SNS COLLEGE OF ENGINEERING

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B.E. - Electronics and Communication Engineering

Fifth Semester

19EC502 – Transmission Lines and Antennas

Regulations 2019

UNIT-I

INTRODUCTION TO LIFE

Prokaryotic and Eukaryotic Cells

Introduction- Higher eukaryotes have multiple organs to perform specific functions such as liver, kidney and heart. Each Organ has specific tissue and each tissue is composed of cells. **"Cell is the structural and functional unit of life"** and it contains all necessary infrastructure to peform all functions. Based on cellular structure, cells are classified as prokaryotic and eukaryotic cells. In most of the cases, prokaryotes are single cells where as eukaryotes are either single cells or part of multicellular tissues system. Besides this, both types of cells have several structural and metabolic differences as given in Table 3.1 and are discussed later in the lecture.

TABLE 3.1 DIFFERENCE BETWEEN PROKARYOTIC AND EUKARYOTIC CELLS

Feature	Prokaryote	Eukaryote
Size	Small, in µm range	Variable size, upto 40µm in diameter.
Genetic material	Circular DNA present in cytosol as free material	DNA in the form of linear chromosome present in well defined double membrane nucleus, no direct connection with cytosol
Replication	Single origin of replication	Multiple origin of replication.

Genes	No Intron	Presence of Intron
Organelles	No membrane bound organelles	Membrane bound orgelles with well defined function.
Cell walls	Very complex cell wall	Except Fungi and plant, eukaryotic cells are devoid of a thick cell wall.
Ribosome	70S	80S
Trancription and translation	Occurs together	Transcription in nucleus and translation in cytosol

Structure of Prokaryotic cells- A prokaryotic cell is much simpler and smaller than eukarotic cells. It lacks membrane bound organelles including nucleus. A typical prokaryotic cells is shown in Figure 3.1, A. The description of different structural feature of prokaryotic cells is as follows-

1. Outer Flagella: A flagellum attached to the bacterial capsule is a central feature of most of the prokaryotic cell especially of the motile bacteria. It provides motion or locomotion to the bacteria and be responsible for chemotaxsis of bacteria. Movement of bacteria towards a chemical gradient (such as glucose) is known as chemotaxsis. Flagellum is a part of cell wall and its motion is regulated by motor proteins present inside the cell. Flagellar motion is an energy consuming process and it is governed by an ATPase present at the bottom of the shaft. It is made up of protein flagellin and reduction or suppression of flagellar protein reduces bacterial infectivity (pathogenicity) and ability to grow.

