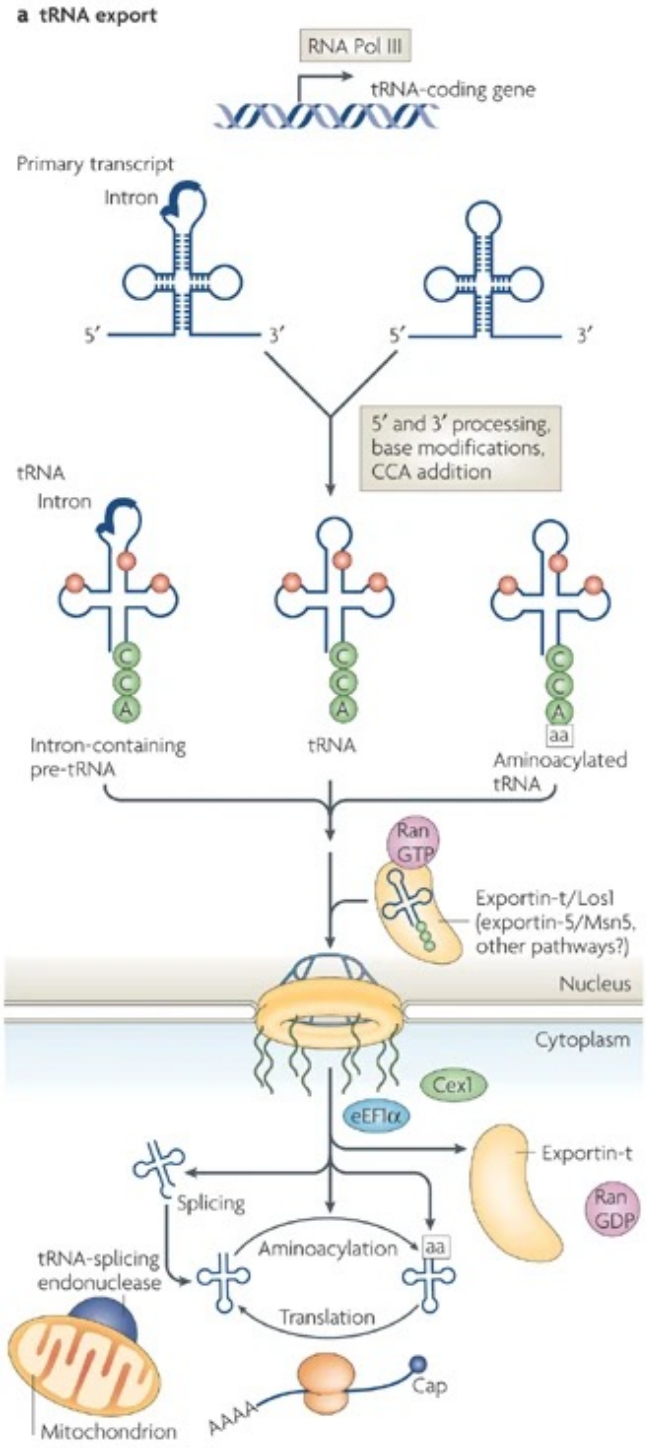


RNA Export

•Small RNAs (tRNAs,microRNAs) follow simple export routes by binding directly to export receptors

•Large RNAs (rRNAs,mRNAs) assemble into complicated ribonucleoprotein (RNP) particles and recruit their exporters via class-specific adaptor proteins.





tRNA EXPORT

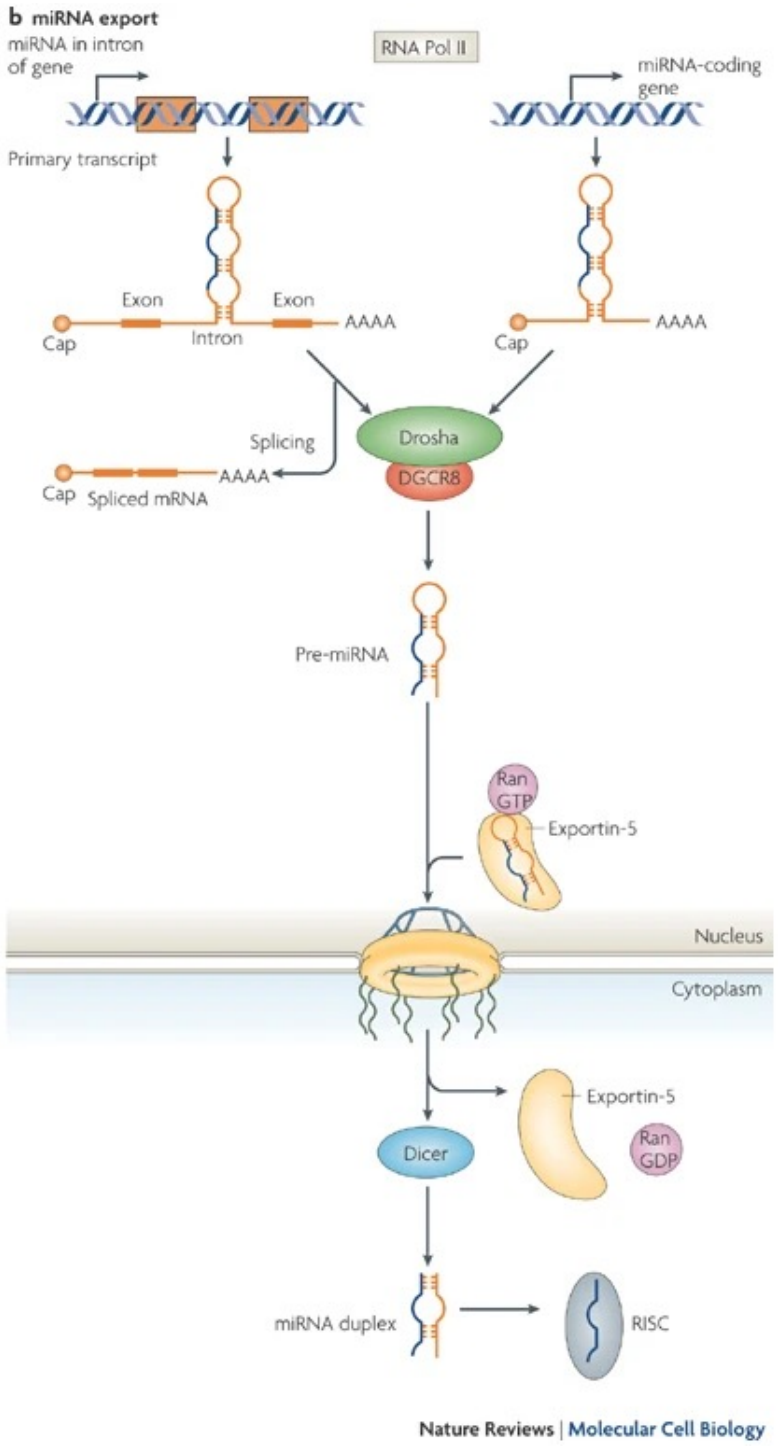
- The class-specific tRNA export receptor **Exportin-t (Los1 in yeast)** binds directly to tRNAs in a RanGTP-dependent manner.Mature tRNAs (containing all base modifications) bind Exp-t 5–10 times more strongly than the unmodified forms

- The NESs in the tRNAs are coded in secondary and tertiary structural elements and properly processed 5' and 3' termini

- After transport of the tRNA–exportin-t–RanGTP complex to the cytoplasm, RanGAP stimulates GTP hydrolysis on Ran, which induces release of the tRNA cargo from its receptor

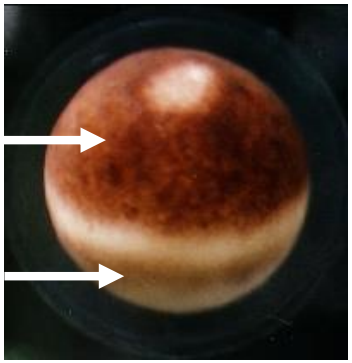
- Exportin-t is the principal tRNA exporter in vertebrate cells, but Exportin-5 is thought to be an auxiliary receptor.

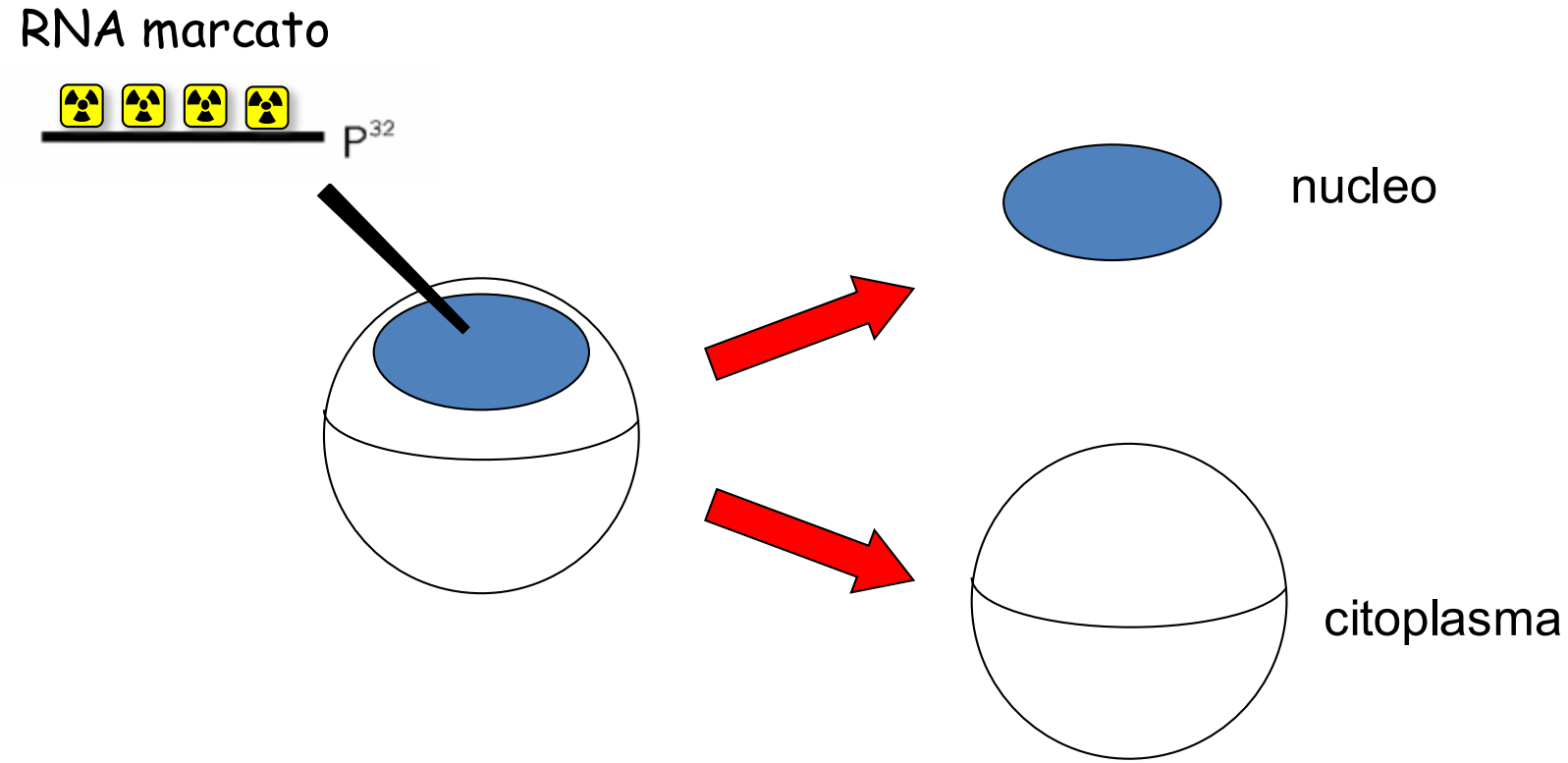
miRNA EXPORT



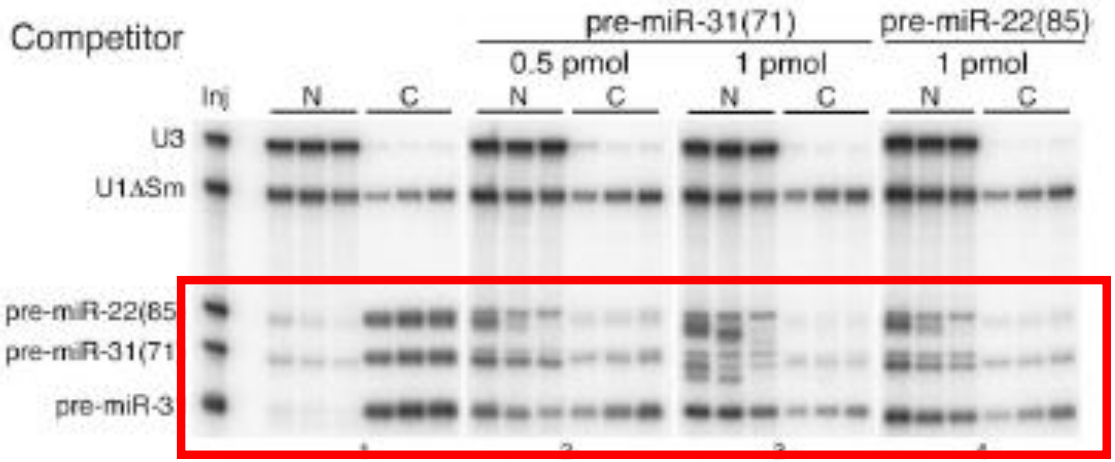
- The ~65-nucleotide **pre-miRNA** is exported to the cytoplasm in a RanGTP-dependent manner by Exportin-5, a member of the karyopherin family.
- During the first maturation step Drosha enzyme generates a double-stranded RNA minihelix with a ~2-nucleotide 3′ overhang, the unique structure of which is recognized both by Exportin-5 and the downstream-acting processing enzyme Dicer in the Cytoplasm.
- After release in the cytoplasm upon GTP hydrolysis on Ran, the pre-miRNA hairpin is released and further cleaved by Dicer.

Oocita di *Xenopus*



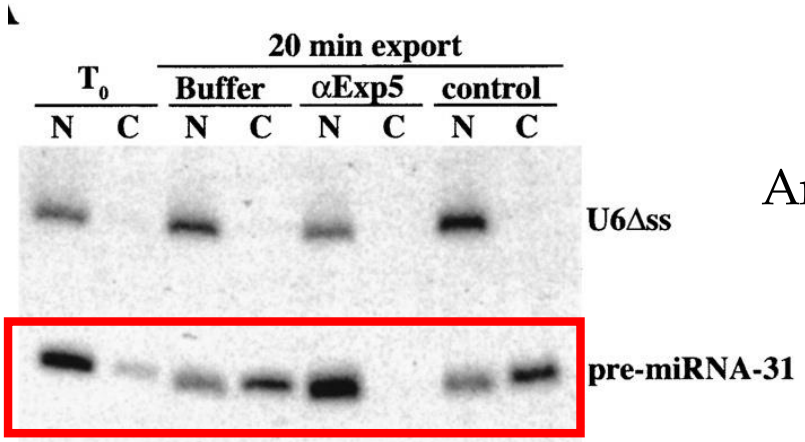


microRNA Export



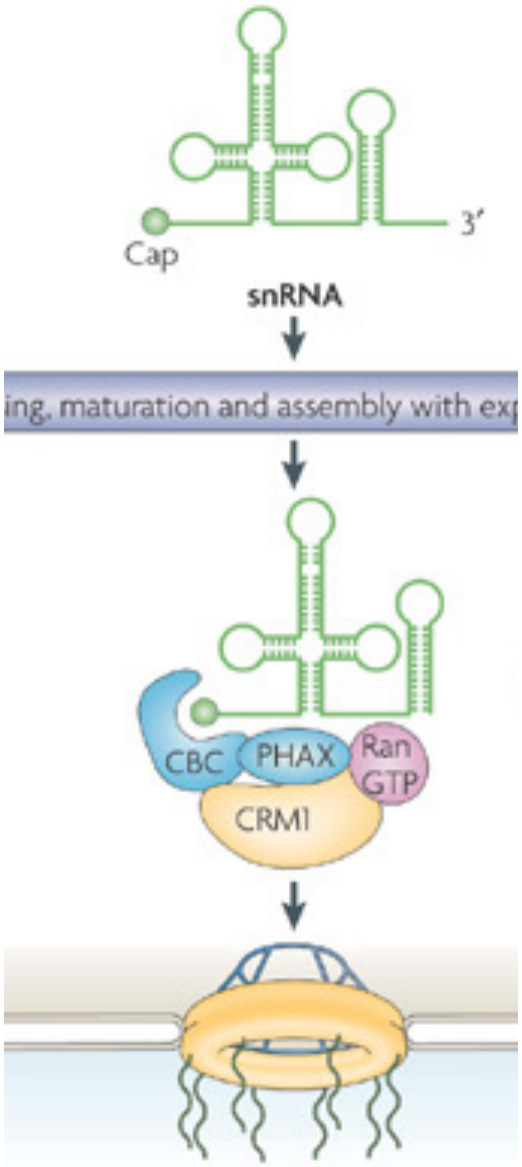
Oocyte microinjection

There is a specific pre-miRNA export receptor



Antibodies against exportin 5 block export of pre-miRNA

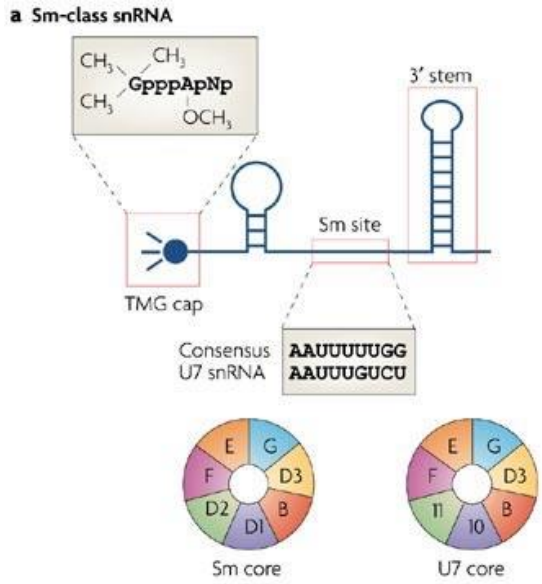
snRNA EXPORT



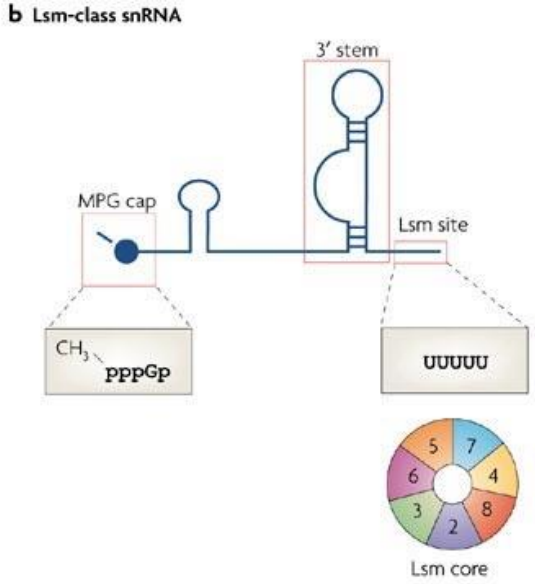
- snRNAs have a nuclear and a cytoplasmic phase. Their maturation is completed in the cytoplasm (except for U6 transcribed by Pol III).
- Their specific exportin is CRM1 (exportin-1) which does not directly interact with the snRNA cargo, but requires the cap-binding complex (CBC) and a NES-containing adaptor protein called PHAX (phosphorylated form) to be targeted to the 5’ cap of the snRNA5
- After export to the cytoplasm, GTP hydrolysis of Ran and dephosphorylation of PHAX are necessary to efficiently dissociate the export complex and release the snRNA

