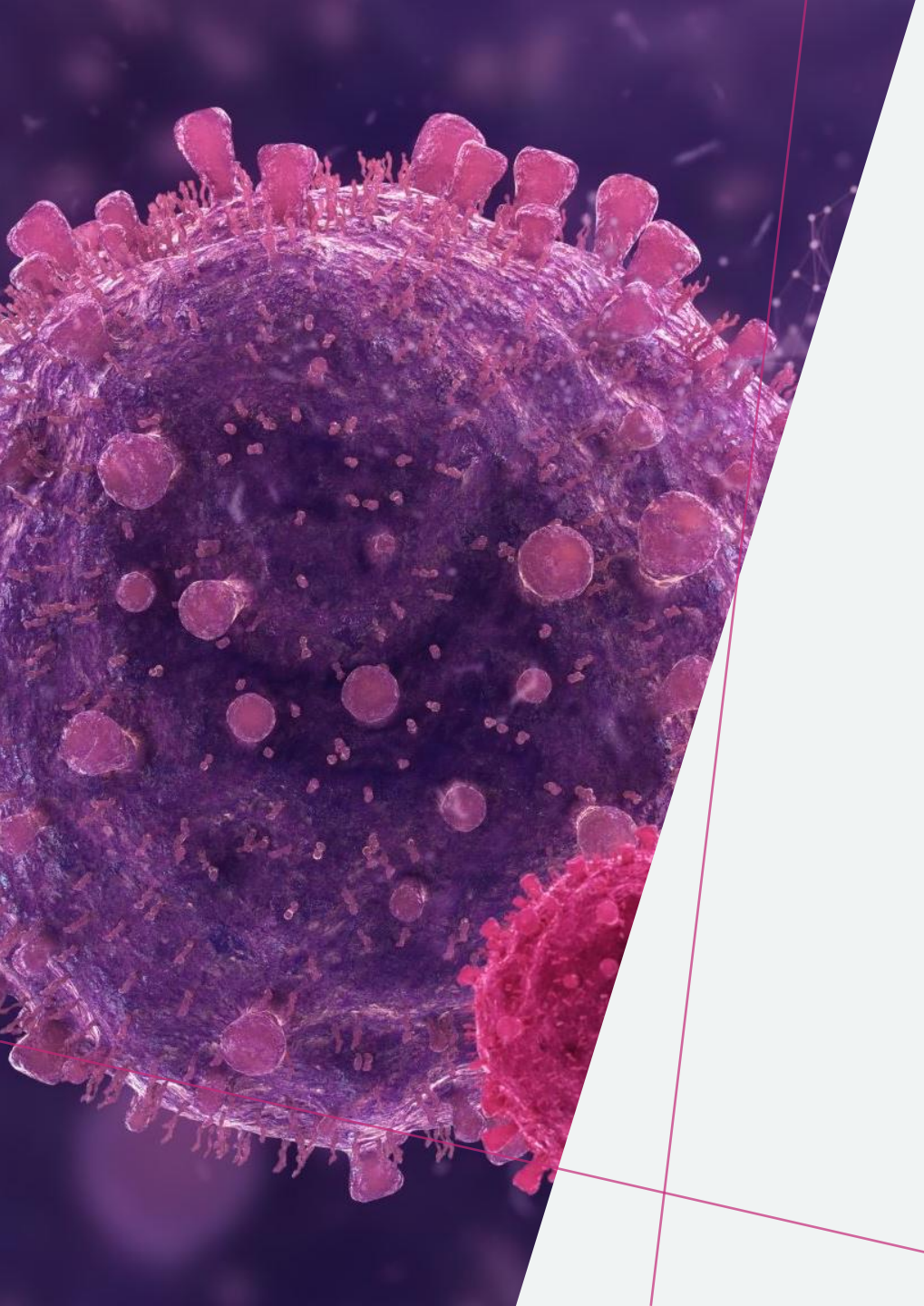
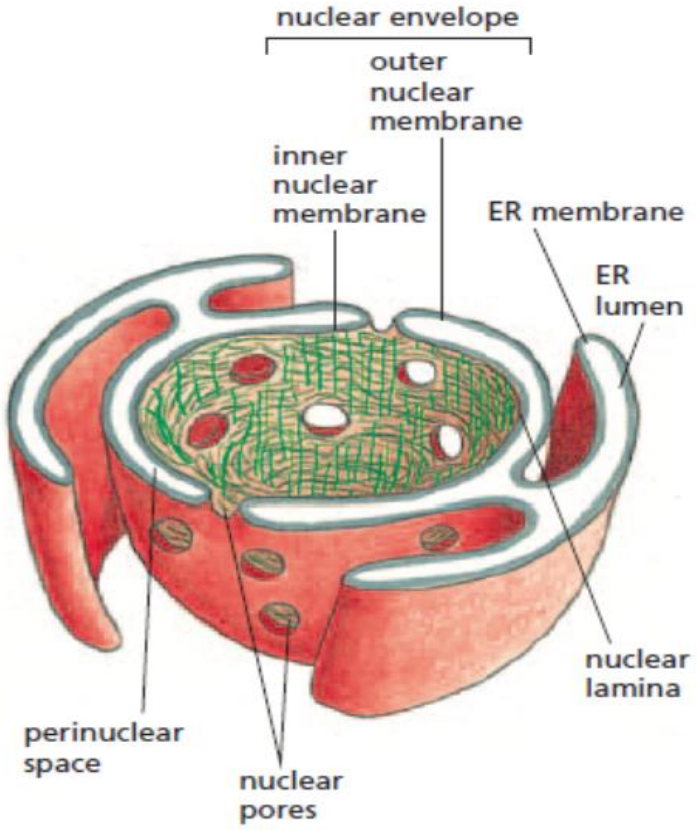