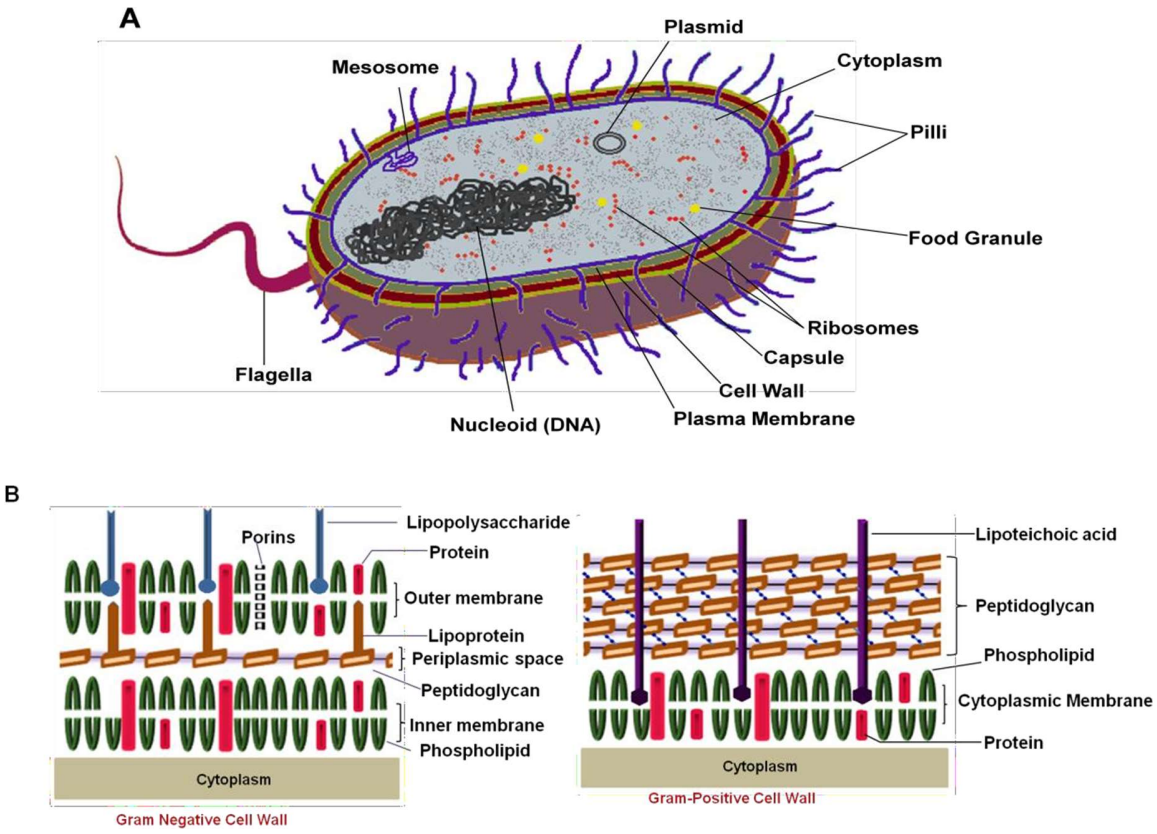

Figure 3.1: Structural details of a typical prokaryotic cell. (A) Whole cell and (B) composition of cell wall of gram negative and positive bacteria.

2. Bacterial surface layers: Bacteria posses 3 anatomical barriers to protect the cells from external damage. Bacterial capsule is the outer most layer and made up of high molecular weight polysaccharides. It is impermeable to the water or other aqueous solvent and it is responsible for antigenicity of bacterial cells. Cell wall in bacteria and its response to gram staining is the basis of classification of bacterial species.

WHAT IS GRAM STAINING? Gram staining is developed by a Danish scientist Hans Christian Gram. This technique differentiates bacterial strains based on their cell wall composition, especially thickness of peptidoglycan layer. A detail staining procedure is given in following paper (**Use of the gram stain in microbiology. Beveridge, TJ (2001) *Biotech Histochem* 76 (3): 111–8. Pubmed ID: 11475313**). During the staining procedure bacterial sample is stained with two dyes, **crystal violet** and **safarin**. During a washing step with non-polar solvents such as alcohol or acetone (decolorization), gram –ve bacteria leave the **blue** stain due to a thin peptidoglycan layer in cell wall whereas gram +ve bacteria retains both stains and appear as **Pink**.

Cell wall composition in gram-ve and gram +ve bacteria is different. Bacterial cell wall has different constituents and be responsible for their reactivity towards gram stain.

A. Peptidoglycan layer: peptidoglycan layer is thick in gram +ve bacteria and thin in gram –ve bacteria. Peptidoglycan is a polymer of NAG (N-acetyl-glucosamine) and NAM (N-acetyl-muramic acid) linked by a β-(1,4) linkage. Sugar polymer are attached to peptide chain composed of amino acids, L-alanine, D-glutamic acid, L-lysine and D-alanine. Peptide chain present in one layer cross linked to the next layer to form a mesh work and be responsible for physical strength of the cell wall. Peptidoglycan synthesis is targeted by antibiotics such as pencillin where as lysozyme (present in human saliva or tears) degrades the peptidoglycan layer by cleaving glycosidic bond connecting NAG-NAM to form polymer.

B. Lipoteichoic acids: Lipoteichoic acid (LTA) are only found in gram +ve bacteria cell wall and it is an important antigenic determinant.

C. Lipopolysaccharides (LPS)- Lipopolysaccharides (LPS) are found only in gram –ve bacterial cell wall and it is an important antigenic determinant.

3. Cytosol and other organelles-Prokaryotic cells do not contain any membrane bound organelle. The organelles are present in cytosol such as ribosome (70S), genetic material where as electron transport chain complexes are embedded within the plasma membrane.

4. Chromosome and extra chromosomal DNA-Prokaryote cell contains genetic material in the form of circular DNA, known as "**bacterial chromosome**". It contains genetic elements for replication, transcription and translation. Bacterial chromosome follows a rolling circle mode of DNA replication. The genes present on chromosome does not contains non coding region (introns) and it is co-translated to protein. Besides main circle DNA, bacteria also contains extra chromosomal circular DNA known as "plasmid". Presence of plasmid containing resistance gene confers resistance towards known antibiotics. Exchange of extra-chromosomal DNA between different bacterial strains is one of the mechanisms responsible for spread of antibiotic resistance across the bacterial population. Details of plasmid and its structural features will be discussed in a later lecture.

Struture of Eukaryotic cell- The eukaryotic cell is much more complex and it contains many membrane bound organelles to perform specific functions. It contains a nucleus isolated from cytosol and enclosed in a well defined double membrane. A typical eukaryotic animal and plant cell is shown in Figure 3.2 and the difference between these types of cells is given in Table 3.2.