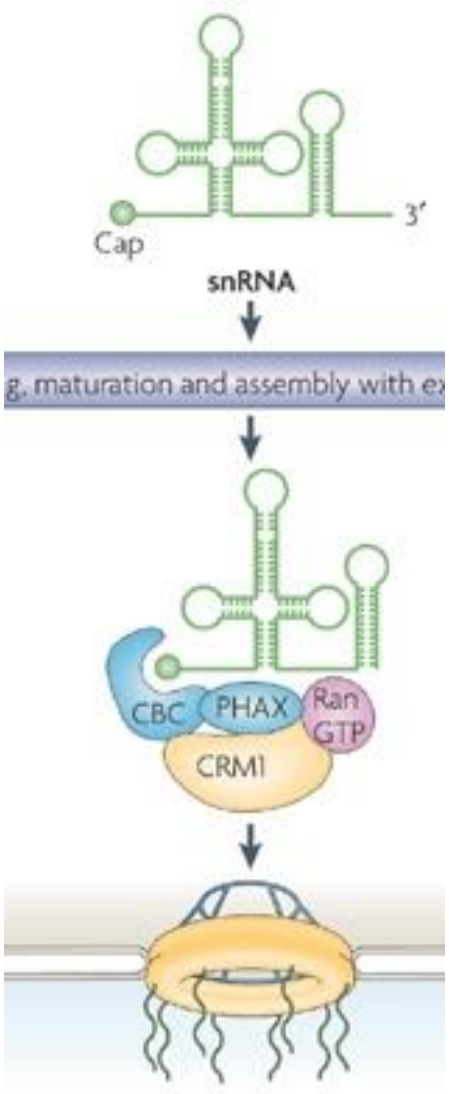
snRNP Biogenesis

U1, U2, U4, U5, U7



U6

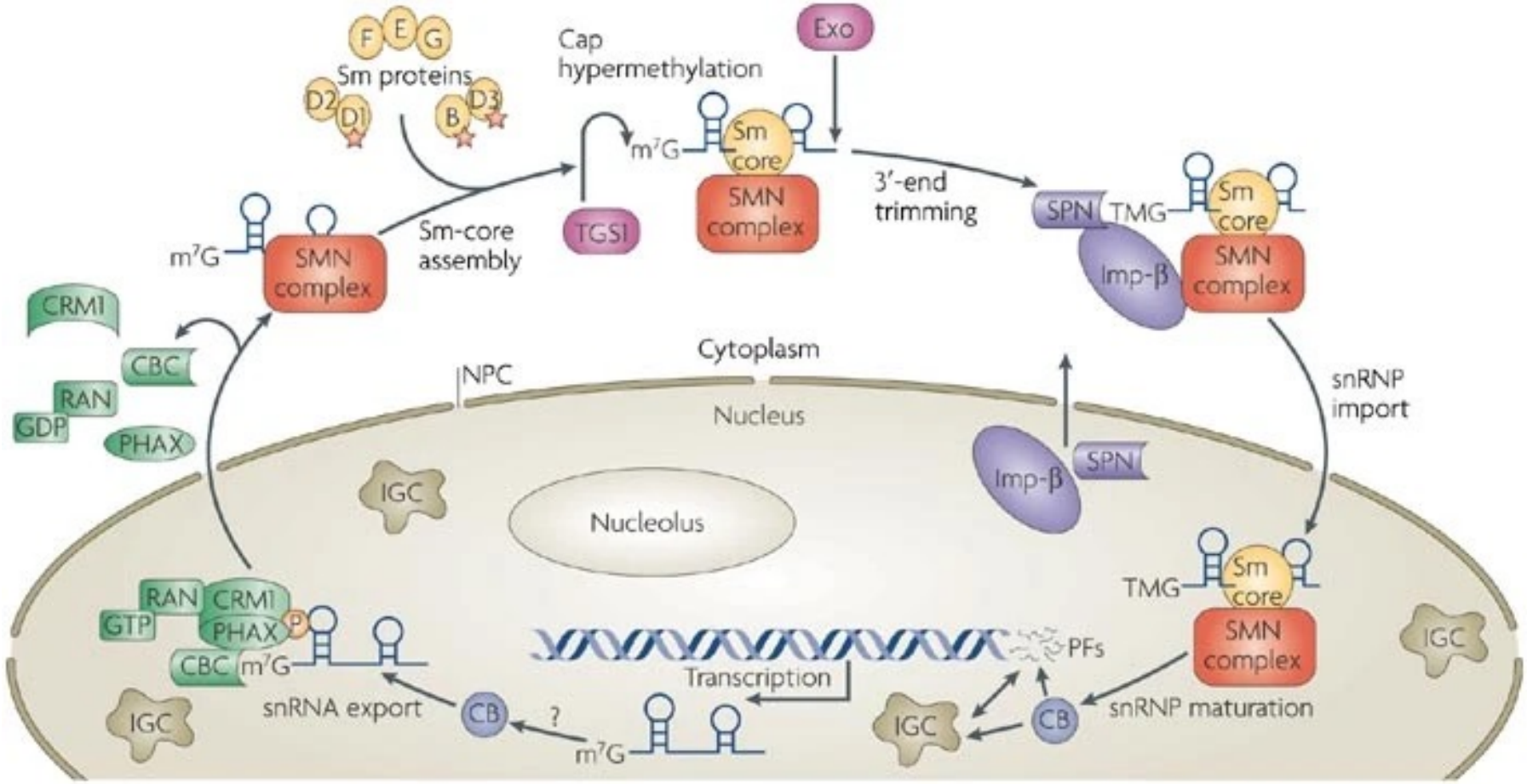




•Their export requires <u>adaptor proteins (**CBC** and **PHAX**)</u> that recruit the export receptor **CRM1**

snRNP Biogenesis

•The life-cycle of Sm-class small nuclear ribonucleoproteins (snRNPs) includes both nuclear and cytoplasmic phases.

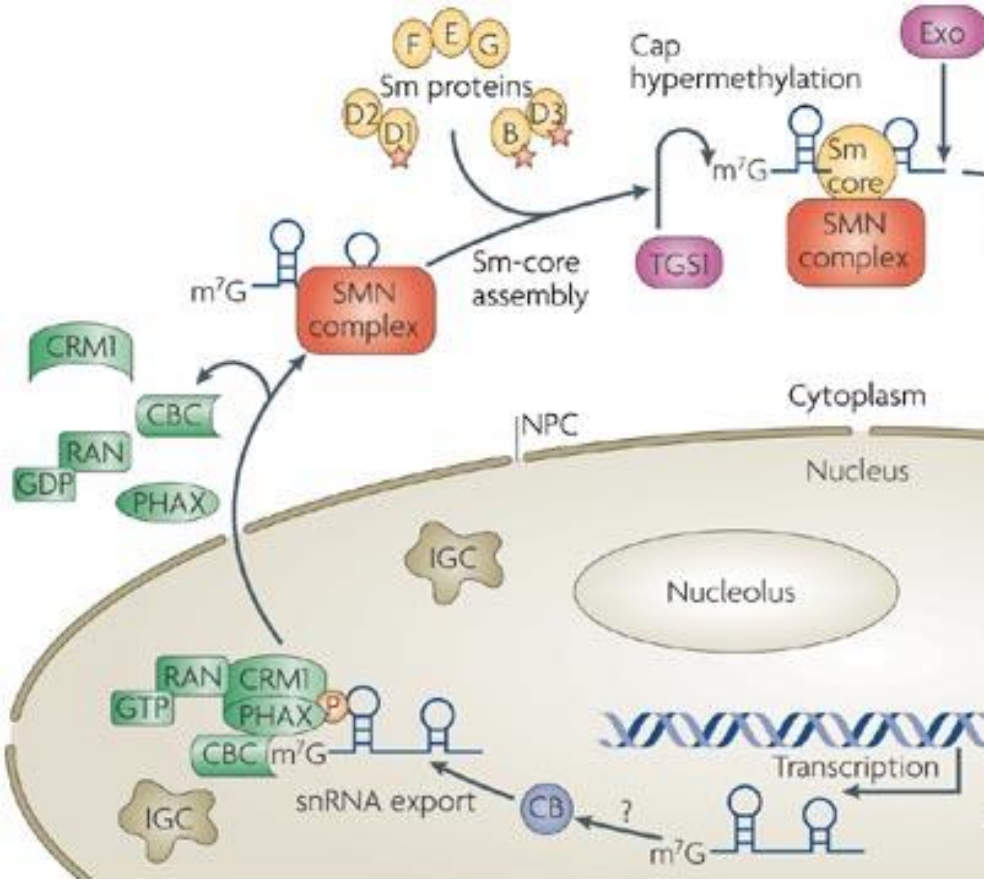


Nature Reviews | Molecular Cell Biology

snRNP Export

•The snRNA-export complex consists of the export receptor **chromosome region maintenance-1** (**CRM1**), the hyperphosphorylated form of the export adaptor **PHAX**, the heterodimeric **cap-binding complex** (**CBC**) and **Ran GTP**

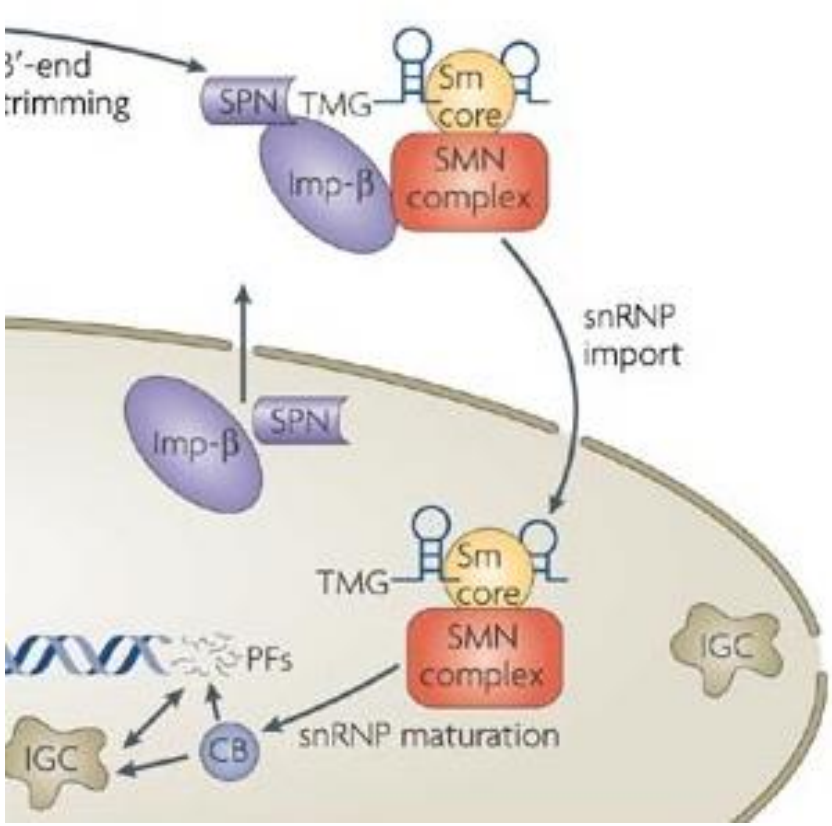
•These factors dissociate from the pre-snRNA in the cytoplasm after binding by the **survival of motor neuron** (**SMN**) complex and dephosphorylation of PHAX



•The SMN complex recognizes specific sequence elements in the snRNAs and recruits a set of seven **Sm proteins**, three of which contain symmetrical dimethylarginine residues (stars), to form the Sm-core RNP. Following assembly of the Sm core, the 7-methylguanosine (m^7G) cap is hypermethylated by **trimethylguanosine synthase-1** (**TGS1**) to form a 2,2,7-trimethylguanosine (**TMG**) cap structure, and the 3' end is trimmed

snRNP Import

•The formation of the TMG cap triggers the assembly of the import complex, which includes the import adaptor **snurportin-1** (**SPN**) and the import receptor **importin-b** (Imp-b). Both SPN and the SMN complex make functional contacts with Imp-b

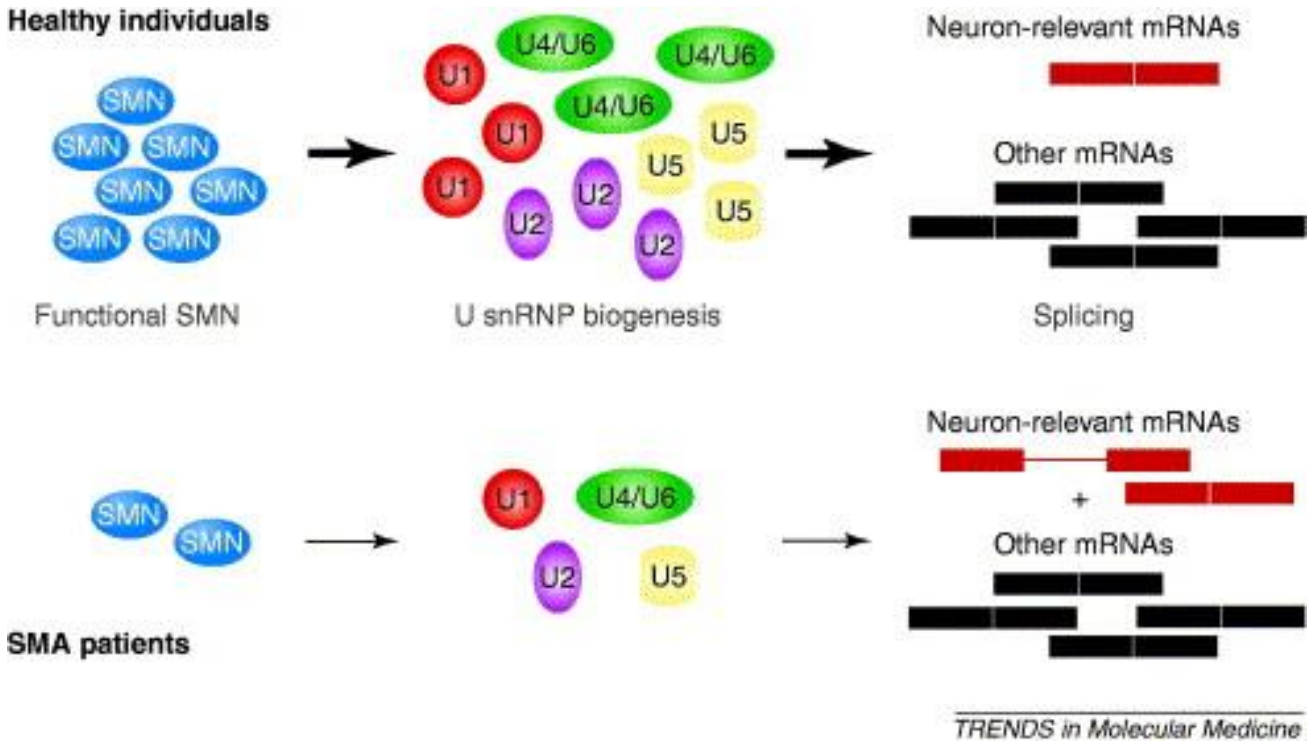


•On nuclear re-entry, the Sm-class snRNPs target to Cajal bodies for snRNP maturation, which includes binding by snRNP-specific proteins and site-specific modification by small Cajal body (sca)RNPs

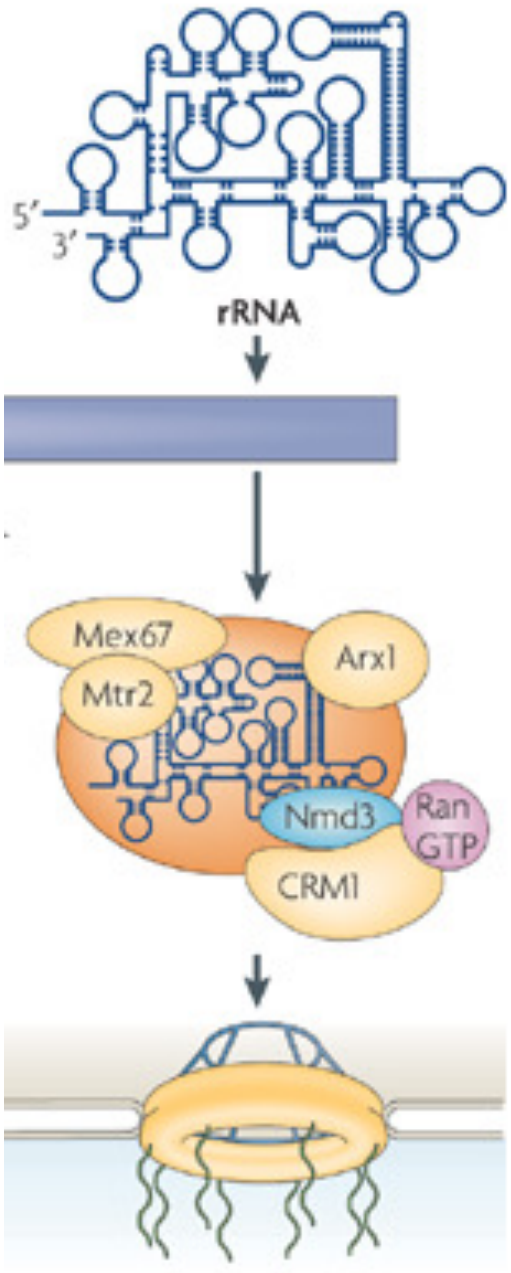
•The newly assembled snRNPs either participate in splicing at **perichromatin fibrils (PFs)** or are stored in **interchromatin granule clusters (IGCs)** for later use

Spinal muscular atrophy: the RNP connection

SMN deficiency results in reduced production of spliceosomal U snRNPs. As a consequence, processing of mRNAs with sub-optimal splice sites (e.g. tissue-specific transcripts) would be compromised. Therefore, inefficient splicing of neuron-relevant mRNAs is the basis for the tissue-specific phenotype in SMA.



rRNA EXPORT



rRNA associate to the ribosomal subunits inside the nucleus.