NUCLEAR PORE COMPLEX

STRUCTURE & FUNCTION

PROTEINS ENTER THE NUCLEUS THROUGH NUCLEAR PORES

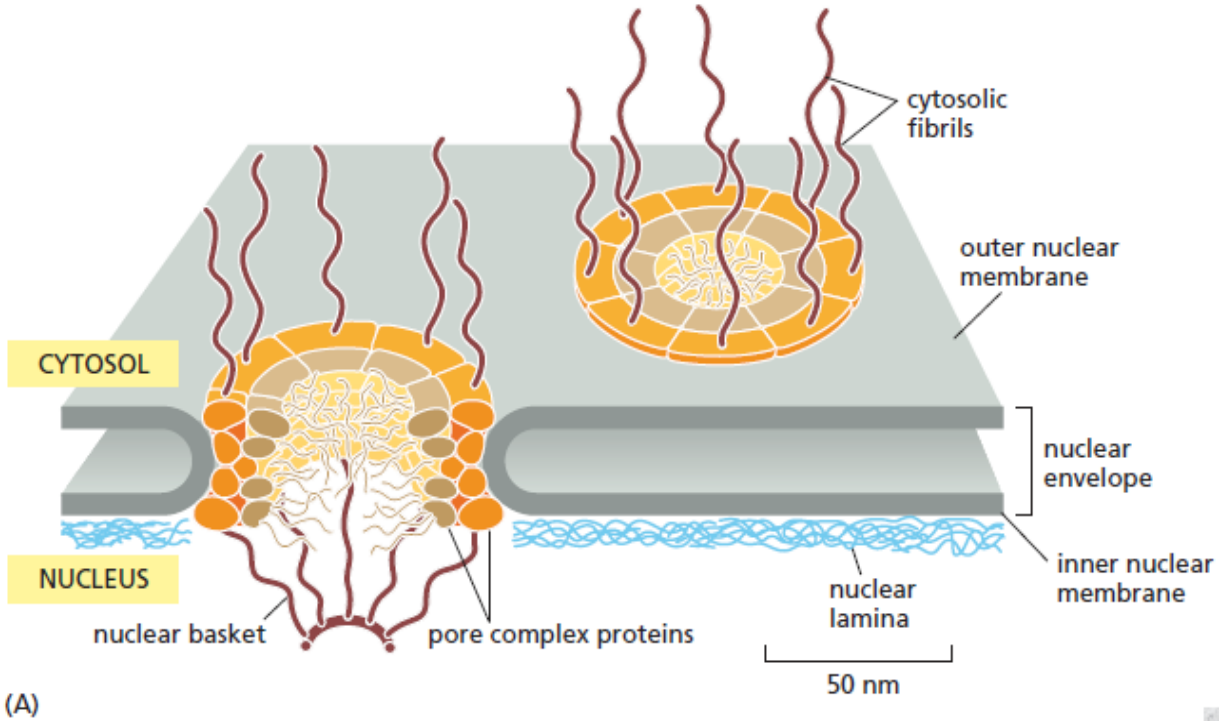
- The nuclear envelope, which encloses the nuclear DNA and defines the nuclear compartment, is formed from two concentric membranes.
- The ***inner nuclear membrane*** contains some proteins that act as binding sites for the chromosomes and others that provide anchorage for the ***nuclear lamina***, a finely woven meshwork of protein filaments that lines the inner face of this membrane and provides structural support for the nuclear envelope.
- The composition of the ***outer nuclear membrane*** closely resembles the membrane of the ER, with which it is continuous.