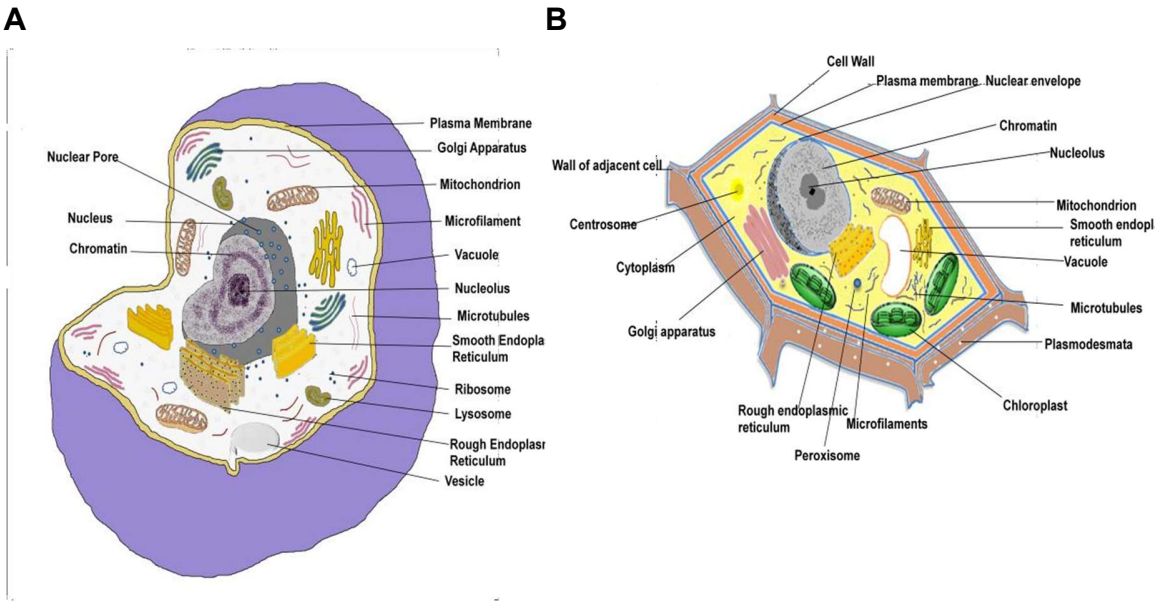

Figure 3.2 : Structure of Eukaryotic cell. (A) Animal Cell (B) Plant Cell

TABLE 3.2 DIFFERENCE BETWEEN ANIMAL AND PLANT CELLS

FEATURE	PLANT CELL	ANIMAL CELL
Cell wall	Present	Mostly absent
Size	Large	Comparatively small
Chlorophyll	Present	Absent
Vacuole	Large Central	Small and many in number
Mitochondria	Few	More
Lysosome	Almost absent	Present
Glyoxysomes	Present	Absent
Cytokinesis	By Plate method	By constriction

The description of different structural feature of eukaryotic cell is as follows-

Different organelles of Eukryotic cells (Animal)

1. Cytosol-Cytosol is the liquid part filled inside the cell and it contains water, salt, macromolecules (protein, lipid, RNA). It has an array of microtubule fiber running through out the cytosol to give vesicular structure to its destination. Besides this, cytosol exhibits “Sol” to “Gel” transition and such transition regulates multiple biochemical and cellular processes.

2. Nucleus-Nucleus is the central processing unit of cell and homologous to the processor in a typical computer (Figure 3.3, A). The liquid filled inside nucleus is called as **nucleoplasm**. It is a viscous liquid containing nucleotides and enzymes to perform replication, transcription, DNA damage repair etc. It contains genetic material (DNA) in a complex fashion involving several proteins (histones) to pack into nuclear bodies or chromosomes. The chromatin in eukarotic nucleus is divided into **euchromatin** or **heterochromatin**. Euchromatin is a part of chromatin where DNA is loosely packed and it is transcriptionally active to form mRNA where as Heterochromatin is more densily packed and it is transcriptionally inactive. Nuclei in eukarytotic cells are present in a double layer of membrane known as **nuclear envelope** (Figure 3.3, B). Outer membrane of nuclear envelope is continuous with the rough endoplasmic reticulum and has ribosome attached to it. The space between these two membranes is called as **perinuclear space**. Nuclear envelope often has **nuclear pore** and as per calculation an average nucleus has 3000-4000 pores per nuclear envelope.