Ribosomal subunit export has to be very efficient: 2 export systems:

- CRM1/Exportin 5 (RAN-GTP-dep.)
- MEX67-MTR2 (yeast), RAN-GTP-indep. (used by mRNAs).

In mammals, the pre-60S subunit is exported by Crm1 or Exportin 5 (Xpo5), whereas the pre-40S subunit is exported by only Crm1. Crm1 recognises the nuclear export signal (NES) of ribosome-bound NMD3 in a RanGTP-dependent manner

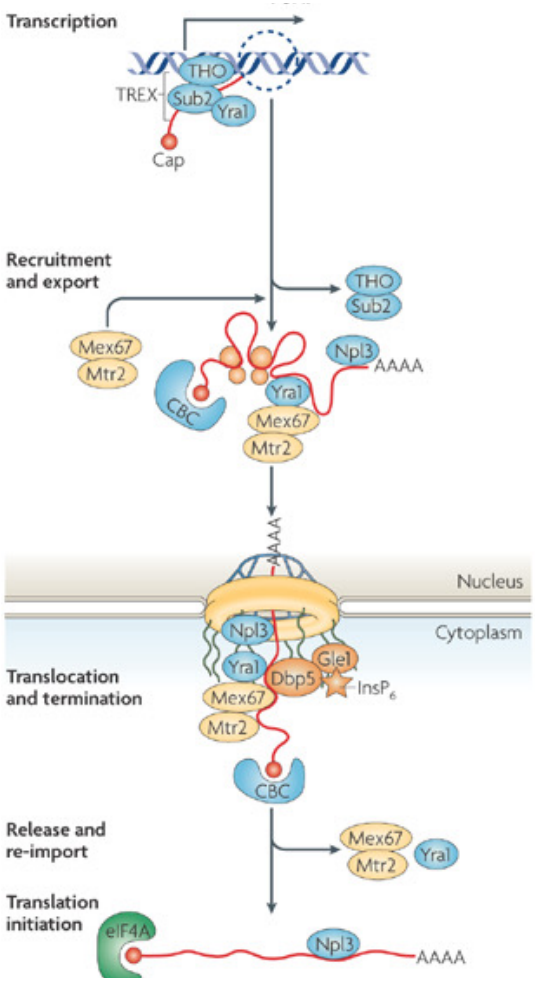
In yeast Mex67/Mtr2 is involved in the export of pre-60S particles together with additional export factors. Crm1 is necessary for nuclear export of the pre-40S ribosome tigether with Rio2 (with NES),the export adapter slx9 and RanGTPc

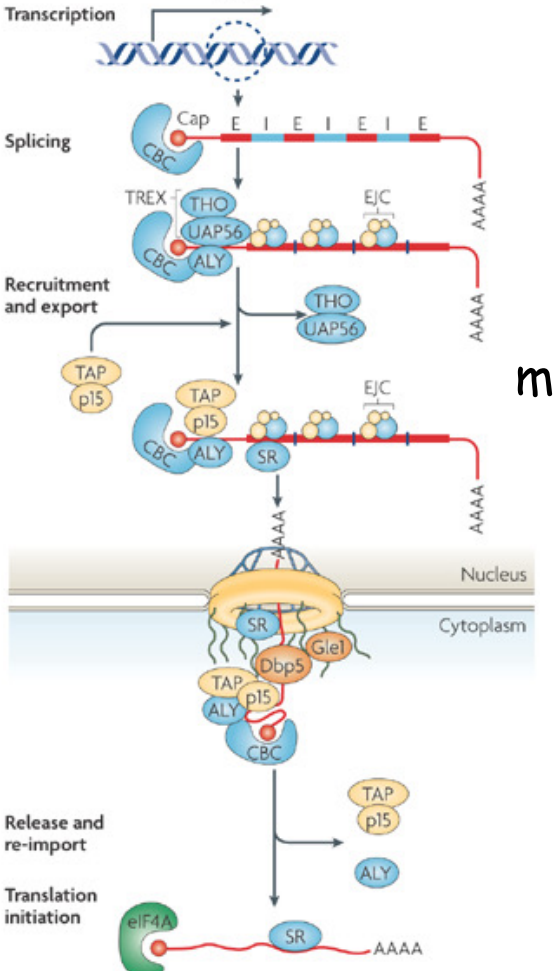
mRNA Export

•mRNAs are channelled into the specific export pathway coordinately with their processing and assembly into messenger (m)RNPs.

•Among the factors bound to the pre-mRNAs are also export <u>adaptors</u> that serve to establish a physical bridge between the mRNA molecule and its export receptor.

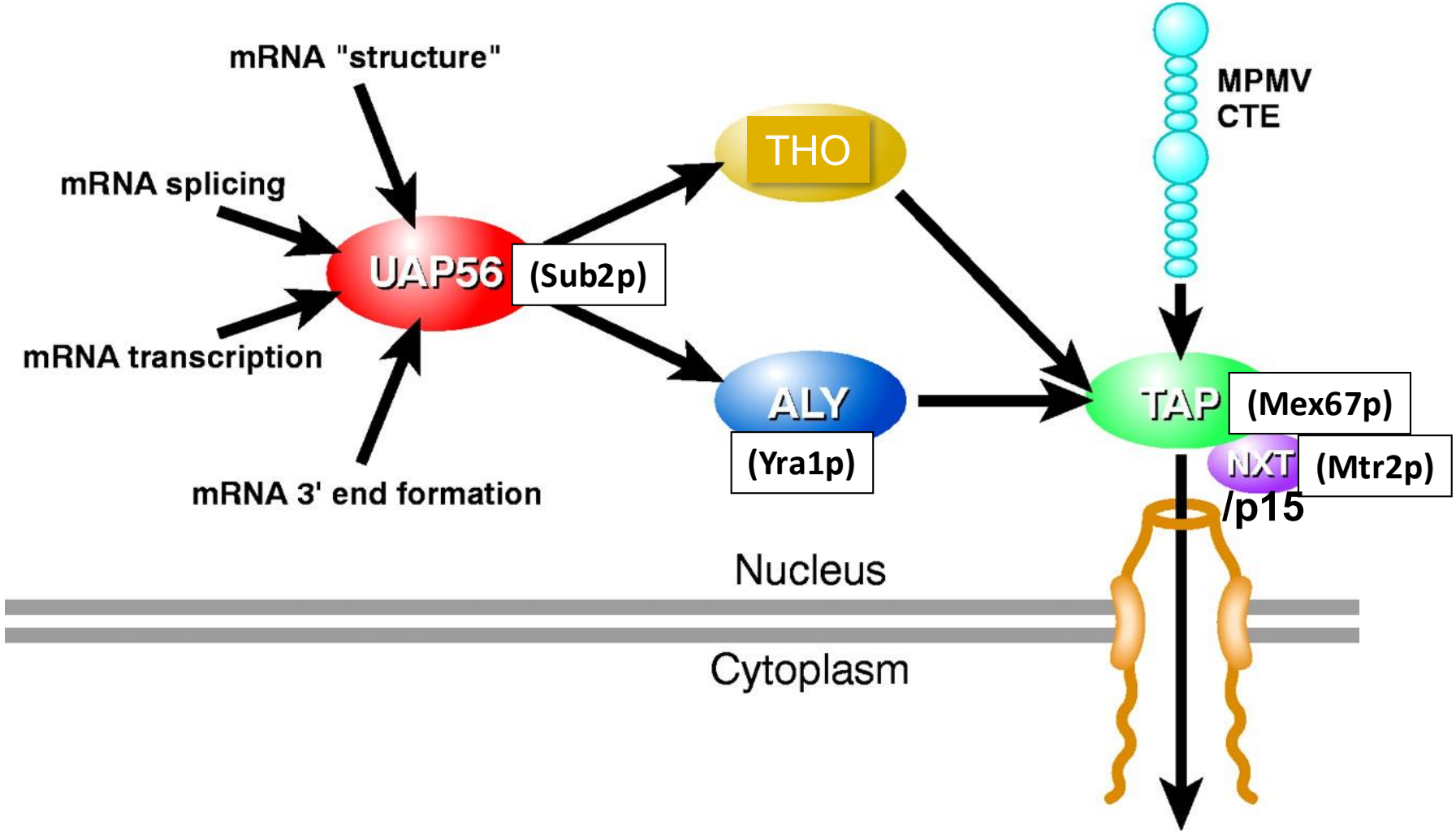
yeast





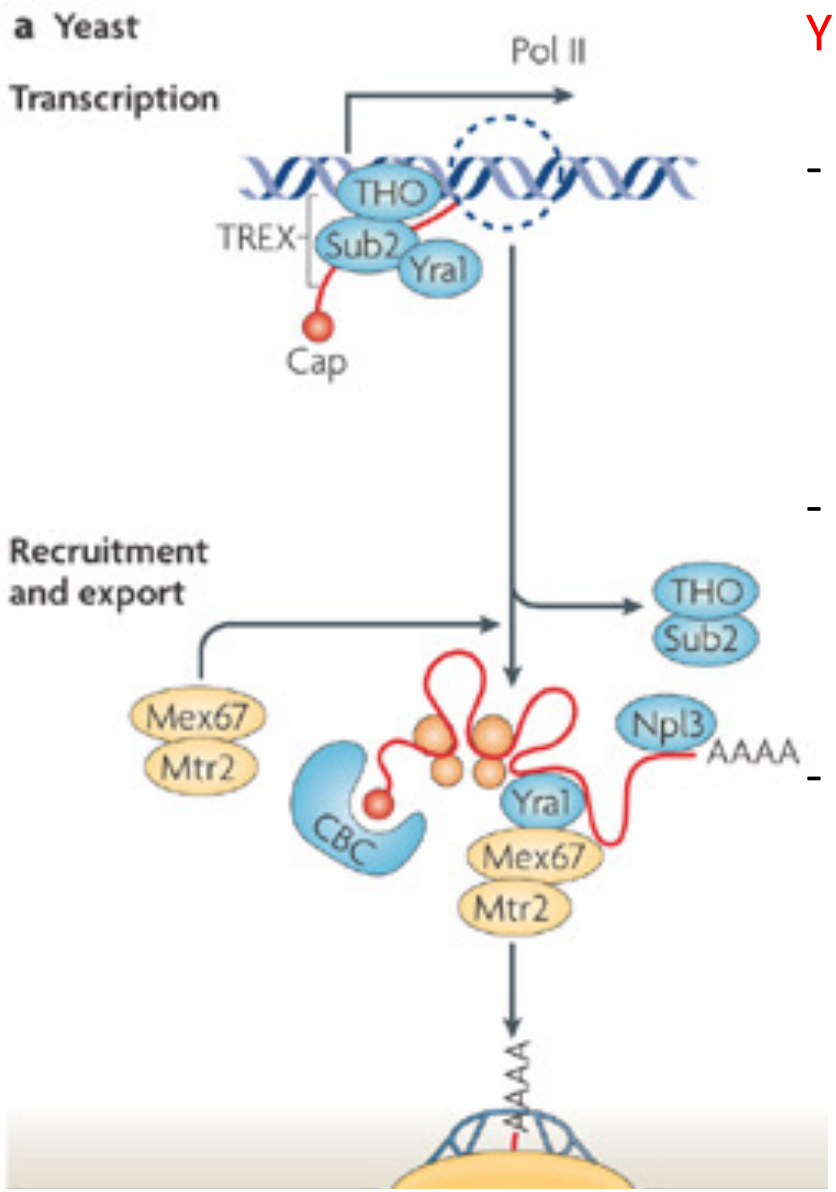
metazoa

Factors involved in Nuclear Export of mRNA



(Corresponding yeast factors indicated in parentheses)

mRNA EXPORT: differences between YEAST and METAZOA



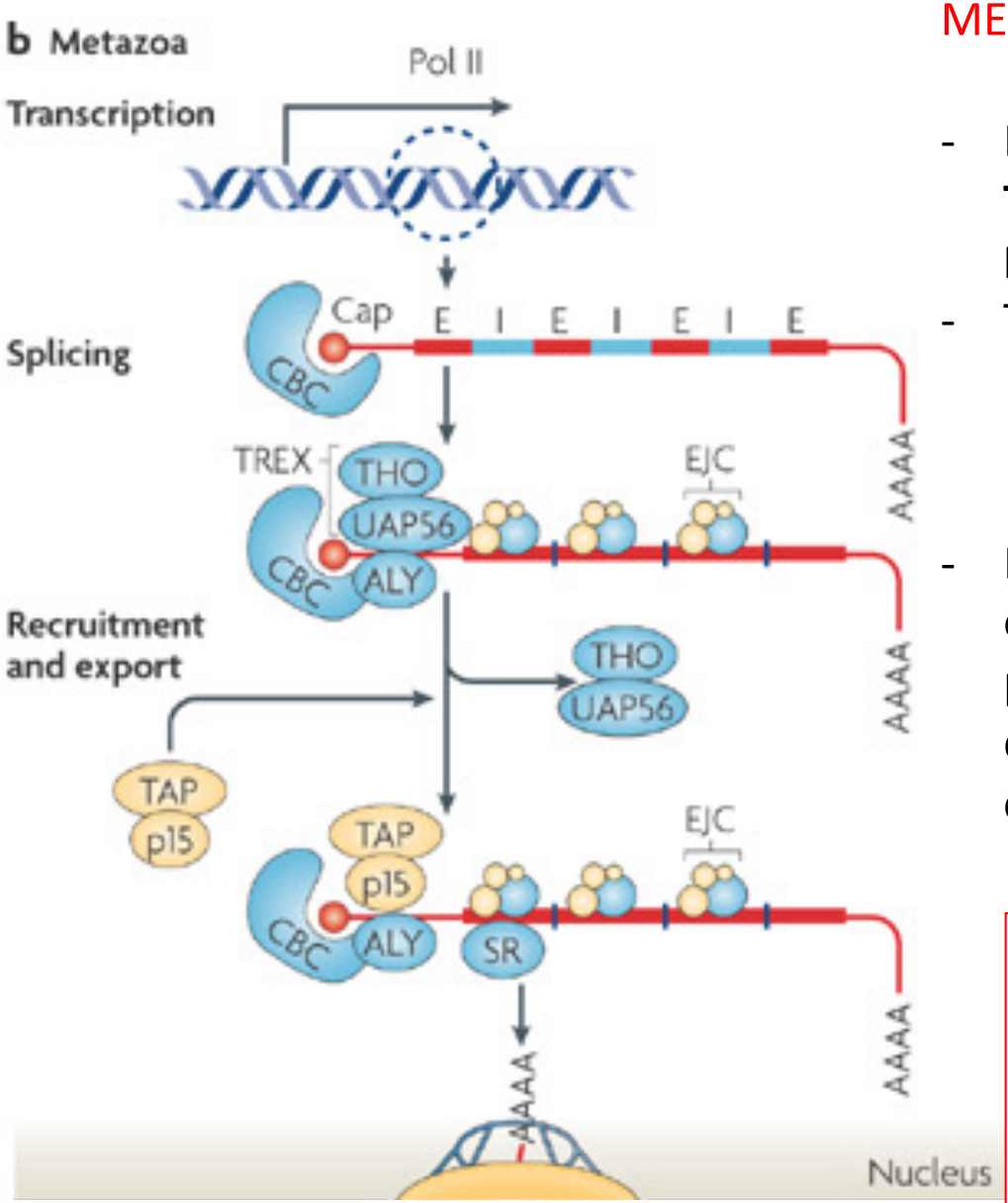
YEAST:

- The export protein is **Mex67** together with **Mtr2,** which recognize the nucleoporins. The Mex67-Mtr2 complex associate to the mRNP complex.

