XII. ENDOCYTOSIS

A. PHAGOCYTOSIS IS THE INGESTION OF LARGE PARTICLES

1) protozoa

2) mammalian cells

   a) macrophages

   b) neutrophils

3) function of mammalian phagocytic cells

   a) defense against microorganisms

   b) scavenge damaged cells and debris

4) phagosomes

5) phagocytosis is a triggered process

6) role of receptors in phagocytosis

   a) fc receptors recognize antibody

   b) some receptors can recognize complement

   c) some receptors can recognize oligosaccharides

B. PINOCYTOSIS IS THE INGESTION OF FLUIDS AND SOLUTES VIA SMALL VESICLES

- types of vesicles, proteins, and organelles involved

   a) clathrin coated pits

   b) clathrin coated vesicles

   c) early endosomes

   d) late endosomes

C. RECEPTOR MEDIATED ENDOCYTOSIS

1) Import of cholesterol

   a) steps involved in cholesterol import

      i) LDL receptors are inserted in the membrane

ii) Fusion of LDL receptors to clathrin coated pits

iii) Internalization of membrane receptors

iv) clathrin coats are shed

v) early endosomes - receptors are recycled

vi) cholesterol proceeds to late endosomes

vii) maturation into lysosomes

viii) meanwhile, lysosomal hydrolases come from the golgi

ix) hydrolysis of cholesterol esters to form free cholesterol

x) LDL receptor is returned to the membrane

xi) cholesterol becomes available to the cell

b) diseases involving defects in LDL receptors

i) defect in the gene for LDL receptor

ii) receptor is lacking, or defective

iii) deficient in the capacity to take up LDL from the blood

iv) resulting high levels of blood cholesterol

v) atherosclerosis, heart attacks

2) <u>other types of receptor mediated endocytosis</u>

a) transferrin

i) a protein that carries iron from the blood

ii) cell surface transferrin receptor

iii) delivers transferrin bound to iron to early endosomes

iv) iron is released from transferrin

v) apotransferrin remains bound to the receptor

vi) apotransferrin is recycled to the cell surface

b) EGF receptor

i) EGF binds to cell surface receptors

ii) intracellular signaling is initiated

iii) EGF receptors bind coated pits only after binding EGF

iv) most receptors end up in lysosomes together with EGF

v) EGF and receptors are degraded

vi) receptor downregulation

D. TRANSCYTOSIS

1) transfer from one membrane to another in polarized cells

2) example, newborn rats receiving antibodies from mothers milk

a) ingestion of milk

b) antibodies bind receptor on the apical side of gut cells

c) internalization via clathrin coated pits

d) vesicles are delivered to early endosomes

e) vesicles bud and fuse with basolateral membrane

f) dissociation of antibodies

g) entry into the bloodstream

h) (in humans the antibodies remain in the gut)