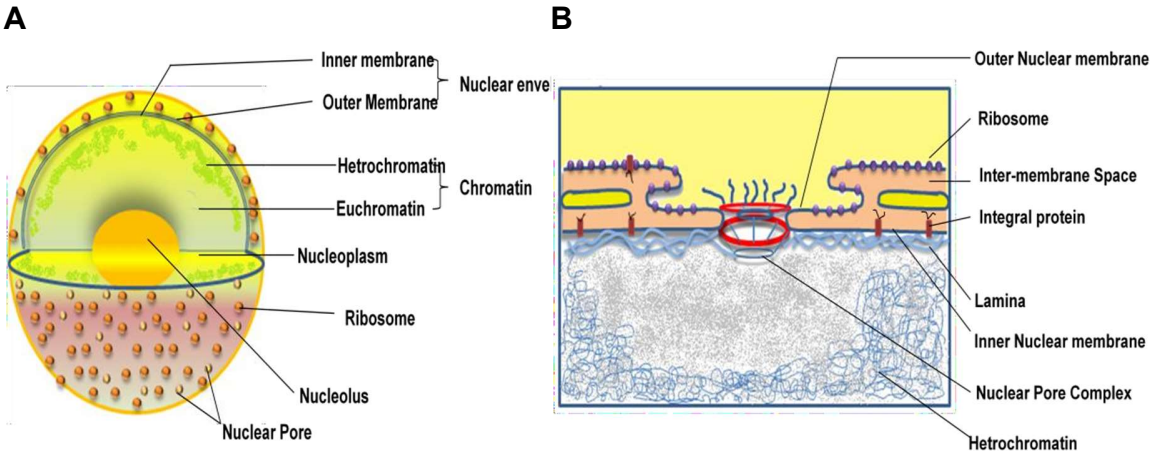

Figure 3.3: Structural details of nucleus. (A) whole and (B) enlarged view of nuclear pore.

Nuclear pore is 100nm is diameter and consists of several proteins. It is a gateway for transfer of material between nucleus and cytosol. RNA formed after transcription from DNA within the nucleus and move out of the nucleus into the cytosol through nuclear pore. Similarly protein from cytosol crosses nuclear pore to initiate replication, transcription and other processes.

Lecture 4: **Prokaryotic and Eukaryotic Cells (Part II)**

Summary of Previous Lecture: In the previous lecture we discussed the structure of prokaryotic cells, differences between prokaryotic and eukaryotic cells and lastly we started the discussion about the structure of eukaryotic cells. In continuation to previous lecture, in the current lecture we will discuss remaining cellular organelles of eukaryotic cells.

1. Mitochondria- It is popularly known as "**power house of the cell**" as the organelle is actively involved in the generation of ATP to run the cellular activities. Mitochondria is a double layered membrane-bound organelle with different structural properties (Figure 4.1, A). Outer membrane is smooth and cover the complete organelle with large number of integral proteins, known as **porins**.

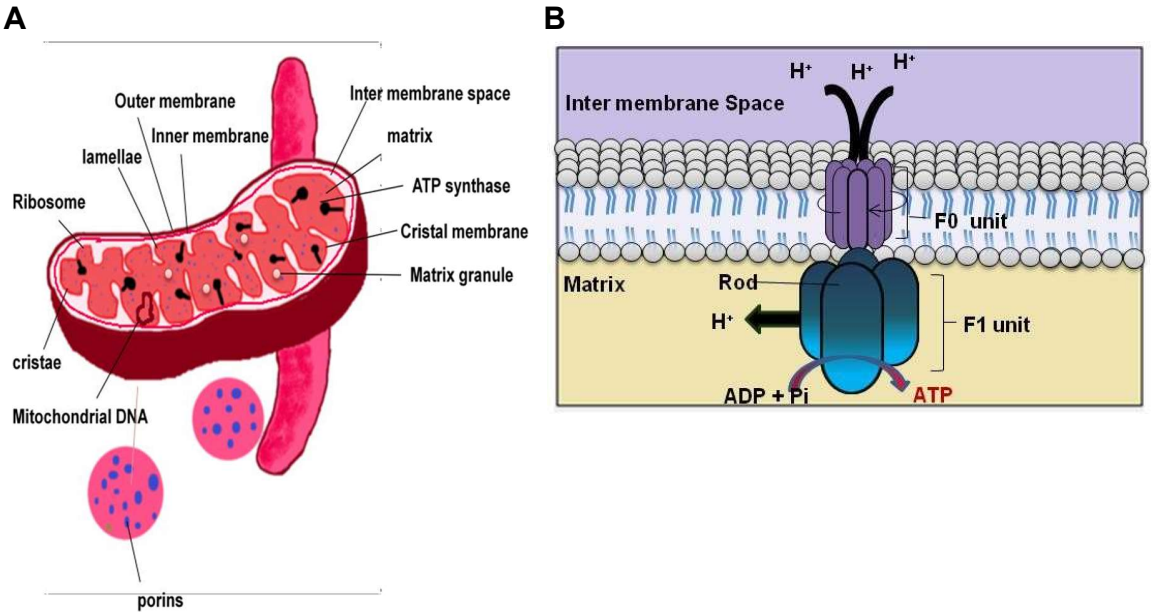

Figure 4.1: Mitochondria. (A) Struture of mitochondria and (B) enlarged view of ATP Synthase.