- Mex67 does not recognize the mRNA directly, but recognizes adaptor proteins, such as **Yra1**.

- The export is connected to **TRANSCRIPTION**: the adaptor protein is associated to the mRNA since its transcription.

mRNA Export



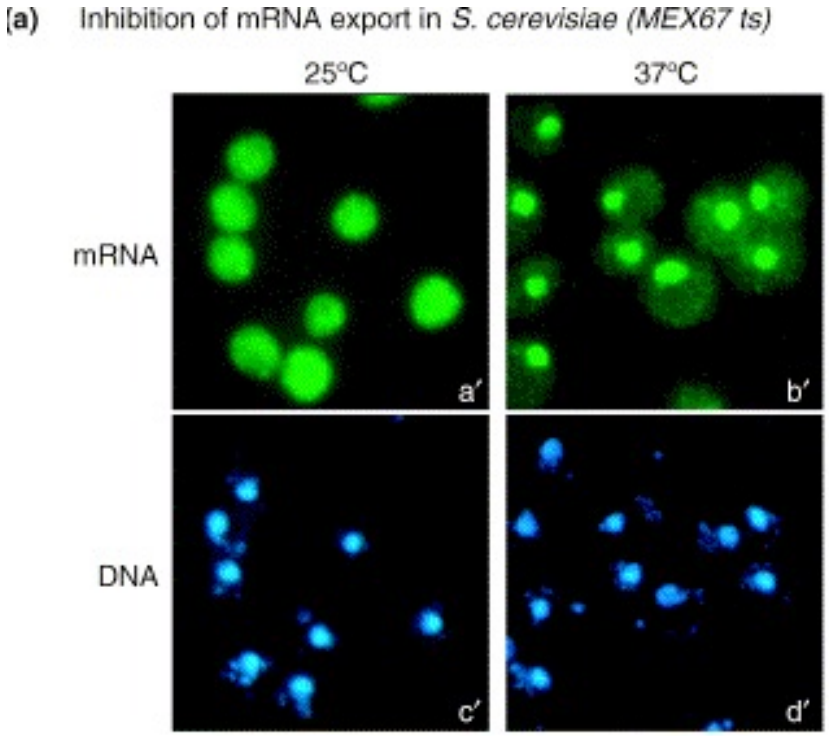
METAZOA:

- Export proteins and adaptors are conserved: **TAP–p15 complex** (also known as **NXF1–NXT1**).
- The export is connected to **SPLICING**.

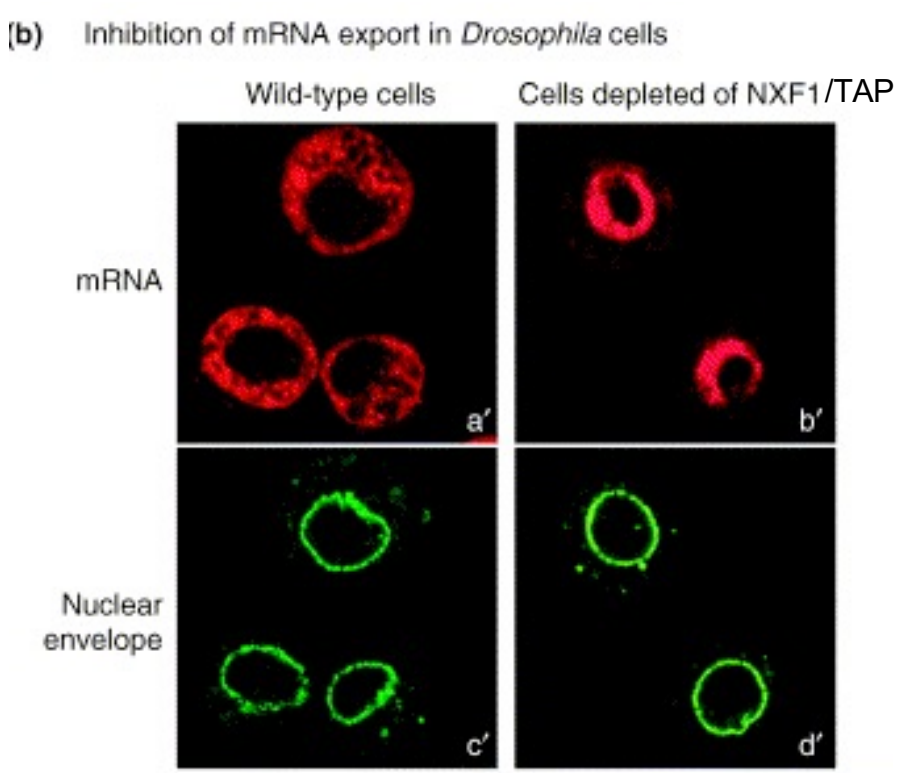
- Besides its physiological role in cellular mRNA export, human TAP transports a set of viral pre-mRNAs to the cytoplasm by binding directly to specific viral RNA elements called constitutive transport elements (CTEs)

The conserved mRNA exporter is structurally unrelated to the karyopherins and is **RanGTP independent** but it can physically interact with the Phe-Gly-rich repeats of FG nucleoporins

Mex67p and TAP (NXF1) are essential for export of bulk mRNA



Shifting the mex67^{ts} strain to the restrictive temperature (37° C) causes nuclear mRNA accumulation.

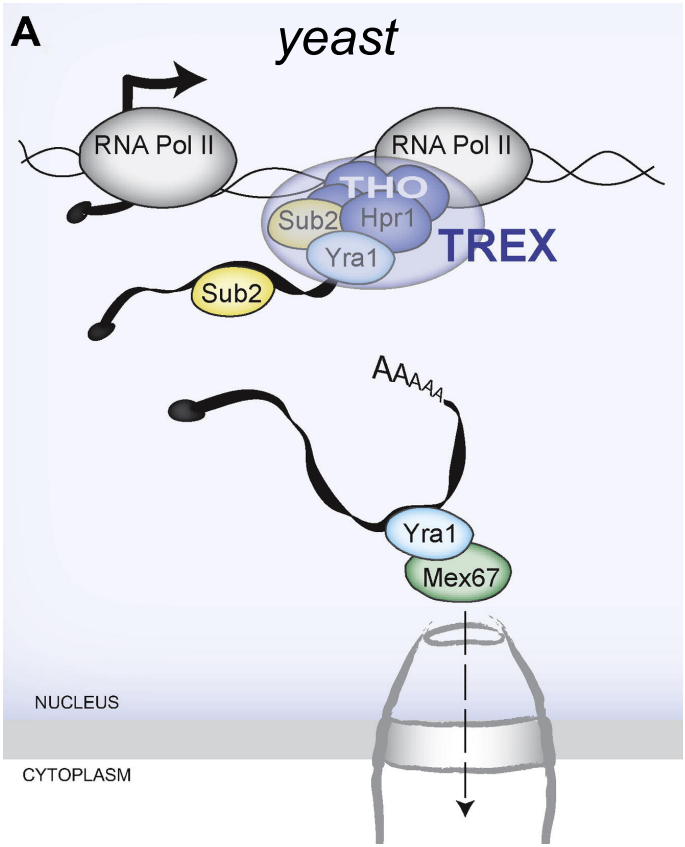


Depletion of Drosophila cells of NXF1(TAP) by double stranded RNA inhibition causes nuclear mRNA accumulation.

In both experiments, poly(A) mRNA was visualized by in situ hybridization with a fluorescently-labeled oligo-dT probe

The Yra1 and Aly/REF adaptors

•Because of its low affinity and low specificity for binding mRNAs, Mex67/TAP requires adaptor proteins to interface with mature transcripts ready for export. So far, the best-characterized adaptor for Mex67/TAP is the essential **Yra1** protein in yeast or **Aly/REF** in higher eukaryotes.



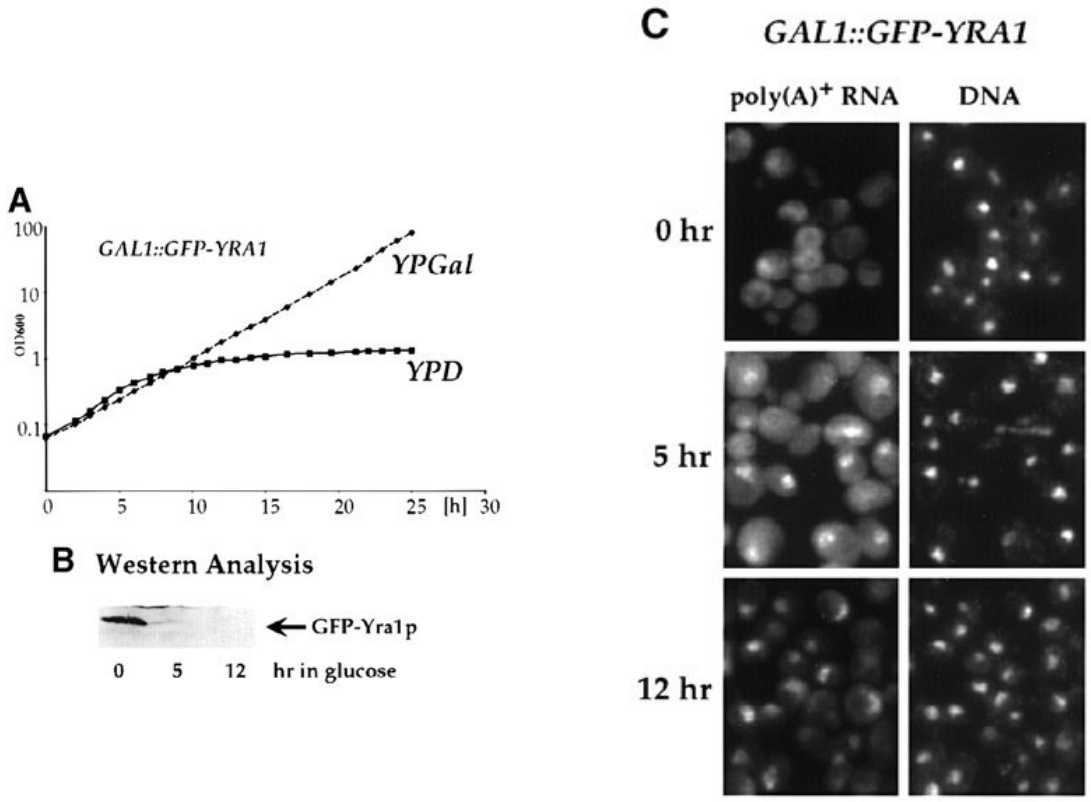
•Thus, Yra1 and ALY/REF form a bridge between an upstream-acting RNA-binding protein and a downstream-acting mRNA export receptor.

Yra1 and its partner Sub2 (UAP56 in metazoans), a DEAD box helicase required for mRNA export, were biochemically purified with the THO complex (consisting of Hpr1, Mft1, Thp2, and Tho2), involved in mRNP biogenesis and export.

The Yra1 and the adaptor Sub2

- Yra1 was identified as an interactor of Mex67p

- Yra1p-depleted cells accumulated poly(A)$^+$ RNA inside the nucleus.

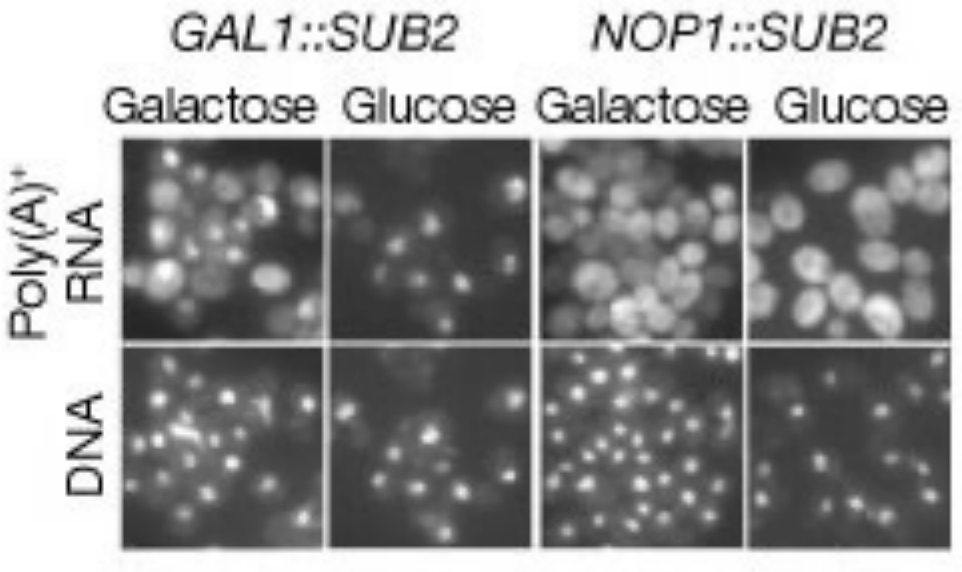


poly(A) mRNA was visualized by in situ hybridization with a fluorescently-labeled oligo-dT probe

Sub2p

•Sub2 (UAP56 in human) was identified as an interactor of Yra1 and it is a member of the DEAD box family of RNA helicases

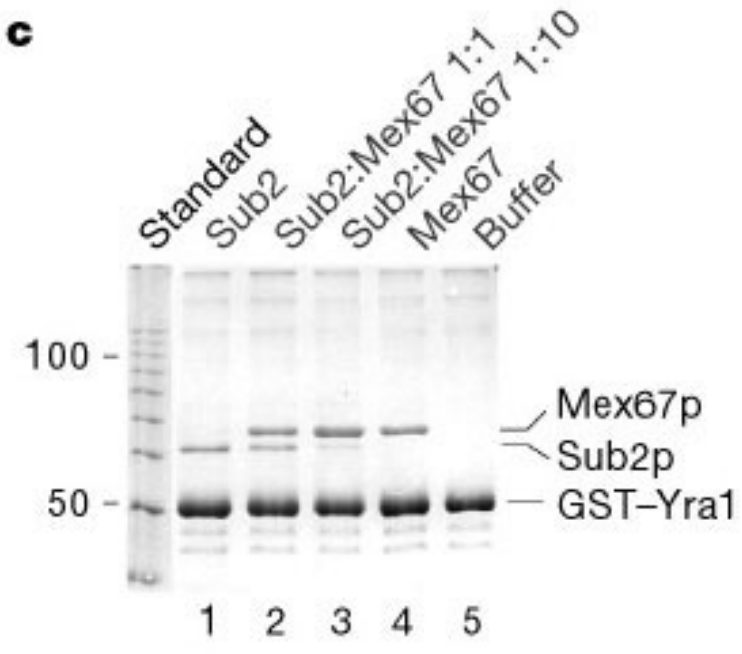
•Sub2p-depleted cells accumulated poly(A)$^+$ RNA inside the nucleus.



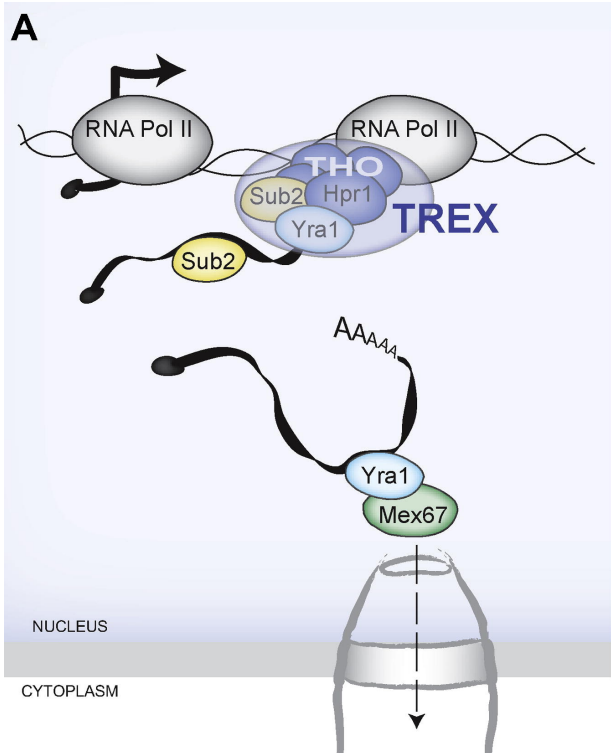
poly(A) mRNA was visualized by in situ hybridization with a fluorescently-labeled oligo-dT probe

Mex67p competes with Sub2p for binding to Yra1p.