E. ENDOCYTOSIS IN POLARIZED CELLS

1) substances enter the cell from both membranes

2) two distinct early endosomes

XIV. LIPID BIOSYNTHESIS IN THE SMOOTH ER

A) PRODUCTION OF LIPIDS

B) PRODUCTION OF PHOSPHATIDYLCHOLINE

1) produced from choline, two fatty acids, and glycerol phosphate

2) enzyme for catalysis are contained within ER membranes

a) active sites face the cytosol

b) required metabolites are in the cytosol

c) phospholipid synthesis occurs exclusively on the cytosolic side

3) steps involved

a) insertion into the membrane

b) determination of the head group

C) PRODUCTION OF OTHER MEMBRANE PHOSPHOLIPIDS

- PE, PS, and PI are formed in the same way

D) PHOSPHOLIPID TRANSLOCATORS - FLIPPASE

1) head group specific

2) in ER a flippase specifically carries choline containing lipids

3) responsible for asymmetric distribution

E) PRODUCTION OF CERAMIDE AND SPHINGOMYELIN

1) condensing serine with a fatty acid

2) formation of sphingosine

3) second fatty acid is added to form ceramide

4) transported to the golgi

5) transfer of phosphocholine head groups - sphingomyelin

F) FORMATION OF CHOLESTEROL

XV. Protein transport into and out of the nucleus

A) Nucleus houses the genome

B) Presence of a nucleus allows regulated gene expression

D) Nuclear lamina underlies the inner membrane

1) fibrous network

2) provides structural support

3) composed of lamins

a) 4 types of lamins

b) fibrous proteins related to intermediate filaments

c) the lamins form filaments

d) The lamins bind integral membrane proteins

e) the lamins provide structural support for the nucleus

f) The lamins are sites for chromatin attachment

E) Nuclear matrix

1) structural framework similar to the cytoskeleton

2) may serve to organize and anchor functional domains

3) its existence is still not certain

4) its molecular composition is not yet defined

F) Transport of molecules in and out of the nucleus

1) inner and outer nuclear membranes differ

a) inner membrane contains proteins that bind nuclear lamina

b) outer membrane resembles ER membrane

2) Bi-directional traffic occurs in and out of nucleus

a) some things move into the nucleus

b) others, such as RNAs, move out of the nucleus

c) Ribosomal proteins go into and then out of the nucleus

3) nuclear pores

a) protein complex of more than 100 proteins

b) contains one or more aqueous channels

c) experiments to test the size of the channels

b) pores are permeable only to small molecules

4) nuclear localization signal

a) usually rich in positively charged amino acids and proline

c) can be located anywhere

b) NLSs were first identified on the T ag encoded by SV40

5) proposed mechanism by which proteins enter the nucleus

a) role of receptors

b) opening in the pore changes size

c) proteins are fully folded

d) NLS is not cleaved off

e) nuclear proteins need to be able to re-enter the nucleus

f) nuclear import uses a pore

6) nuclear import of protein is an important part of gene regulation

7) breakdown and reformation of nuclear envelope during mitosis

a) nuclear lamins get phosphorylated

b) nuclear lamina depolymerize

c) nuclear pore complexes disassemble at the same time

d) nuclear envelope breaks up into vesicles

e) vesicles and nuclear contents disperse

f) nuclear lamins get dephosphorylated

g) nuclear lamina repolymerizes

h) lamina bind the nuclear vesicles which fuse with each other

i) enveloped chromosomes come together

j) nuclear pore complexes reassemble

8) transport between cytoplasm and nucleus can be regulated

G) Nucleolus

1) Contain no membranes

2) sites of rRNA transcription and processing for ribosome assembly

a) ribosomes contain 5S, 5.8S, 18S and28S rRNA

b) nucleolus is organized around rRNAs.

c) 5.8S, 18S, and 28S are transcribed as a single 45S precursor

d) (transcription of 5S RNA takes place outside the nucleolus)

3) nucleoli consist of three distinguishable regions

a) fibrillar center

b) dense fibrillar component

d) granular component

4) steps involved in ribosomal assembly

a) transcription of rRNA genes in the nucleolus

b) transcription of genes for ribosomal proteins outside the nucleolus

c) mRNA for ribosomal proteins are translated in the cytoplasm

d) ribosomal proteins are imported into the nucleolus

e) ribosomal proteins assemble on the pre rRNA

f) processing of the rRNA with attached proteins

g) formation of preribosomal particle

h) export of the 2 subunits into the cytoplasm

5) RNA export from the nucleus also requires a signal

a) the signal in mRNA is a 5' CAP

b) tRNA and rRNA assemble with proteins

XVI. Transport of proteins into the mitochondria and peroxisomes

A) Structure of the Mitochondria

1) double membrane

2) intermembrane space

3) Inner membrane folds to form cristae

a) contains enzymes

b) impermeable to most things

4) The interior of the organelle is the matrix

a) contains enzymes

b) contains mitochondrial genetic system

5) outer membrane contains porins

- large aqueous channels - permeable to most things

7) Mitochondria contain DNA and ribosomes

8) most of the proteins are imported from the cytoplasm.

B) Protein transport into the Mitochondria

1) mitochondrial precursor proteins have signal peptides

a) signal peptides are diverse in sequence

b) usually 20-80 amino acids long

c) at the amino terminus

d) signal peptides probably bind receptors

e) cleaved by peptidases

2) Steps involved in transport of proteins into mitochondria

a) signal peptide, receptor on the outer membrane

b) proteins pass through both membranes at once

c) signal peptidase cleaves the peptide

3) Role of chaperonins

a) cytoplasmic hsp70 binds newly synthesized proteins

b) hsp70 is released after ATP hydrolysis

c) mitochondrial hsp70 receives the proteins once they enter

d) mhsp70 releases the protein in an ATP dependent step

e) proteins are pulled through the matrix

f) mhsp60 attaches to the protein and facilitates folding

4) Some proteins are destined for inner membrane or intermediate space via 3 potential pathways

a) second signal sequence

b) stop transfer pathway

c) some proteins are released into the intermediate space

C) Transport of proteins into peroxisomes requires short signal sequences

1) peroxisomes have a single membrane

2) proteins often have a 3 aa sequence near the C terminus

3) A receptor protein recognizes the signal sequence