- The outer nuclear membrane is continuous with the ER membrane. The double membrane of the nuclear envelope is penetrated by nuclear pores. The ribosomes that are normally bound to the cytosolic surface of the ER membrane and outer nuclear membrane are not shown.

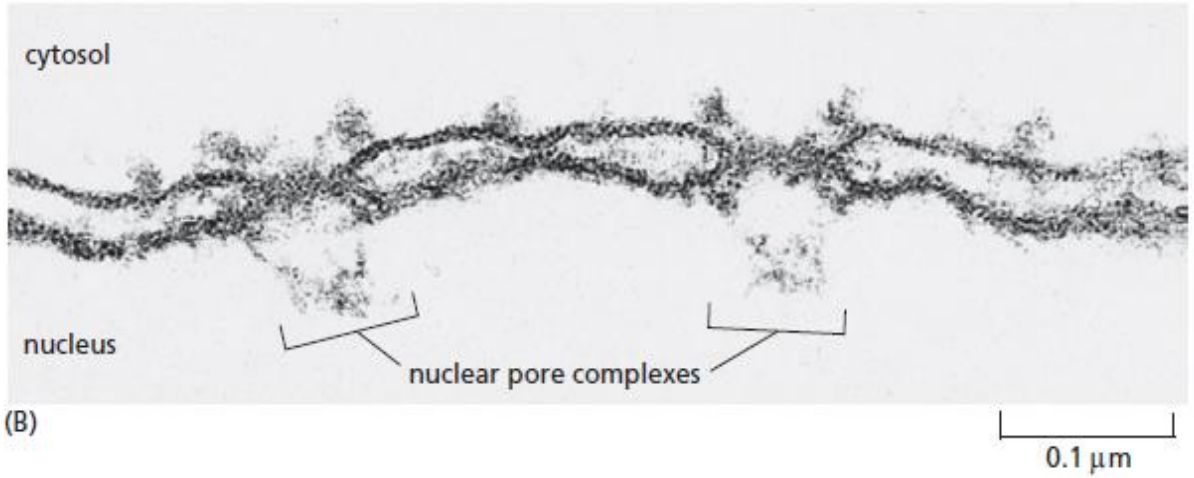
NUCLEAR PORE COMPLEX

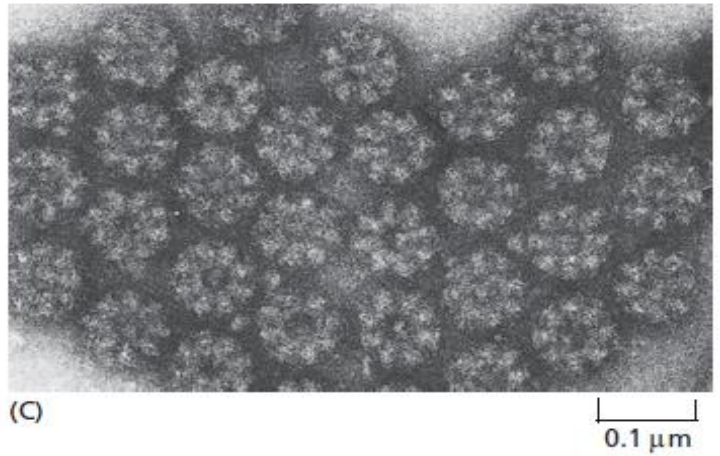
- The nuclear envelope in all eukaryotic cells is perforated by nuclear pores that form the gates through which molecules enter or leave the nucleus.
- A nuclear pore is a large, elaborate structure composed of a complex of about 30 different proteins.
- Many of the proteins that line the nuclear pore contain extensive, unstructured regions in which the polypeptide chains are largely disordered.
- These disordered segments form a soft, tangled meshwork—that fills the center of the channel, preventing the passage of large molecules but allowing small, water-soluble molecules to pass freely and non-selectively between the nucleus and the cytosol.



- The nuclear pore complex forms a gate through which selected macromolecules and larger complexes enter or exit the nucleus.
- (A) Drawing of a small region of the nuclear envelope showing two pores. Protein fibrils protrude from both sides of the pore complex; on the nuclear side, they converge to form a basketlike structure. The spacing between the fibrils is wide enough that the fibrils do not obstruct access to the pores.

ELECTRON MICROGRAPH





- (B) Electron micrograph of a region of nuclear envelope showing a side view of two nuclear pores (brackets).
- (C) Electron micrograph showing a face-on view of nuclear pore protein complexes; the membranes have been extracted with detergent.

(B, courtesy of Werner W. Franke; C, courtesy of Ron Milligan.)