•Mex67p competed with Sub2p for binding to Yra1p when Mex67p and Sub2p were present in equimolar amounts

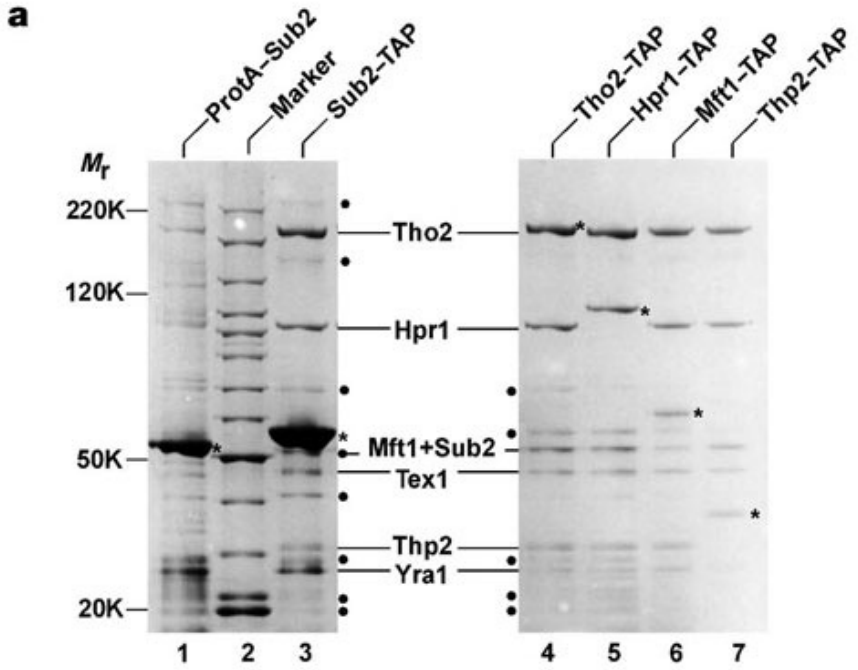


Competition assay



Yra1 and Sub2 are associated with the THO complex

•Yra1 and its partner **Sub2** (**UAP56** in metazoans), a DEAD box helicase required for mRNA export, were biochemically purified with the THO complex (consisting of Hpr1, Mft1, Thp2, and Tho2), involved in mRNP biogenesis and export. Likewise, Aly/REF and UAP56 have been found to interact with the human counterparts of the complex



Tho2, Hpr1, Mft1 and Thp2 are all components of the **THO complex**

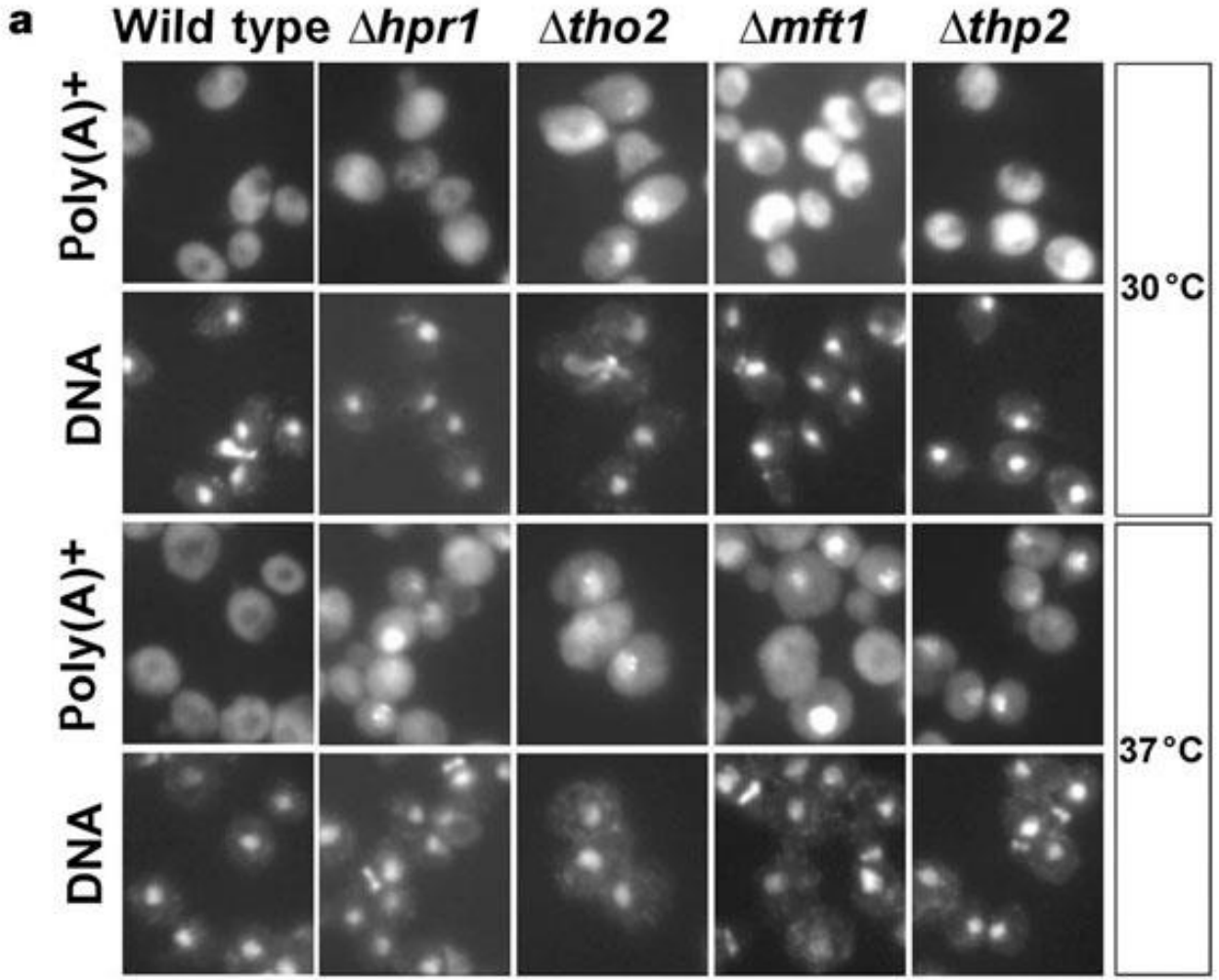
THO Complex

- THO was identified as a four proteins complex containing proteins encoded by *THO2* and *HPR1*, two genes previously identified by hyper-recombination mutations, and *MFT1* and *THP2*. The null mutations of each of the four genes are viable and yield similar phenotypes of impairment of transcription elongation and transcription-dependent hyper-recombination between direct repeats.

- It was previously proposed that the THO complex has a functional role related to RNAP II transcription elongation.

- THO and RNA export factors are functionally related. For example, *sub2*, *yra1*, *mex67* and *mtr2* mutants show similar defective transcription and hyper-recombination phenotypes to THO mutants.

Deletion of Tho2, Hpr1, Mft1 and Thp2 results in mRNA export defect



poly(A) mRNA was visualized by in situ hybridization with a fluorescently-labeled oligo-dT probe

Factors that influence mRNA export

The coupling of nuclear mRNA biogenesis to the recruitment of the export machinery is largely coordinated by the transport/export complex (TREX)

Yeast	Metazoans
Mex67p	TAP
Mtr2p	NXT1
Sub2p	UAP56 (Hel)
Yra1p and Nab2	Aly (Ref1)/THOC4
Tho2p	THOC2
Hpr1p	THOC1
Mft1p	THOC3
Thp2p	THOC5/6/7
Tex1	?
	?

mRNA export receptor

mRNA export adaptors

TREX Complex

THO Sub Complex

The TREX complex contains THO and 2 mRNA export factors

- In yeast, the multi-subunit TREX complex plays a role in coupling transcription to mRNA export. This complex contains the mRNA export factors **Sub2p** and **Yra1p** as well as the **THO complex**, which functions in transcription elongation.
- Human TREX contains Aly/REF, UAP56, and the human counterpart of the yeast THO complex. The human THO complex specifically associates with spliced mRNA and not with unspliced pre-mRNA.
- Recent data indicate that recruitment of the human TREX complex to spliced mRNA occurs by a splicing-coupled mechanism rather than by the direct transcription-coupled mechanism that occurs in yeast.
- Deletion mutants of THO components show defects in transcription elongation and have a hyper-recombination phenotype. The elongation defects and hyper-recombination appear to be due to the presence of RNA/DNA hybrids that form between the nascent RNA and the DNA template.

The TREX complex contains THO and 2 mRNA export factors

- In yeast, the multi-subunit TREX complex plays a role in coupling transcription to mRNA export. This complex contains the mRNA export factors **Sub2p** and **Yra1p** as well as the **THO complex**, which functions in transcription elongation.

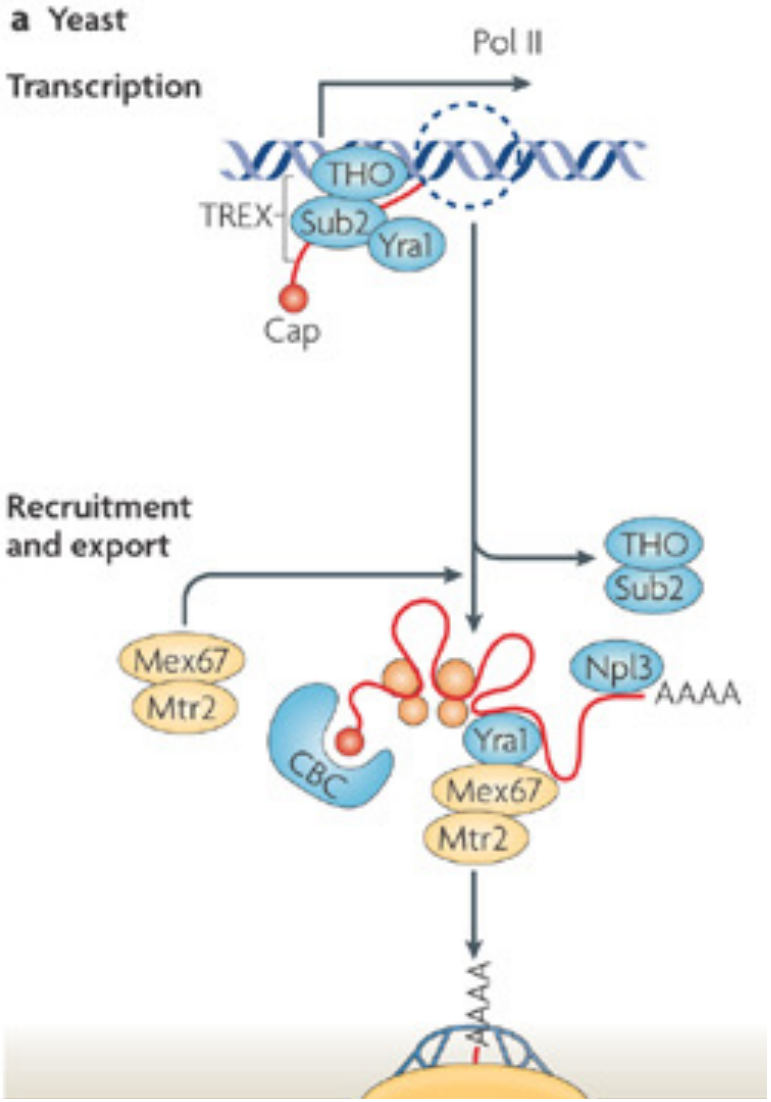
mRNA EXPORT: YEAST
In the nucleus…

The exporter MEX67 has **not** RNA binding domains

YRA1 (Adaptor) is the RNA binding protein which bridges mRNA and the exporter MEX67

SUB2 interacts with YRA1 **(provides competence for the export to the mRNA)**

SUB2 is a RAN-helicase recurited by the THO elongation complex during transcription.



mRNA EXPORT: YEAST in the nucleus…

The exporter MEX67 has **not** RNA binding domains

MEX67 interactor with RNA binding domains is **YRA1 (Adaptor): a** bridge between the mRNA and the exporter

SUB2 interacts with YRA1 **(provides competence for the export to the mRNA)**

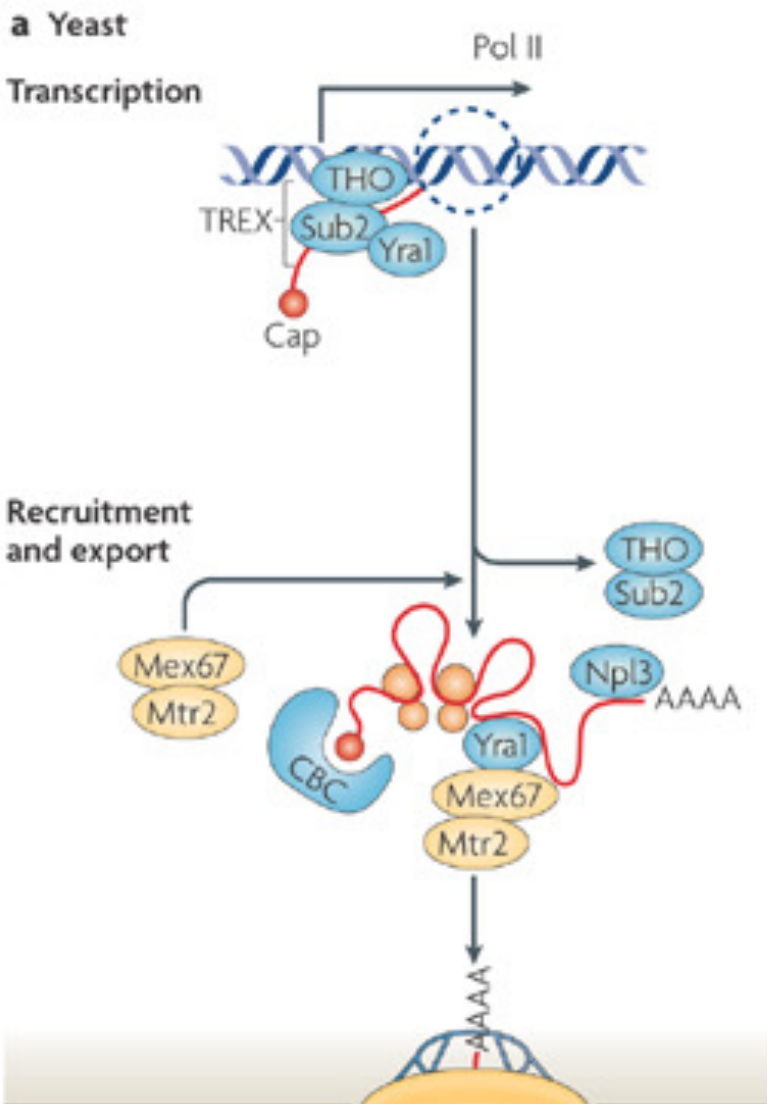
SUB2 is a RAN-helicase recurited by the THO elongation complex during transcription.

1. When YRA1 interacts with SUB2, it cannot bind MEX67 -> **mRNA+SUB2+YRA1= not ready for the export**

2. At the end of transcription, **SUB2 detaches** from the mRNA and YRA1 -> **YRA1 + MEX67** -> **mRNA ready to be exported.**

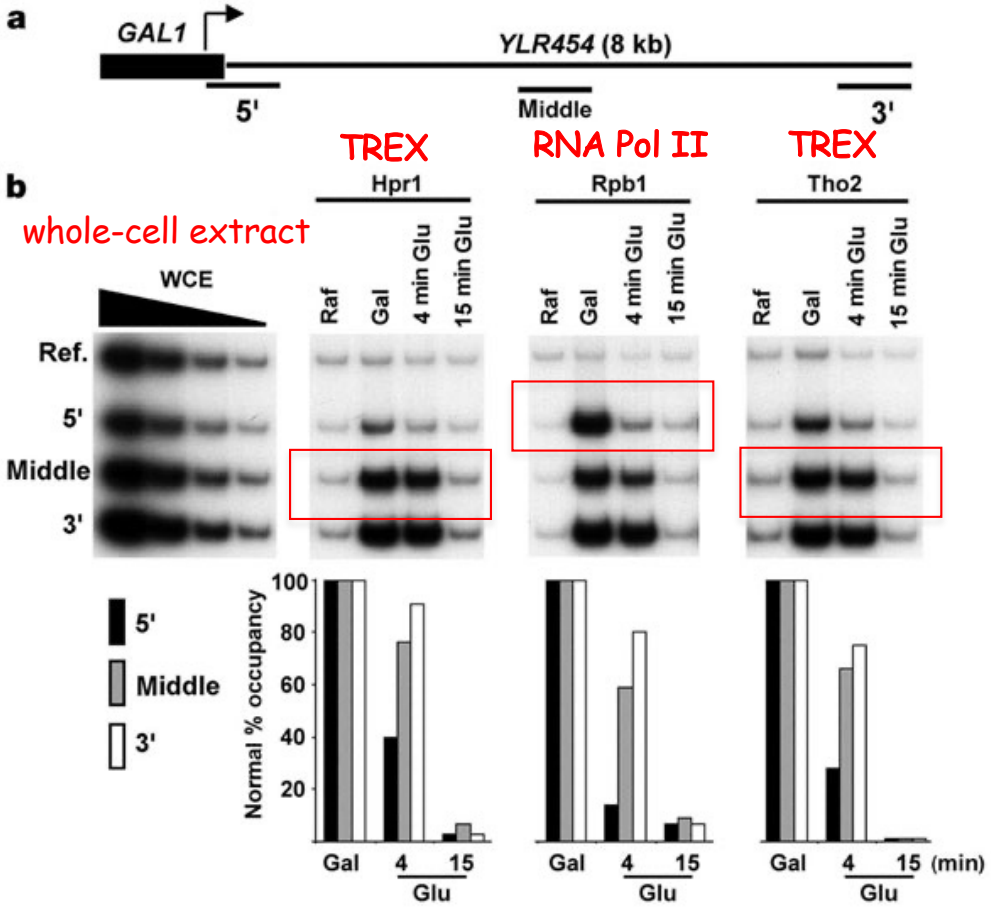
THO elongation complex is therefore important also for export: **THO+SUB2+YRA1= TREX** complex

TREX complex associates to the mRNA **during transcription** elongation, in yeast