Porin allows free movement of molecules less than 5000da within and outside mitochondria. Where as larger molecules or proteins moves into the mitochondria through transporters involving signal peptides known as "**mitochondrial targeting sequence**". Inner membrane is folded into membrane projections to form **cristae**. Cristae occupies major area of membrane surface and house machinery for anaerobic oxdidation and electron transport chain to produce ATP. Due to presence of inner and outer membrane, mitochondria can be divided into 2 compartments: first in between the inner and outer membrane, known as **intermembrane space** and second inside the inner membrane known as **matrix**. The proteins present in intermembrane space have a role in executing "**programmed cell death**" or "**apoptosis**". Matrix is the liquid part present in the inner most compartment of the mitochondria and it contains ribosome, DNA, RNA, enzymes to run Kreb's cycle and other proteins. Mitochondrial DNA is circular and it has full machinery to synthesize its own RNA (mRNA, rRNA and t-RNA) and proteins. Marked differences exist between mitochondrial DNA and DNA present in nucleus and these differences are not discussed here due to space constrain. Electron transport chain components (complex I to complex V) are integral proteins, present in the inner membrane of mitochondria. During metabolic reactions such as glycolysis, Kreb's cycle [metabolic reaction are discussed later] produces large amount of reducing equivalent in the form of $NADH_2$ and $FADH_2$. Electron transport chain process reducing equivalent and flow of the electron through different complexes (Complex I to Complex IV) causes generation of proton gradient across the membrane. Proton expelled in the intermembrane space returned back to the matrix through complex V (ATP synthase) to generates ATP. ATP synthase (Figure 4.1, B) is a mushroom shaped multimeric protein complex, mainly composed of two proteins Fo and F1. Fo is a membrane bound portion where as F1 is the complex present into the lumen towards matrix. F_oF_1 complex of mitochondria harvest the proton motive force to catalyze phosphorylation reaction involving ADP and phosphate to generate ATP.

Functions of mitochondria-

1. Production of ATP

2. Generation of **Reactive Oxygen Species** (ROS) in immune cells to kill infectious agents.

3. Used to track tree of a family.

3. Role in programmed cell death or **"apoptosis"**

Apoptosis: Apoptosis is the programmed cell death involving a series of events involving cellular metalloprotease known as caspases. In an adverse event of exposure of cell to the cyto-toxic agent or environmental condition, it activates cell surface signaling to activate cytosolic caspases. In addition, it disturbs mitochondrial membrane potential to cause the release of CytC. Ultimately, these cellular events activates DNase activity within nucleus and degrade genominc DNA to cause cell death.

2. Chloroplast-Chloroplasts are found in plant, algae and other lower invertebrates such as euglena. Contrasting to mitochondria, chloroplast has outer membrane, an inner membrane and then light pigment containing inner most thylakoid membrane (Figure 4.2, A). Outer membrane is porous to the small molecules but protein or large molecules are transported by **TOC** (translocon on the outer chloroplast membrane) complex. Movement of material passed through outer membrane gets into the inner membrane through **TIC** (translocon on the inner chloroplast membrane) complex. In between outer and inner membrane is intermembrane space filled with aqueous liquid.

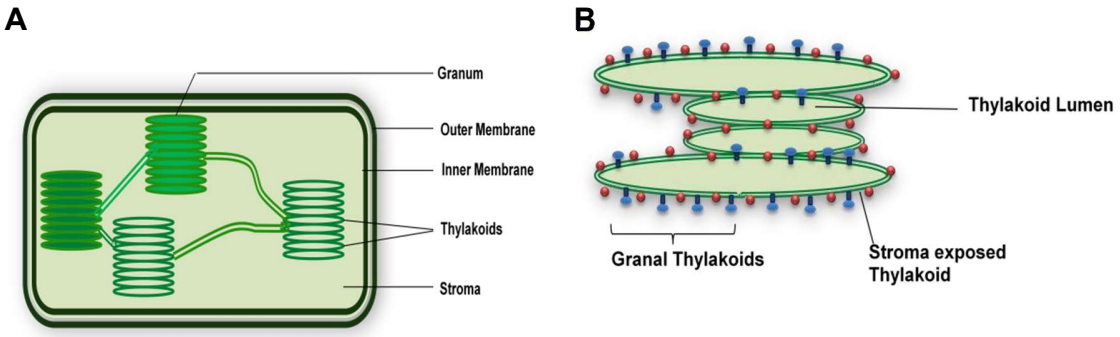

Figure 4.2: (A) Struture of Chloroplast, (B) Arrangement of

thylakoid membrane in chloroplast.