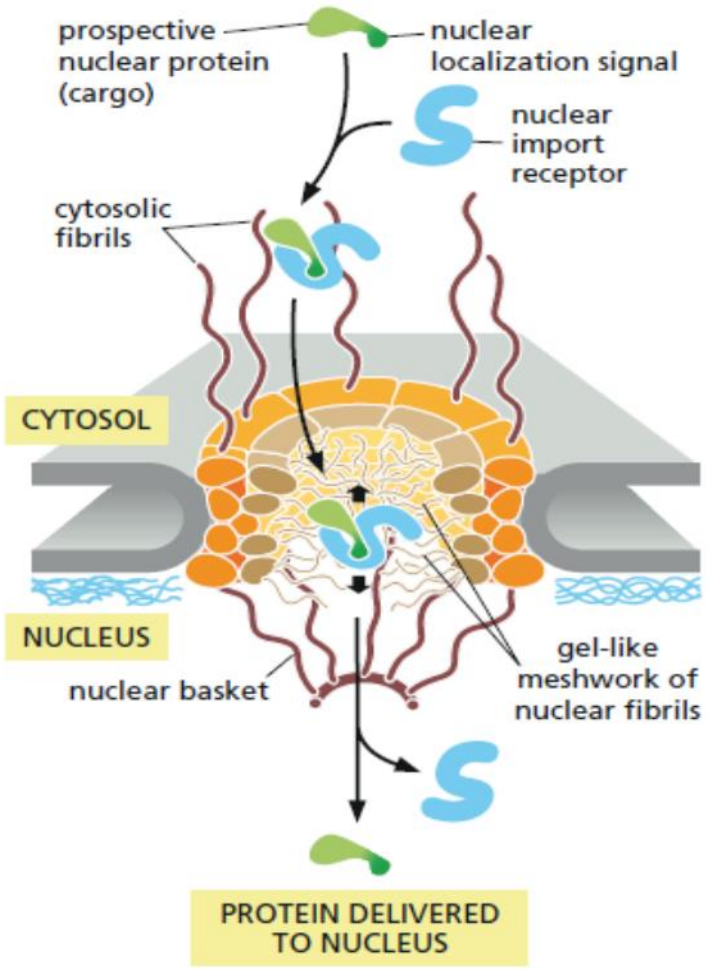
NUCLEAR LOCALIZATION SIGNAL

- Selected larger molecules and macromolecular complexes need to pass through nuclear pores. RNA molecules, which are synthesized in the nucleus, and ribosomal subunits, which are assembled there, must be exported to the cytosol. And newly made proteins that are destined for the nucleus must be imported from the cytosol.
- To gain entry to a pore, these large molecules and macromolecular complexes must display an appropriate sorting signal. The signal sequence that directs a protein from the cytosol into the nucleus, called a ***nuclear localization signal***, typically consists of ***one or two short sequences containing several positively charged lysines or arginines***.

Import into nucleus	-Pro-Pro-Lys-Lys-Lys-Arg-Lys-Val-
Export from nucleus	-Met-Glu-Glu-Leu-Ser-Gln-Ala-Leu-Ala-Ser-Ser-Phe-

NUCLEAR IMPORT RECEPTORS

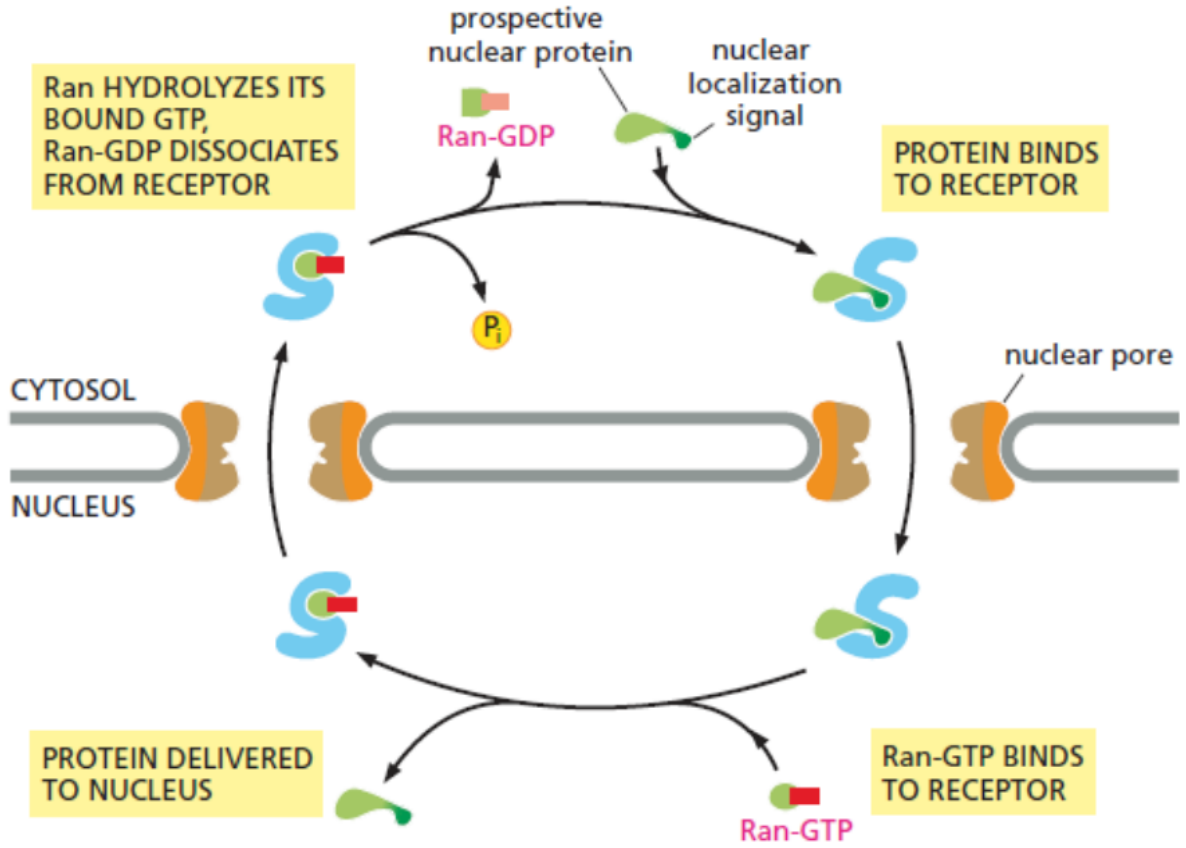
- The nuclear localization signal on proteins destined for the nucleus is recognized by cytosolic proteins called ***nuclear import receptors***.
- **These receptors help direct a newly synthesized protein to a nuclear pore by interacting with the tentacle-like fibrils that extend from the rim of the pore into the cytosol**.
- Once there, the nuclear import receptor penetrates the pore by grabbing onto short, repeated amino acid sequences within the tangle of nuclear pore proteins that fill the center of the pore.
- When the nuclear pore is empty, these repeated sequences bind to one another, forming a loosely packed gel. Nuclear import receptors interrupt these interactions, and they open a local passageway through the meshwork. The import receptors simply bump along from one repeat sequence to the next, until they enter the nucleus and deliver their cargo. The empty receptor then returns to the cytosol via the nuclear pore for reuse.