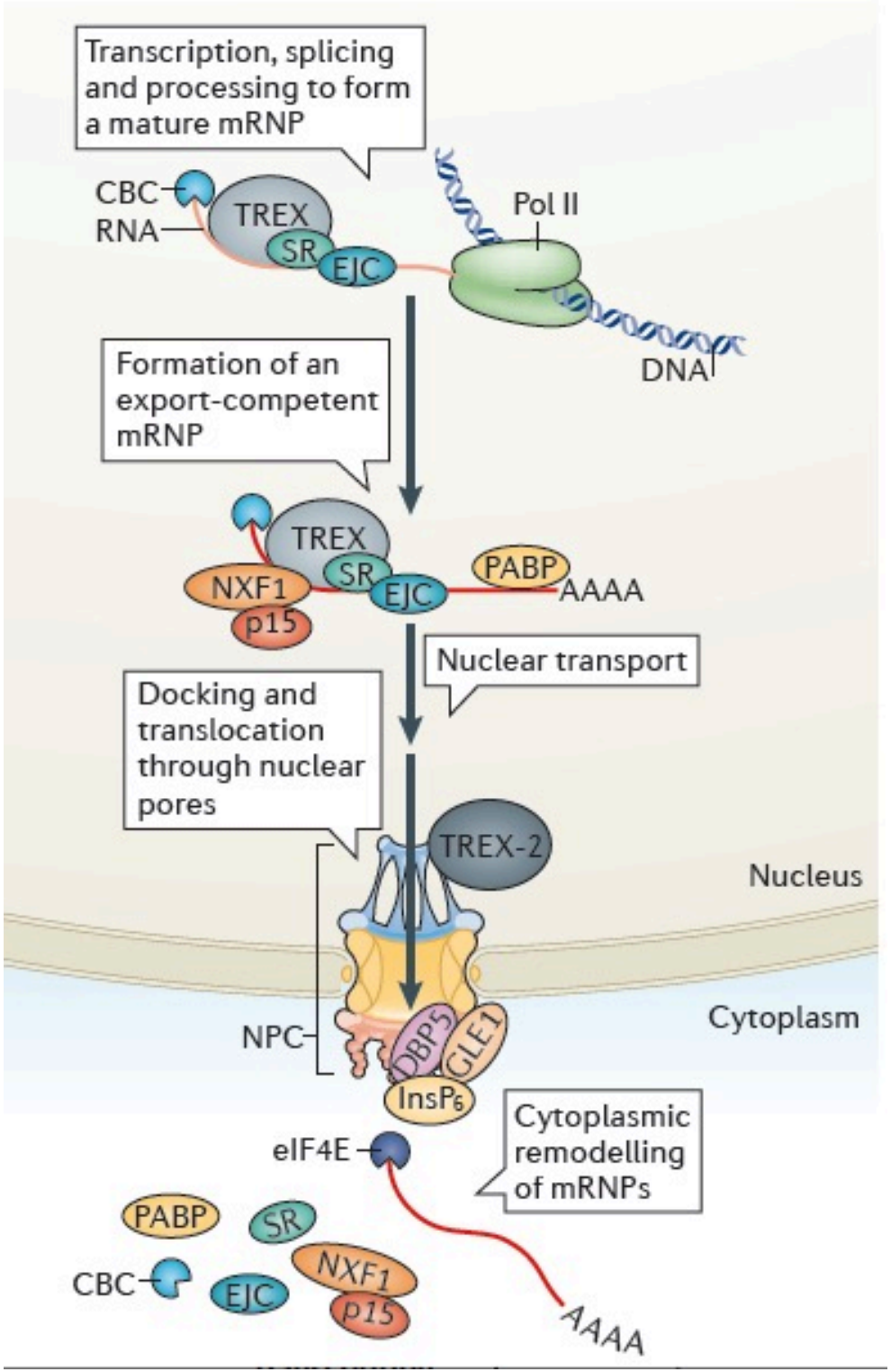


TREX complex operates in coupling transcription elongation to mRNA export

•The TREX complex is specifically recruited to the transcribing gene and travels with the polymerase during transcriptional elongation.



chromatin immunoprecipitation assay in strains containing a long yeast gene (ORF YLR454) under the control of the regulatable GAL1 promoter



In Metazoa TREX complex is recruited to the transcribing gene by different mechanisms:

1. CAP-dependent manner: CAP binding subunit CBP80 interacts with the ALY/REF component of TREX. Suggenting why mRNAs are exported in a 5'➔3' direction.

2. Splicing-dependent manner: UAP56 interacts with the splicing factors U2AF2 and together with Aly/REF and TAP-p15 interatct with the exon-junction complex which is deposited as a consequence of splicing 20–24 nucleotides upstream of every exon-exon junction in the spliced mRNA.

3. transcription dependent manner: SPT6 elongation factor recruits IWS1 to act ad bridging protein for Aly/REF

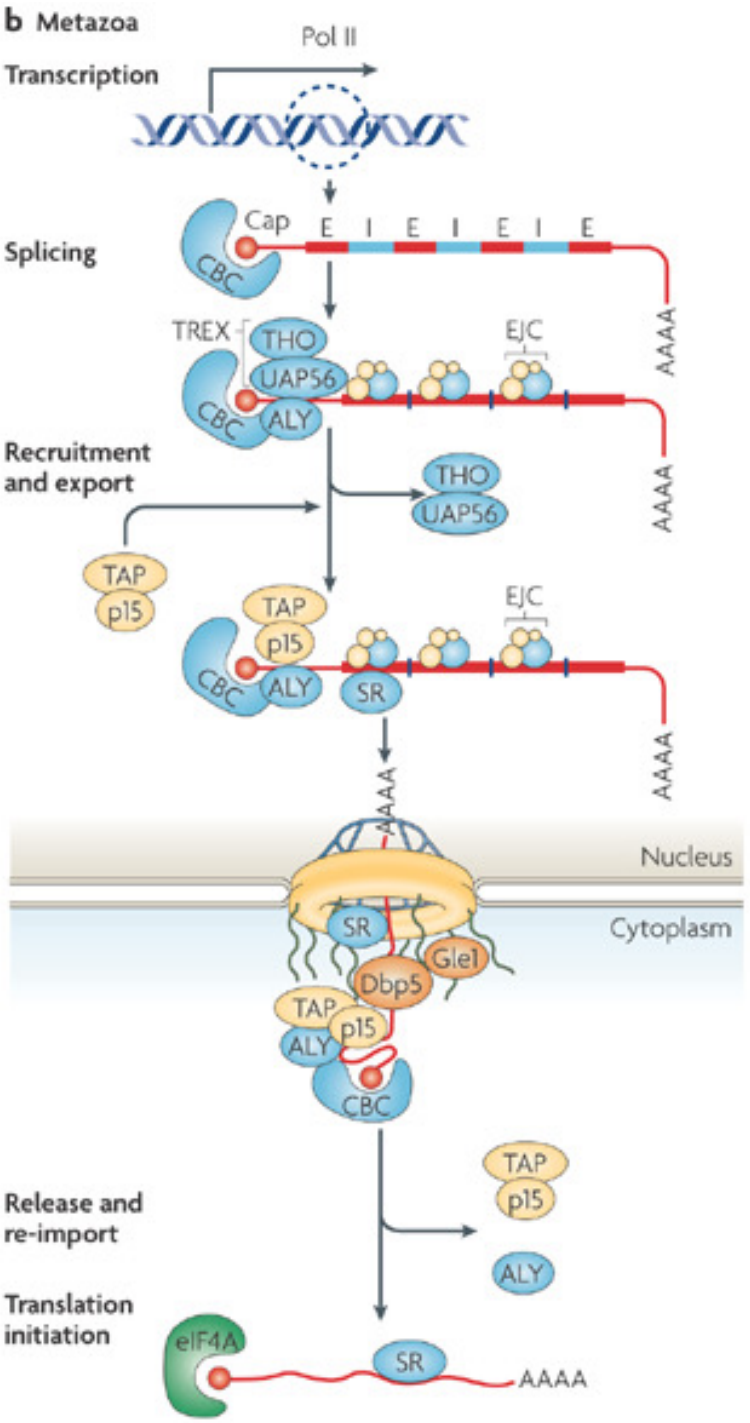
1. Metazoan TREX contains Aly/REF (YRA1), UAP56 (SUB2), and the metazoan counterpart of the yeast THO complex.

2. Human TREX complex binds only to **spliced mRNAs** by a **splicing-coupled mechanism,** rather than by the direct transcription-coupled mechanism that occurs in yeast.

3. TREX is recruited by the **cap-binding complex** and by the **exon-junction-complex**

4. In human ALY/REF (YRA1) is recruited to the mRNP **via UAP56 (SUB2) during splicing, in an ATP dependent manner**

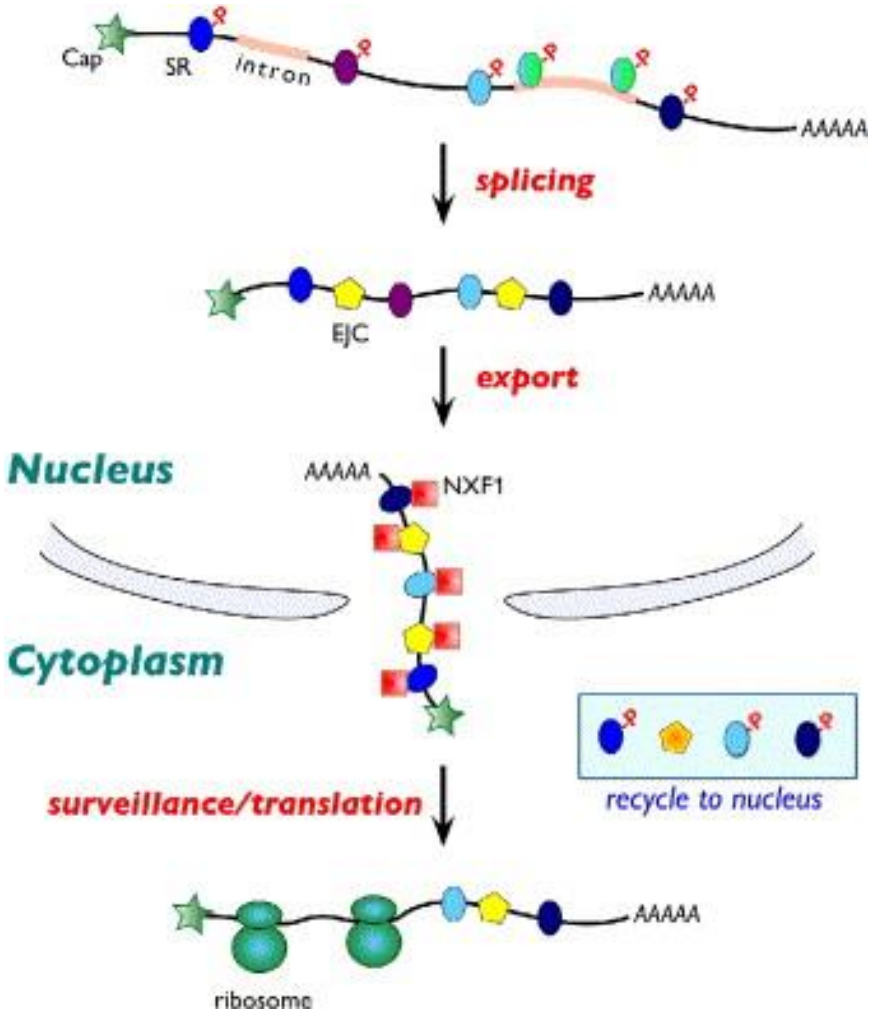
5. Aly/REF, in contrast to Yra1, which is essential for mRNA export in yeast, is required but **not essential** for bulk cellular mRNA export. This suggests the existence of additional mRNA export adaptors in metazoa



Export competency is linked to splicing in metazoa

•The N-terminal domains of the **SR proteins** bind the same N-terminal portion of TAP (NXF1) that associates with the REF/Aly family of export adapter proteins. **TAP interacts preferentially with shuttling SR proteins that are hypophosphorylated.**

•SR proteins are initially recruited to pre-mRNAs for splicing in their hyperphosphorylated forms, they become partially dephosphorylated during the splicing reaction and more avidly bind TAP (NXF1). **The phosphorylation state of bound SR proteins contributes to the ability of the export machinery to discriminate between spliced and unspliced mRNPs**



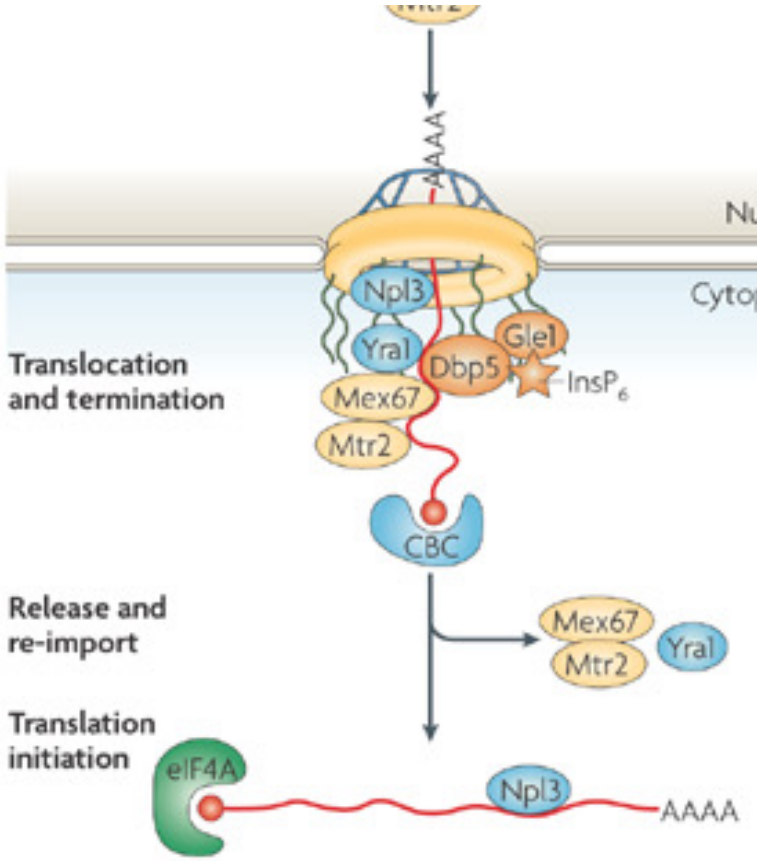
TAP=Mex67
Aly=Yra1

Directionality and termination of mRNA export

•ATP-dependent RNA helicases such as **Dbp5** which are involved in mRNA export, could trigger an irreversible ATP-driven mRNP rearrangement at the cytoplasmic side of the NPC

•Dbp5 exhibits a very low ATP-dependent RNA-helicase activity, which can be stimulated by **Gle1,** an essential mRNA export factor that is also asymmetrically located at the cytoplasmic nuclear pore filaments

•Maximal stimulation of the ATPase activity of Dbp5 requires the signalling molecule **inositol heaxakisphospate** (**$InsP_6$**) which regulates the interaction between Gle1 and Dbp5

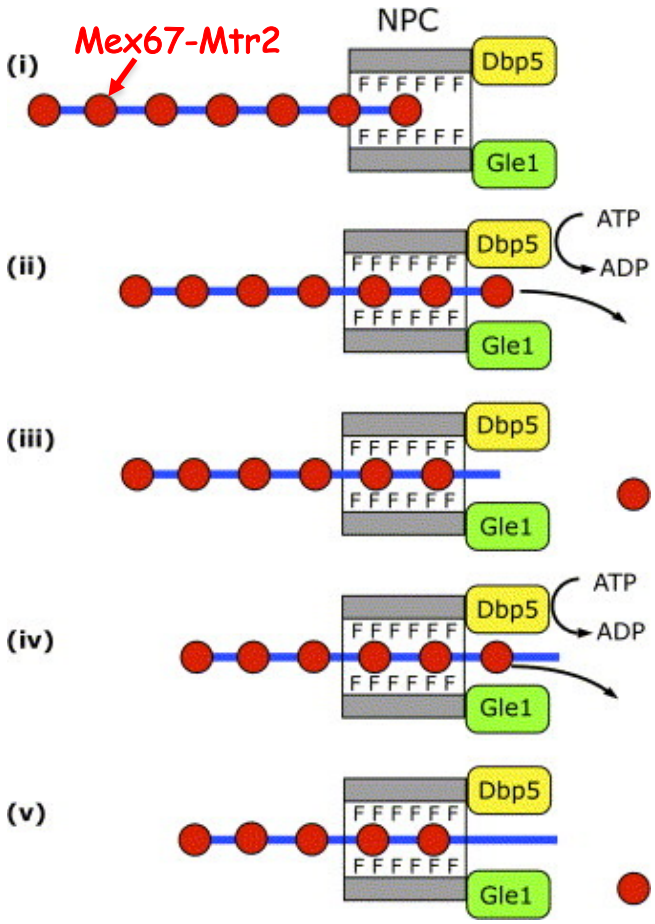


Directionality and termination of mRNA export

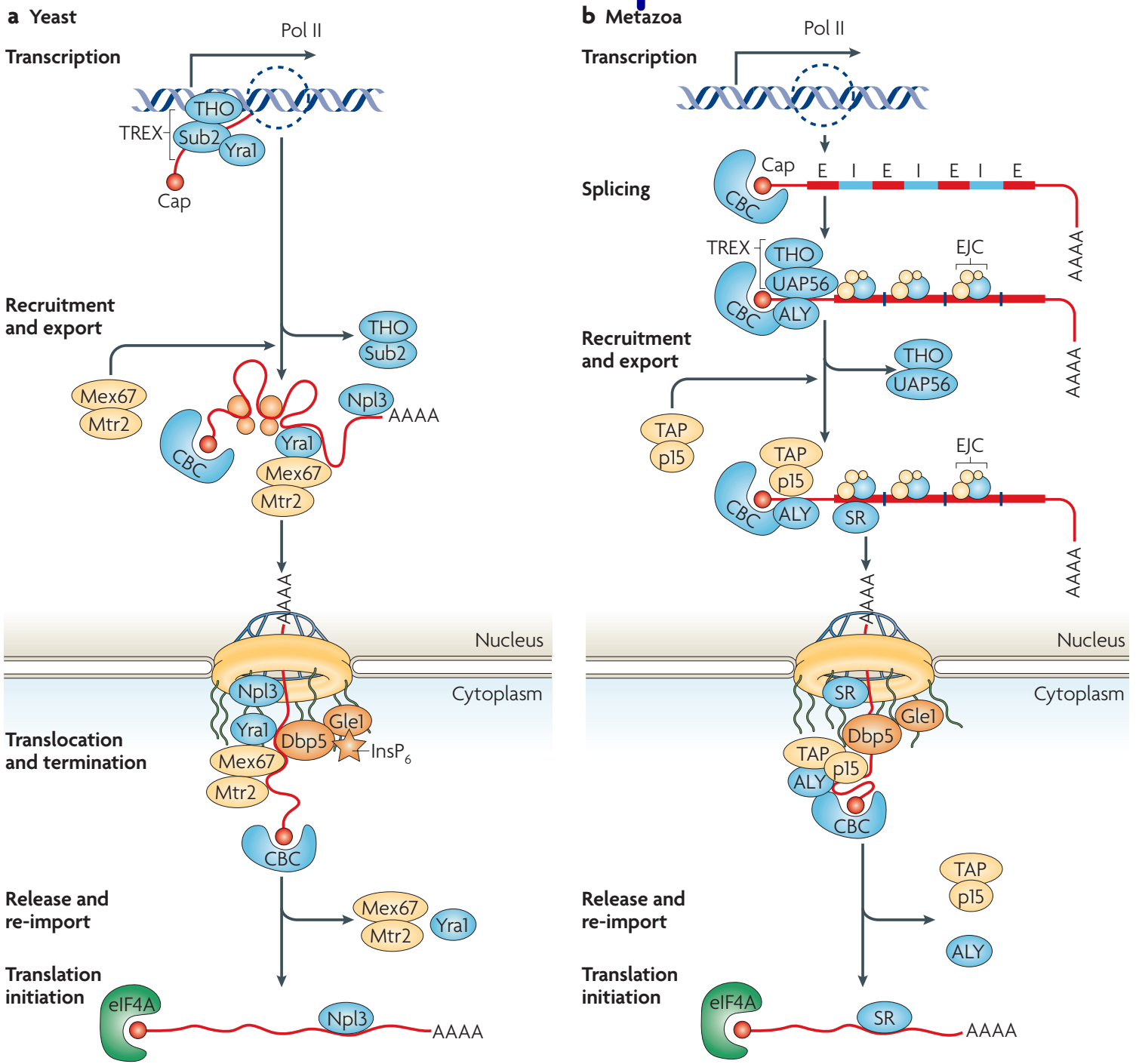
•Interactions between **Mex67:Mtr2** and **FG-nucleoporins (F)** that line the NPC transport channel facilitate movement of the mRNP