The inner membrane of the chloroplast further folds to a flattend membrane system known as **thylakoids**. The photosynthsis machinery such as light absorbing pigments, electron carriers and ATP synthesizing machinery is present on inner membrane as intergral protein complex. Thylakoid membranes are arranged like stack of coin to form **granum** (Figure 4.2, B). The granum throughout the chloroplast are connected by tubule to share the material. Over-all structure of chloroplast is similar to mitochondria but it has few significant structural and biochemical differences. Thylakoid membrane contains photosynthetic green colored pigment chlorophyll.

$$\mathbf{6CO_2 + 6H_2O + Solar\ Energy} \rightarrow C_6H_{12}O_6 + 6O_2 \quad \text{.....(4.1)}$$

Photosynthesis is an assimilation reaction involving CO_2 and water to produce sugar in the presence of solar energy (photons) that catalyzes fusion reaction as given Eq. 4.1. The photo system present on thylakoid membrane consists of two photo system, **photo system-I (PS-I)** and **photo system complex II (PS-II)**. PS-II absorbs the photon from solar energy to excite the electron to the higher energy state, and catalyze water break down into the proton and oxygen. The electron pass through multiple electron carrier and during this proton are exported out of the thylakoid membrane into the lumen. The proton passes through **ATP synthase** and returns back into the stroma to generate ATP. The electron from PS-II is eventually been received by PS-I and been excited after absorbing photon from sun light to high energy state. The energy associated with these electrons are used to generate NADPH in the stroma. Hence as a result of photosynthesis, solar energy is trapped by photo synthesis apparatus to generate **ATP** and **NADPH** into the lumen. Both of them are used to run **Calvin cycle** to assimilate environmental CO_2 to form sugar.

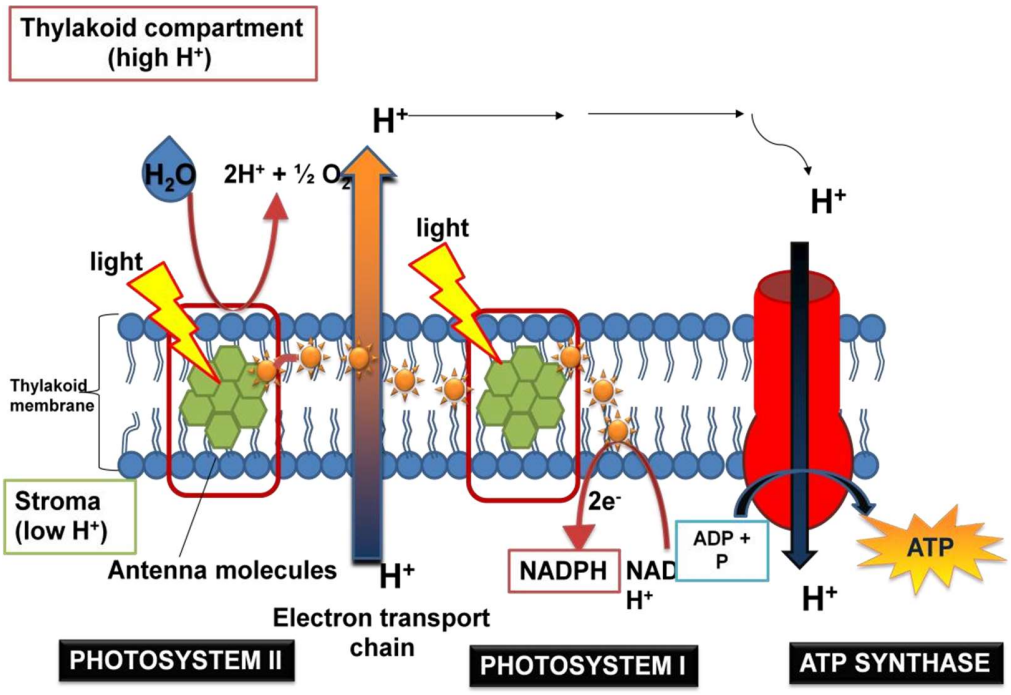

Figure 4.3: Different Steps of Photosynthesis.

5. Organelles of Vesicular Trafficking System: The main function of these organelles is to manage the distribution of material (food particles or proteins) throughout the cell. 3 different organelles such as endoplasmic reticulum, Golgi apparatus and lysosome, co-ordinately work together to maintains vesicular transport of material across the cell (Figure 4.3). Eukaryotic cell takes up the solid material from outside through a process called "**endocytosis**" whereas uptake of liquid is through a process called as "**pinocytosis**". Similarly material is secreted out of the cells through "**exocytosis**". In addition, intravesicular system delivers protein synthesized in endoplasmic reticulum to different organelles.