- Prospective nuclear proteins are imported from the cytosol through nuclear pores. The proteins contain a nuclear localization signal that is recognized by nuclear import receptors, which interact with the cytosolic fibrils that extend from the rim of the pore. As indicated by the short *black* arrows, after being captured, the receptors move randomly with their cargo through the gel-like meshwork of nuclear fibrils, until nuclear entry triggers cargo release. After cargo delivery, the receptors return to the cytosol via nuclear pores for reuse. Similar types of transport receptors, operating in the reverse direction, export mRNAs from the nucleus. These sets of import and export receptors have a similar basic structure.

ROLE OF RAN PROTEIN

- Like any process that creates order, the import of nuclear proteins requires energy. In this case, the energy is provided by the hydrolysis of GTP, mediated by a monomeric GTPase named Ran. This GTP hydrolysis drives nuclear transport in the appropriate direction. Nuclear pore proteins operate this molecular gate at an amazing speed, rapidly pumping macromolecules in both directions through each pore.
- Nuclear pores transport proteins in their fully folded conformation and ribosomal components as assembled particles. This feature distinguishes the nuclear transport mechanism from the mechanisms that transport proteins into most other organelles.



- ❑ Energy supplied by GTP hydrolysis drives nuclear transport.
- ❑ A nuclear import receptor picks up a prospective nuclear protein in the cytosol and enters the nucleus.
- ❑ There it encounters a small monomeric GTPase called Ran, which carries a molecule of GTP.
- ❑ This Ran-GTP binds to the import receptor, causing it to release the nuclear protein.
- ❑ Having discharged its cargo in the nucleus, the receptor—still carrying Ran-GTP—is transported back through the pore to the cytosol.
- ❑ There, an accessory protein (not shown) triggers Ran to hydrolyze its bound GTP.
- ❑ Ran-GDP falls off the import receptor, which is then free to bind another protein destined for the nucleus.
- ❑ A similar cycle operates to export mRNAs and ribosomal subunits from the nucleus into the cytosol, using nuclear export receptors that recognize nuclear export signals.