•When one of the Mex67:Mtr2 complexes reaches the cytoplasmic face of the NPC, it is removed from the mRNP by the **DEAD-box helicase Dbp5**, the ATPase activity of which is stimulated by **Gle1** and **$InsP_6$**

•Removal of **Mex67:Mtr2** prevents this segment of the mRNP from moving back into the transport channel and so functions as a molecular ratchet.

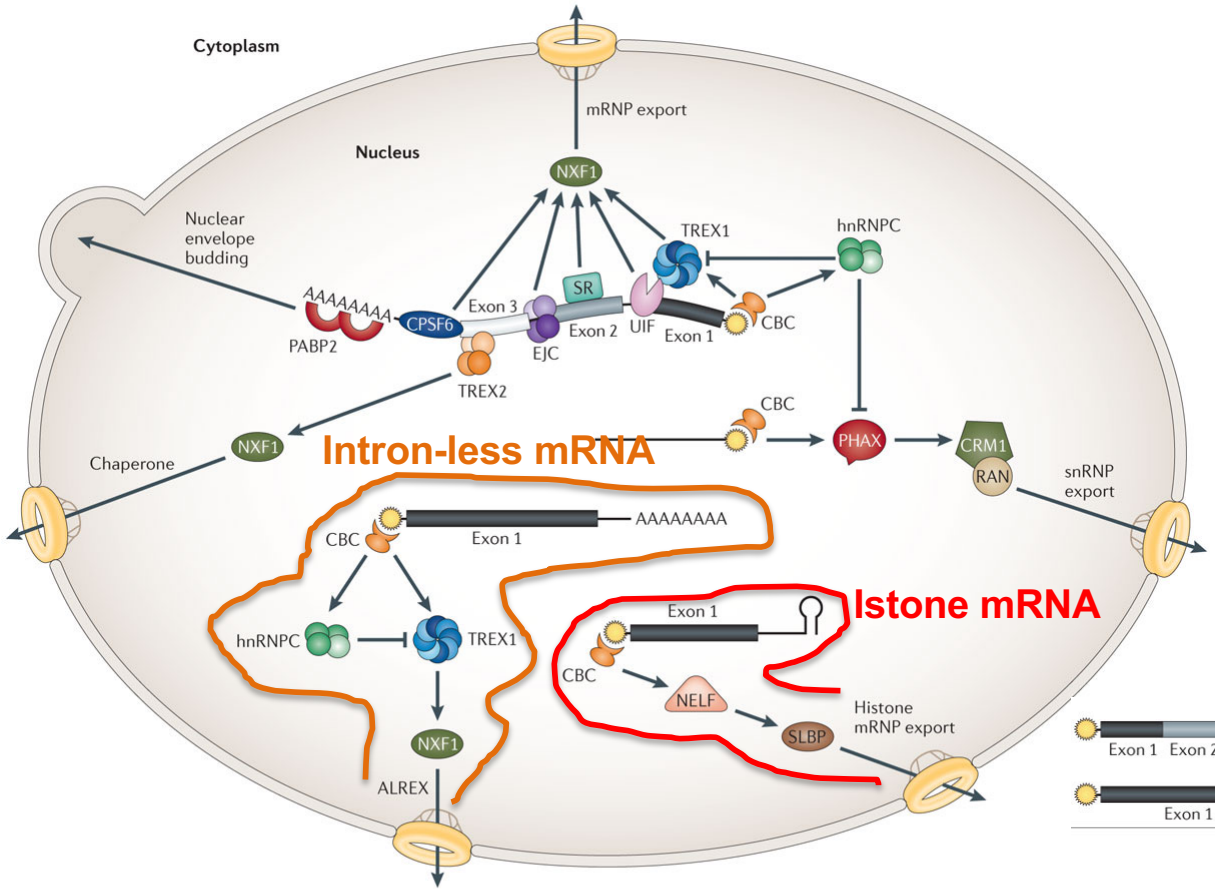


mRNA Export

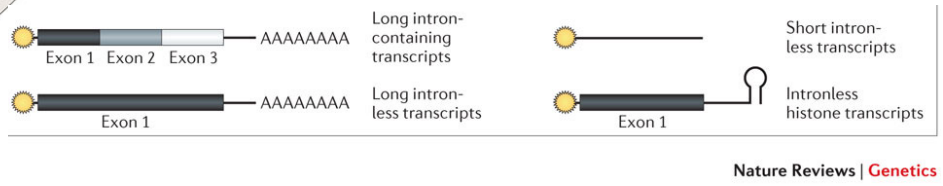

a Yeast
Transcription
Pol II
THO
TREX
Sub2
Yra1
Cap
Recruitment and export
THO
Sub2
Mex67
Mtr2
Npl3
AAAA
CBC
Yra1
Mex67
Mtr2
AAAA
Nucleus
Cytoplasm
Translocation and termination
Npl3
Yra1
Gle1
Dbp5
InsP6
Mex67
Mtr2
CBC
Release and re-import
Mex67
Mtr2
Yra1
Translation initiation
eIF4A
Npl3
AAAA
b Metazoa
Transcription
Pol II
Splicing
Cap
E I E I E I E
CBC
TREX
THO
UAP56
ALY
EJC
AAAA
Recruitment and export
THO
UAP56
TAP
p15
TAP
p15
ALY
CBC
SR
EJC
AAAA
AAAA
Nucleus
Cytoplasm
SR
Gle1
Dbp5
TAP
p15
ALY
CBC
Release and re-import
TAP
p15
ALY
Translation initiation
eIF4A
SR
AAAA

Alternative mRNA export (AREX)

TREX also participates in the nuclear export of several **intronless transcripts** independently of splicing. Efficient export requires the presence of GC-rich export-promoting sequences at the 5′ end of these transcripts. Recruitment of the TREX complex and **UAP56** to the 5′ end of intronless mRNAs (e.g. heat-shock protein 70) occurs through interaction with the cap-binding complex (CBC). This suggests that, similarly to yeast, in metazoans TREX1 functions in co-transcriptional loading of mRNA export factors to a subset of nascent transcripts. This alternative mRNA export pathway, which is termed **alternative RNA export** (**ALREX**), requires NXF1 (TAP) and the CBC.

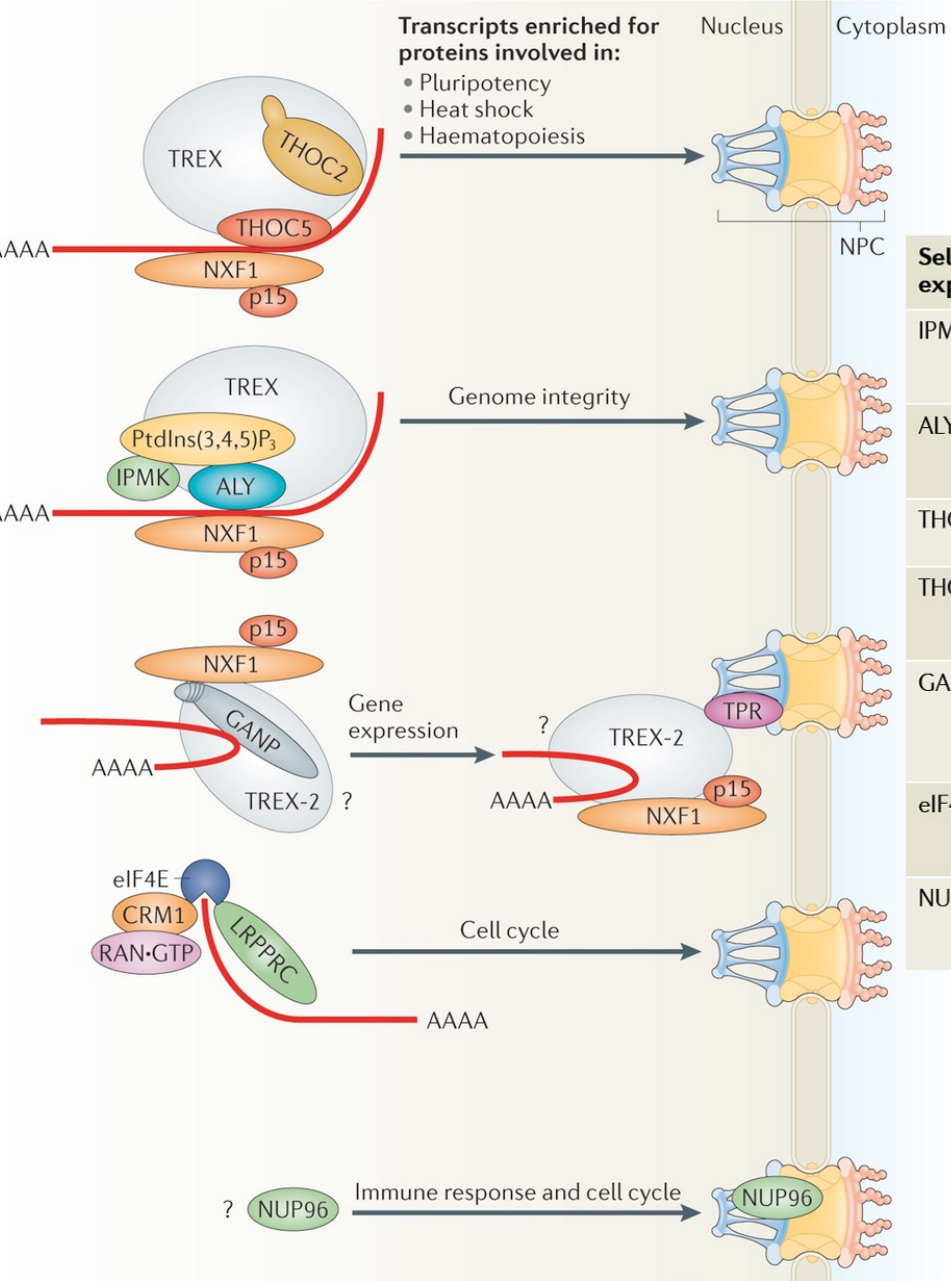


By contrast, **histone mRNAs**, which also lack introns, are exported to the cytoplasm by the **stem–loop-binding protein** (**SLBP**) which is recruited to the 3′ end of histone transcripts through interaction with the **CBC** and **negative elongation factor** (**NELF**)



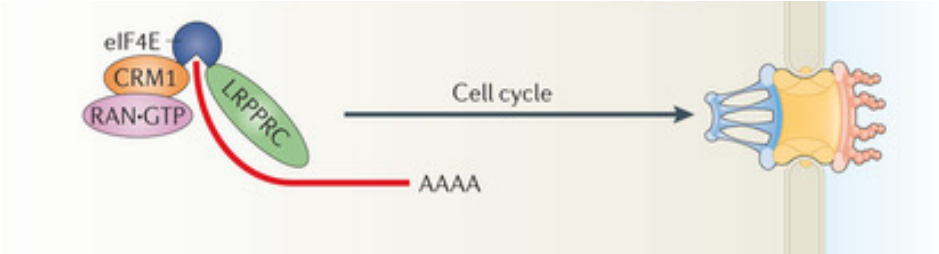
Another complex, which recently was shown to connect transcription with export of the matured mRNA is TREX-2, composed of Sac3, Thp1, Sem1, Sus1 and Cdc31. TREX-2 together with the SAGA complex are involved in docking transcribing genes to the NPC in a process called“gene gating” that allows preferential export of these mRNAs.

Control of mammalian gene expression by selective mRNA export



Selective mRNA export factor	Component of TREX or TREX-2	Function	Biological pathway (transcript examples)
IPMK	Neither	Kinase involved in synthesis of inositol phosphates and PtdIns(3,4,5)P_3	Genome duplication and repair; cell cycle (*RAD51*, *CHEK1* and *FANCD2*)
ALY	TREX	RNA-binding adaptor of TREX	Genome duplication and repair; cell cycle; heat shock (*RAD51*, *CHEK1*, *FANCD2* and HSP70)
THOC2	TREX	Core subunit of THO and TREX	Maintenance of pluripotency (*NANOG*, *SOX2* and *KLF4*)
THOC5	TREX	RNA-binding metazoa-specific subunit of TREX	Maintenance of pluripotency; haematopoiesis; heat shock (*NANOG*, *SOX2*, *KLF4* and HSP70)
GANP	TREX-2	Scaffold protein for TREX-2; exports a subset of mRNA, the transport of which depends on NXF1	Gene expression; RNA processing (*SP1*, *RPS23* and *ARPP19*)
eIF4E	Neither	Binds to m^{7}G cap of mRNA; export is CRM1-dependent and NXF1-independent	Cell proliferation and cell survival (cyclin D1 and *MYC*)
NUP96	Neither	Component of nuclear pore complex	Immune response; cell cycle (MHC I, MHC II, β2 microglobulin and *CDK6*)

Crm1 dependent mRNA Export

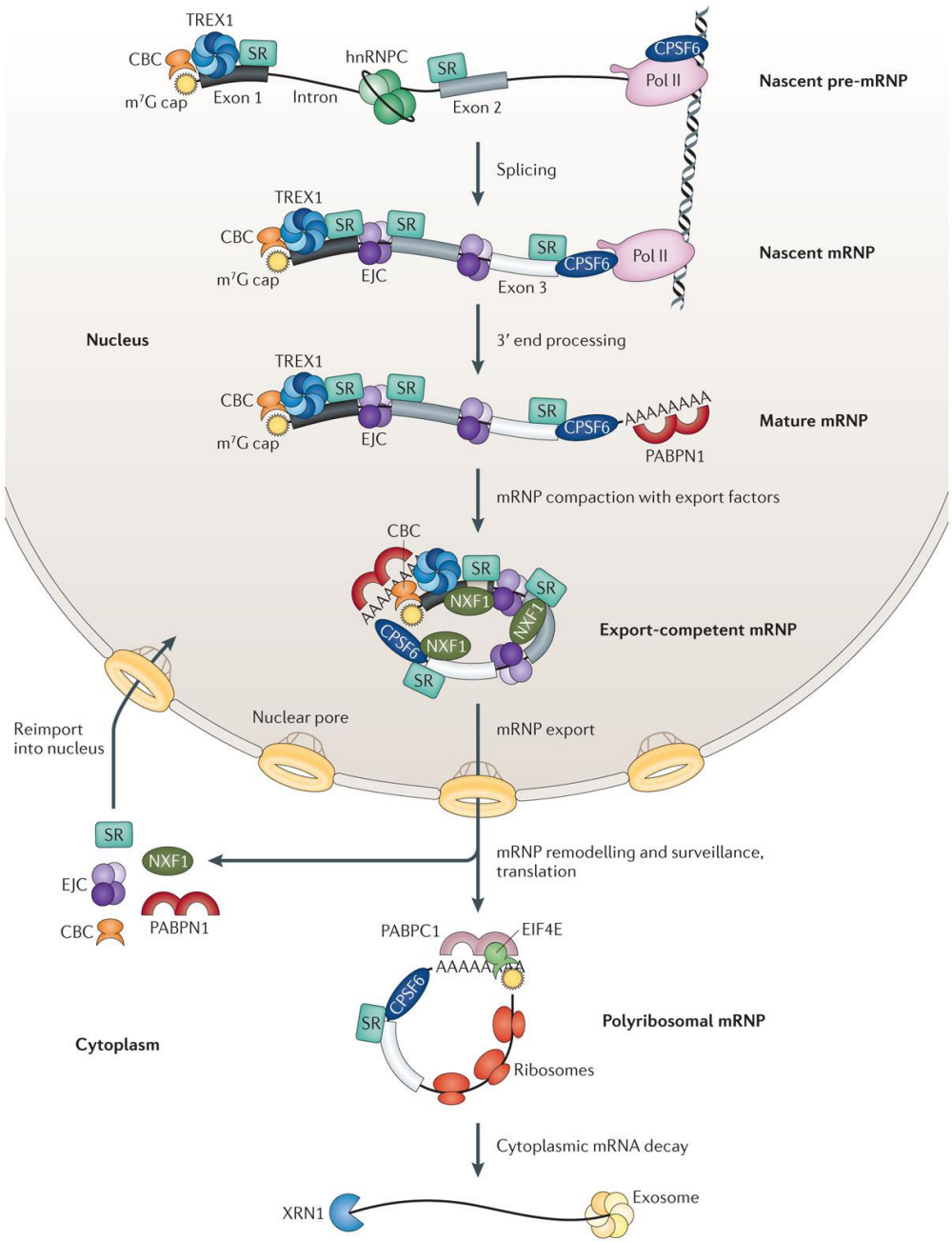


•The general RanGTP-dependent protein export receptor **CRM1** can be involved in the nuclear export of a subset of transcripts, such as mRNAs of several protooncogenes and cytokines, that contain AU-rich elements.