During endocytosis, material present outside the cells binds to the cells surface through cell surface receptors and trap it in a membraneous structure called as **endosome**. Endosomal vesicles are fused with the lysosomes to form late endosome. In late endosome, with the help of lysosomal enzymes material is digested and then endosome is fused with the Golgi bodies and deliver the content for further distribution. In the similar manner, during secretion, vesicles originate from Golgi bodies and fuse with the plasma membrane to release the content outside of the cell.

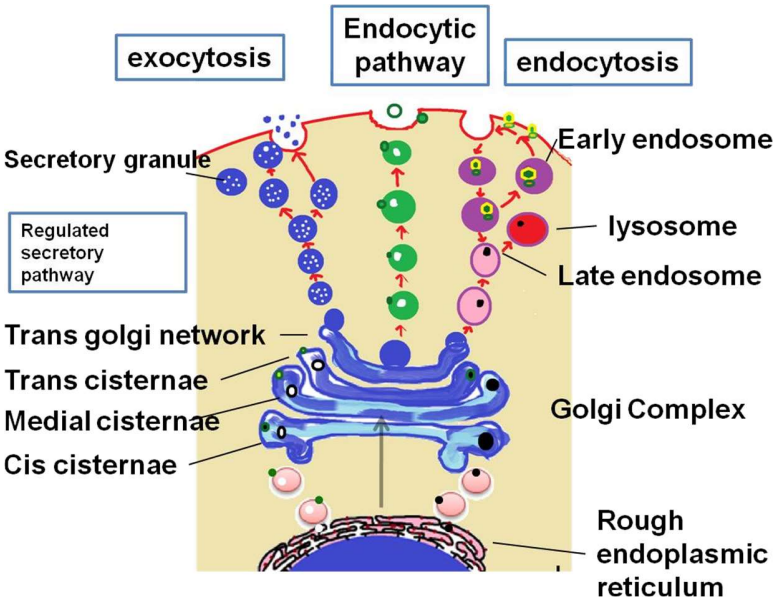

Figure 4.3: Intra cellular vesicular trafficking system of cell.

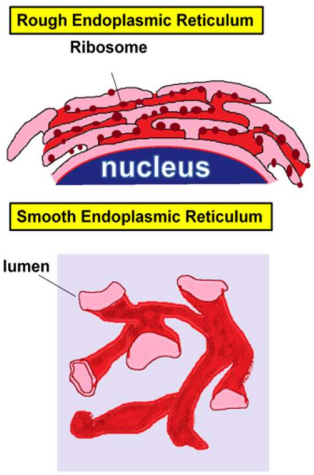

Figure 4.4: Endoplasmic reticulum.

Endoplasmic Reticulum- The vesicular network starts from nuclear membrane and spread throughout the cytosol constitutes endoplasmic reticulum (Figure 4.4). There are two different types of endoplasmic reticuli present in the cell, 1) Rough endoplasmic reticulum (RER), and 2) smooth endoplasmic reticulum (SER). RER has ribosome attached to it to give a rough appearance whereas smooth endoplasmic reticulum is devoid of ribosomes. Protein synthesis on ribosome attached to RER are sorted into 3 different catagories, such as integral membrane proteins, proteins for secretion and protein destined for different organelles. Proteins are synthesized with a n-signal peptide and these signal peptides are recognized by signal recognition particle on their the target organelles. For example, if a protein is synthesized with a signal peptide for mitochondria, it will attach to signal recognition particle and receptor onto the outer mitochondrial membrane to deliver the protein. The proteins without any signal peptide tags are supposed to remain in the cytsol.

Functions of endoplasmic reticulum:

1. Synthesis of steroid hormone in gonad cells.
2. Detoxification
3. Ca^{2+} sequestration
4. Synthesis of protein, phospholipid and carbohydrate.
5. Protein sorting to different organelles.
6. Protein modifications such as glycosylation etc.

Golgi Bodies- Golgi bodies were first visualized by a metallic stain invented by **Camillo golgi** and it is made of flattend, disk like cisternae arranged in a stacked manner to give 3 distinct zones (Figure 4.5). **Cis-face** receives material or vesicles from endoplasmic reticulum, **medial Golgi** is the actual place where protein are covalently modified with the sugar. **Trans Golgi** is the face of Golgi towards plasma membrane and this site sorts vesicle for their destined organelles or plasma membrane.