•CRM1 itself does not bind to RNA, instead recruiting NES-containing adaptor proteins that bind directly to RNA or to other RBPs. For example, AU-rich elements are recognized by RBP Hu-antigen R (HuR; also known as ELAVL1) and its protein ligands, which interact with CRM1

•CRM1 acts in the nuclear export of a number of unspliced and partially spliced viral mRNAs. These viral mRNAs can bind adaptor proteins that contain NESs (for example, HIV Rev, adenovirus E1b 55 kDa), thereby targeting the transport receptor CRM1.

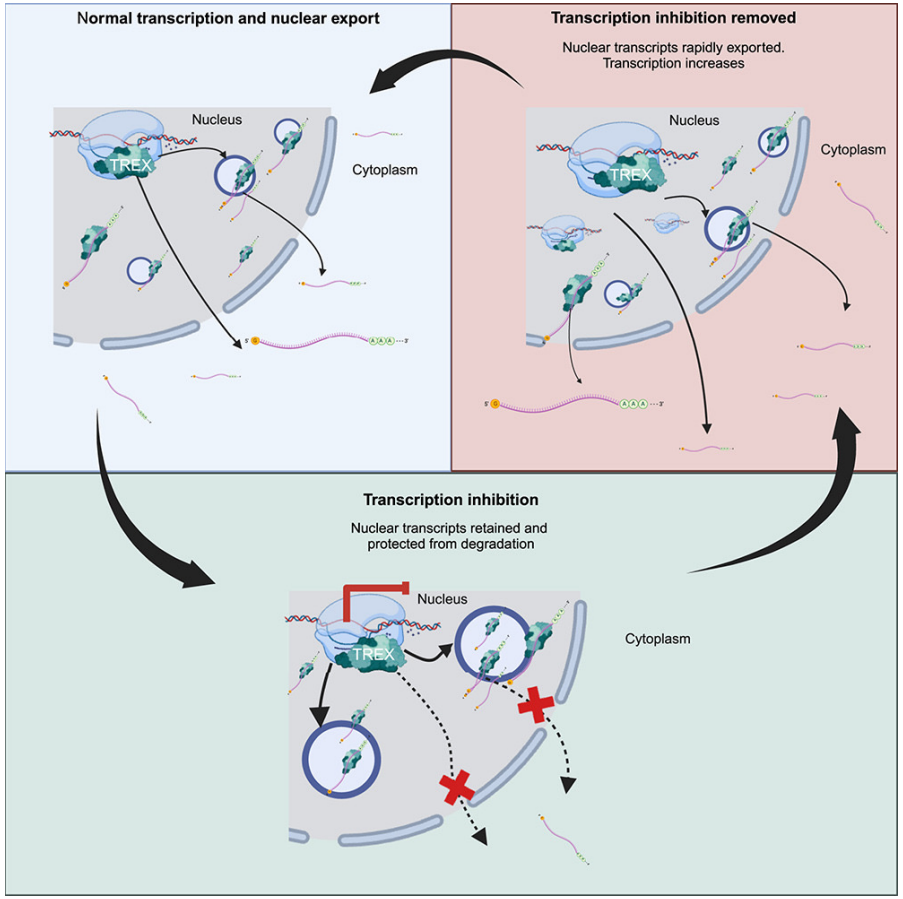
Messenger RNA (mRNA) is never alone



mRNA is coated and compacted by **RNA-binding proteins (RBPs)**, forming large messenger ribonucleoprotein particles (mRNPs). RBPs assemble on nascent and mature mRNAs. They serve as structural elements for mRNP packaging and modify the output of gene expression at all steps of the mRNA life cycle: transcription, splicing, 3′ end processing, capping, **nuclear export**, localization, translation and mRNA stability.

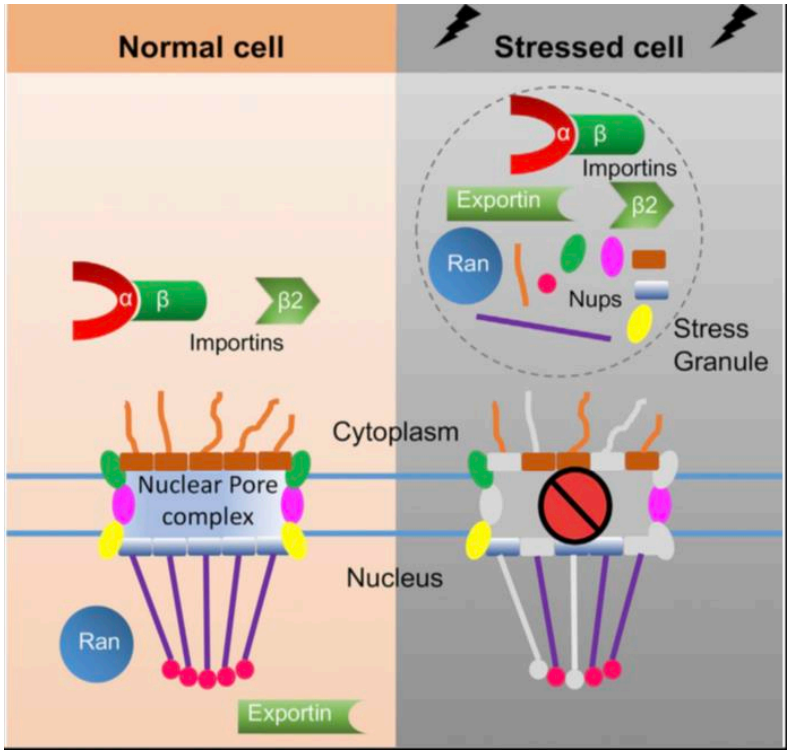
mRNA export factors store nascent transcripts within nuclear speckles as an adaptive response to transient global inhibition of transcription

Retained transcripts are fully processed and accumulate in proportion to the expression level of the genes from which they emanate. The TREX mRNA export complex plays an integral role in directing nascent transcripts to nuclear speckles where they are bound to NXF1, protected from degradation, and poised for rapid export following reinitiation of transcription



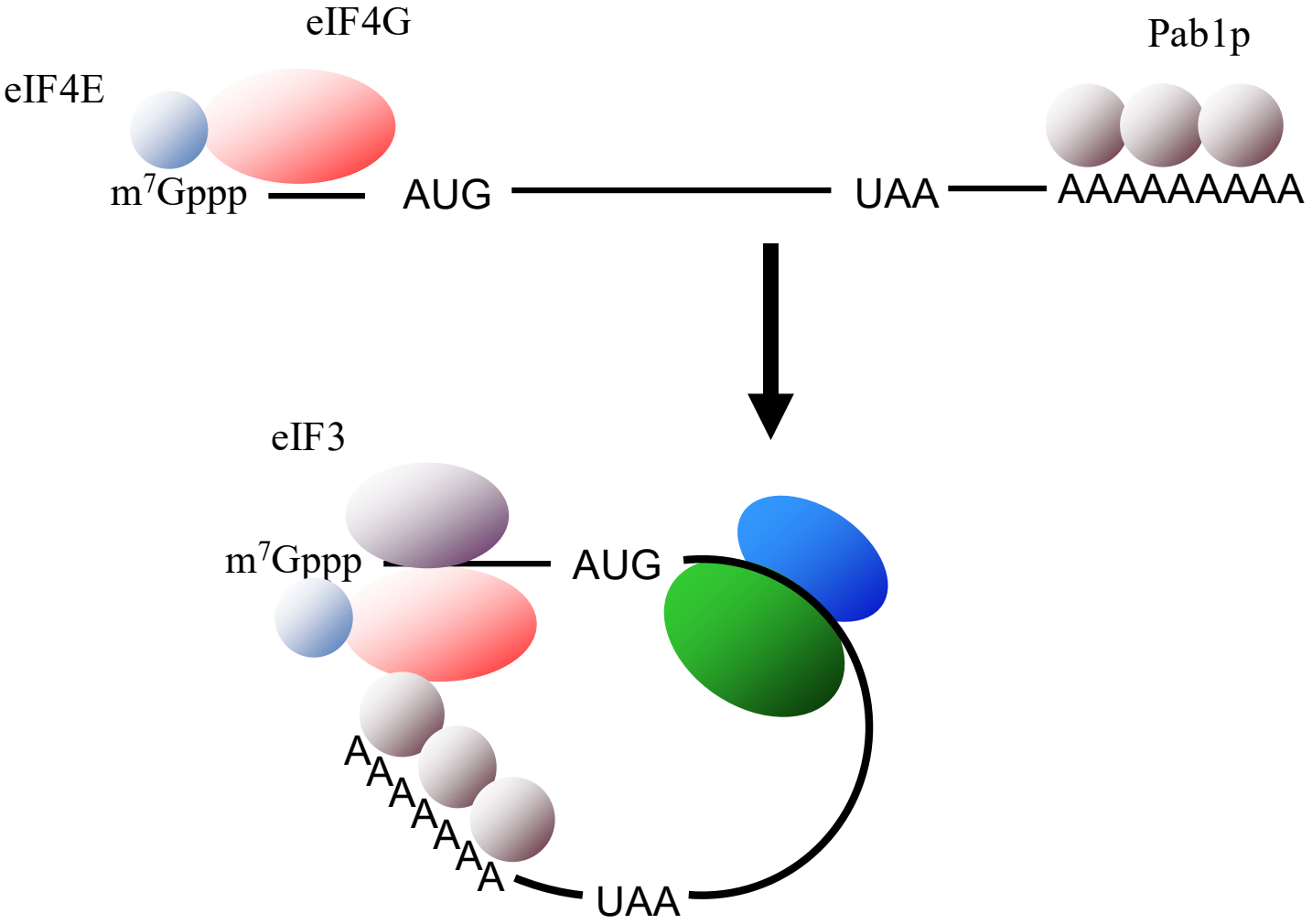
Neurodegeneration: problems at the nuclear pore

Defects in nucleocytoplasmic transport have been identified as a key pathogenic event in amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD) mediated by a GGGGCC hexanucleotide repeat expansion in *C9ORF72*, the most common genetic cause of ALS/FTD. Moreover, cellular stress disrupts nucleocytoplasmic transport by localizing critical nucleocytoplasmic transport factors into stress granules, RNA/protein complexes that play a crucial role in ALS pathogenesis. Importantly, inhibiting stress granule assembly, suppresses nucleocytoplasmic transport defects as well as neurodegeneration in C9ORF72-mediated ALS/FTD.



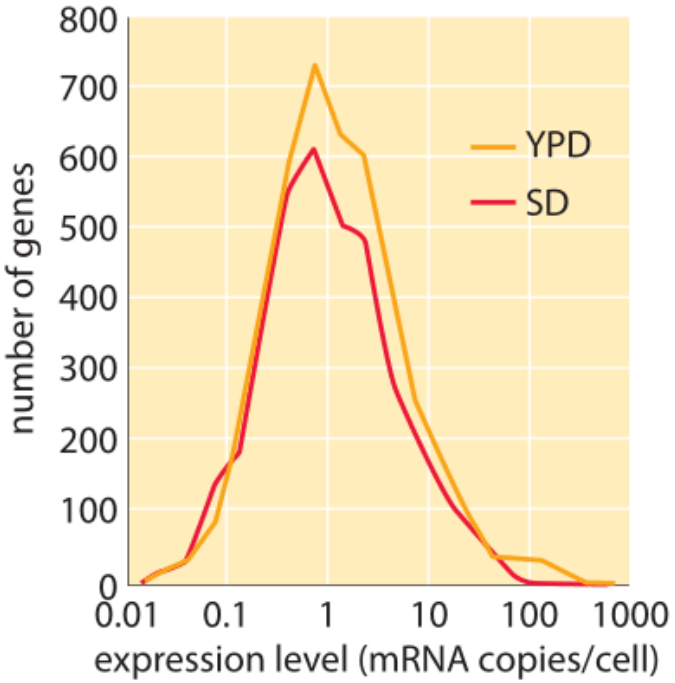
Zhang et al., 2018. Cell

mRNA


eIF4G
eIF4E
Pab1p
m7Gppp
AUG
UAA
AAAAAAAAA
eIF3
m7Gppp
AUG
AAAAAAA
UAA

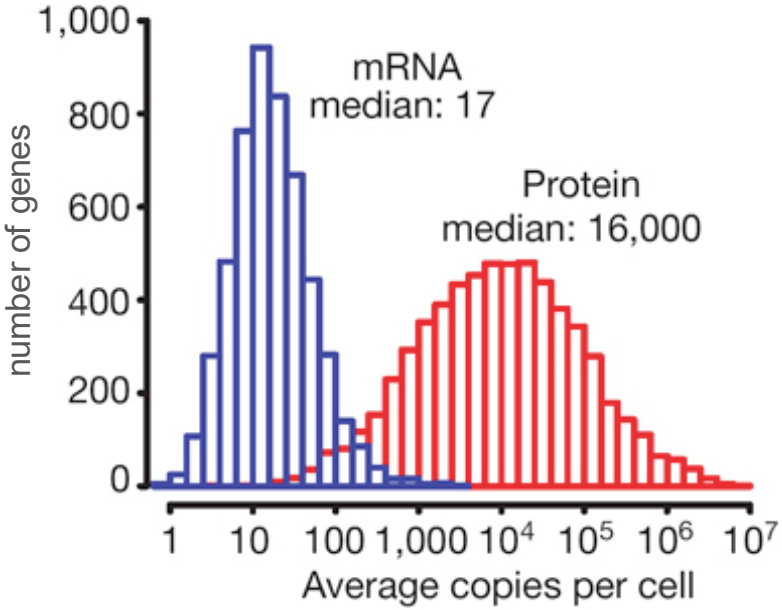
mRNA levels in eukaryotic cells

yeast



Marguerat et al., 2012 Cell

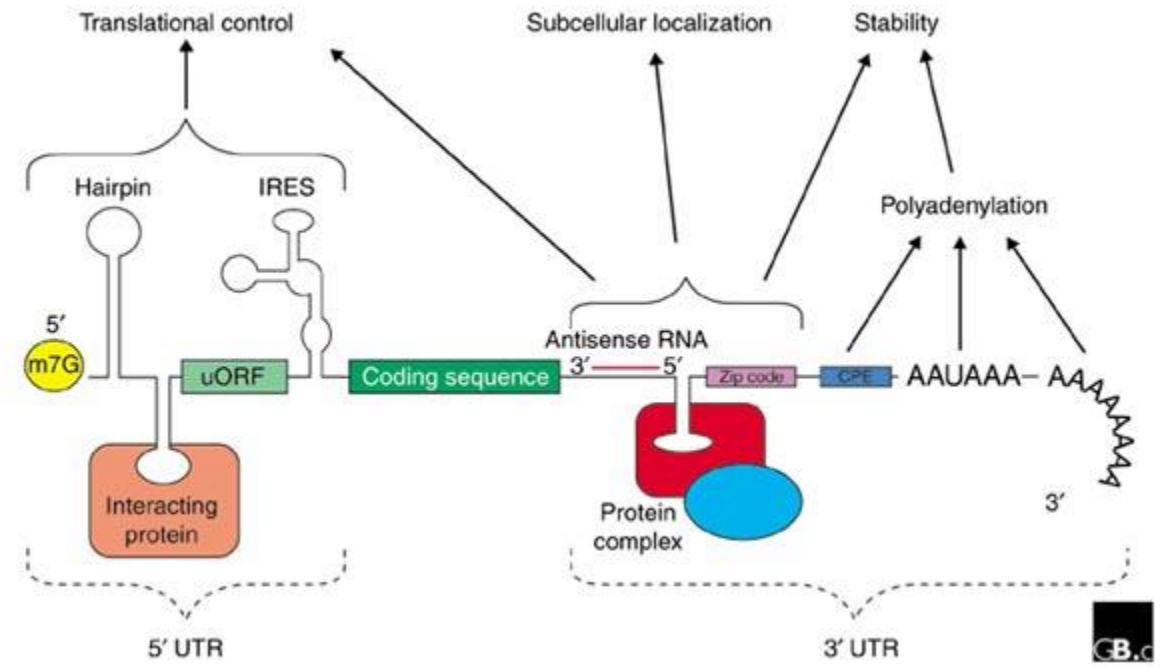
Mammalian cells



Schwanhäusser et al., 2011 Nature

Regulatory elements of mRNA

mRNAs contain several regulatory elements recognized by specific regulatory factors: **proteins** and **RNAs,** which allow fine-tuned regulation of gene expression.



Pathways by which eukaryotic mRNAs are degraded