Functions of golgi bodies

1. Protein sorting

2. Protein modifications (Glycosylation)

3. Proteolysis

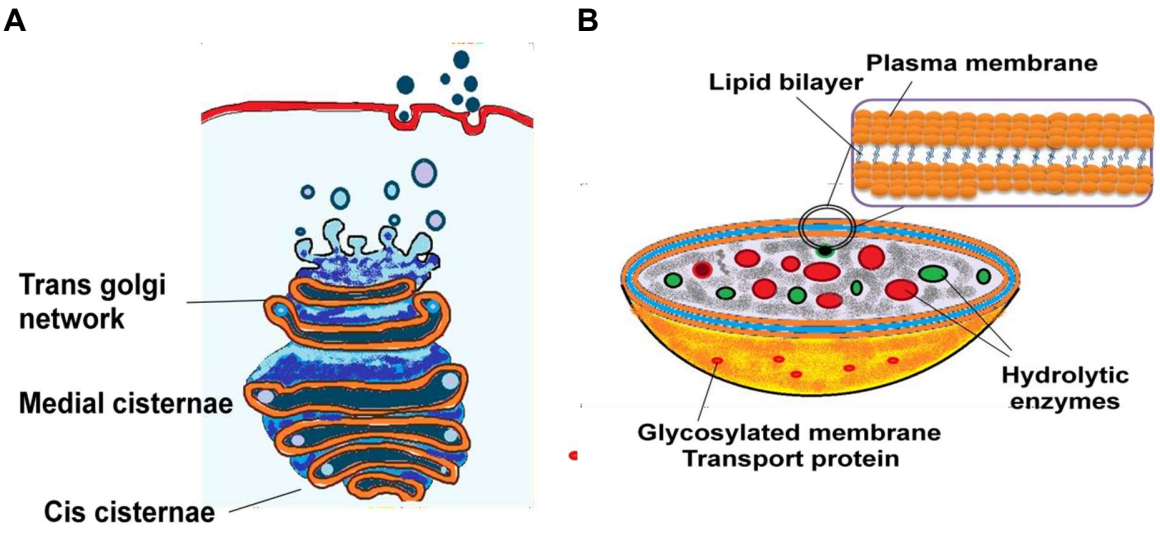

Figure 4.5: Scematic structure of (A) Golgi bodies and (B) Lysosome.

Lysosomes-Lysosomes are discovered by De Duve. They are membrane bound organelles and an important component of intracellular vesicular system (Figure 4.5). They are popularly known as suicidal bags due to their role in autophagy, a cellular process probably operates in cells during starvation to meet their energy requirements. [for more details of molecular mechanism of autophagy and underlying signaling mechanism could be find here: **Annu Rev Genet. 2009;43:67-93. Regulation mechanisms and signaling pathways of autophagy**]. Lysosome lumen is extremely acidic and contains protease, cytolytic enzymes to degrade the ingested material.

Functions of lysosomes

1. Degradation of ingested food material for delivery through vesicular system.

2. Degradation of pathogenic bacteria

3. Degradation of old protein.

Quiz

Q1: Which organelle is the destination for destruction of cellular proteins ?

Answer: Lysosome. The proteins that are either aged or misfolded are sent to lysosome for degradation. Lysosome with the help of acidic environment and proteases degrade protein into the smaller peptide. In addition, bacteria or other pathogenic organism also follows the same path to degrade into smaller pieces for antigen presentation in immune cells. Besides lysosome mediated protein degradation, proteins targeted for degradation are also sent to the proteasome complex. Proteasome complex is a non-membranous multimeric protein complex present in cytosol as free particles and they identify and degrade aged and misfolded proteins. **[Student can refer to following article for further details of proteasome: Structural biology of the proteasome. Annu Rev Biophys. 2013;42:29-49. doi: 10.1146].**

Q2: Which organelle can be visualzed by basic dye Hematoxylin ?

Answer: Nucleus. Nucleus contains genomic DNA (deoxy-ribonucleic acid) and then internal pH is acidic. As a result hematoxylin concentrates into the nucleus and visualize genomic DNA. It can also stain circular DNA in mitochondria but sensitivity of the dye is not optimal for visualization of mitochondrial DNA.

Q3: Treatment of mitochondria with molecule X destroys the proton gradient. These molecular are called as …….

Ans: Uncoupler. The electron transport chain consists of 4 complexes involved in relaying electron and complex V for harvesting proton gradient. Any molecule which can destroy the membrane permeability via making pores will eventually destroy the protein gradient and ultimately affects the ATP production.

Q4. Decribe the structural details of the molecular complex responsible for harvesting proton gradient in the mitochondria?

Ans: ATP Synthase. Please go through the structural details of ATP synthase from web and attempt to collect the information to describe the structure of ATP synthase.