WEEK 2

Unit 1_3: THE CYTOSKELETON	**Where? How long?**	***Explanations /Questions***
Introduction The cytoskeleton is a network of protein fibers that can be compared to the "skeleton of the cell" but also its "muscles" too. The cytoskeleton's function is to allow movement of organelles and vesicles within the cytoplasm, to keep the cell's shape and to allow cell's contraction. The cytoskeleton is a highly dynamic structure that is continuously reorganized as a cell changes shape, divides, and responds to its environment. There are three types of fibers within the cytoskeleton: microfilaments, intermediate filaments, and microtubules.	10 minutes	From cyto (cell) and skeleton, the cytoskeleton is a coherent whole of biological polymers constituting the skeleton of the cell. As medical students, it is essential to know how cells change shape, move, divide and respond to their environment.
Expectations During this section you will find out what is cytoskeleton, what it does, and what is made of. You will review the current concepts of stability and dynamics of the cytoskeleton.		
Course outline Microtubules Intermediate filaments Microfilaments		
Learning outcomes		
At the end of this unit, you should be able to: • Describe the cytoskeleton • Contrast the structure and function of microfilaments, intermediate filaments, and microtubules • Demonstrate the structure of the cilia and flagella • Illustrate the role of microtubules in intracellular transport and cell division		

ACTIVITY AHEAD OF LESSON: CYTOSKELETON.			
Purpose:	The cytoskeleton is a multifunctional network which is providing structural support to the cell, a basis for active transport mechanisms and a system for the production and transduction of mechanical force in the cell. This lecture will allow you to understand how the cytoskeleton keeps cellular organization by linking together many cellular components, mediating communication across the entire cell and consequently has a high effect on cellular functions.	30 minutes	
Over to you:	This video on cytoskeleton explains the different types of cytoskeletal elements and their function Watch the video. Then watch it again, making the notes suggested in the below activity. ***Be certain to note anything that you don't understand.***		
Activity:	List the types of protein fibers in the cytoskeleton. What will happen if a cell's cytoskeleton is damaged?		

ROLE AND COMPONENTS OF CYTOSKELETON

The cytoskeleton provides a supportive network of tubules and filaments in the cytoplasm of eukaryotic cells. The cytoskeleton is a dynamic structure responsible for cellular movement, changes in cell shape and contraction of muscle cells. It provides the machinery for intracellular movement of organelles.
It is composed of three types of supportive structures:

- **microtubules,**
- **intermediate filaments**
- **microfilaments**

Microtubules

Microtubules are polymers of tubulin that undergo rapid assembly and disassembly. They are found in the cytoplasmic matrix of all eukaryotic cells. Microtubules have been implicated in a large number of cell functions, including mitotic and meiotic chromosomal movement, transport of vesicles from one site to another within the cell, and ciliary and flagellar motility.

Structure

The major component of microtubules is **tubulin,** a protein dimer composed of two different polypeptides, α-tubulin and β-tubulin.
Microtubules are hollow cylinders with a diameter of approximately 24 nm.
A cross-sectional view shows that the wall of the cylinder is made up of **13 protofilaments,** each consisting of an alignment of **tubulin dimers.**
The individual protofilaments are arranged such that the α- and β-tubulin subunits alternate along the length of the protofilament.
The tubules have a diameter of approximately 24nm and the wall is 5-6 nm thick.

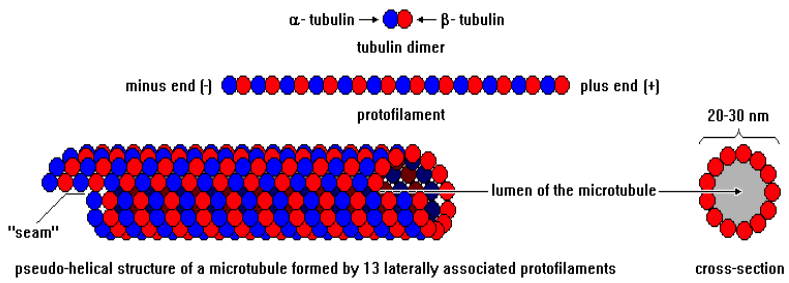


Figure 1.2. Illustration e of microtubules,by Jana Michejdy, September 2006, mikrotubula [image] accessed February 15,2019, https://commons.wikimedia.org/wiki/File:Mikrotubula007.PNG

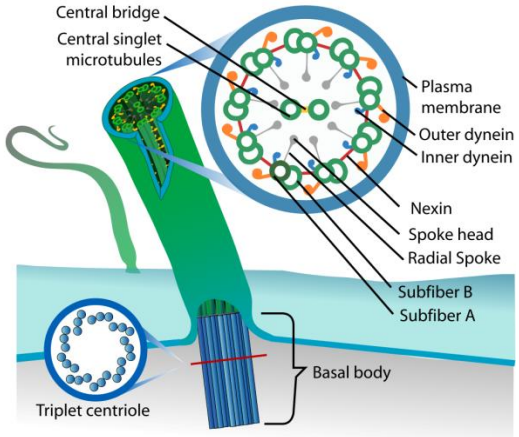
20 minutes

Notes to tutor

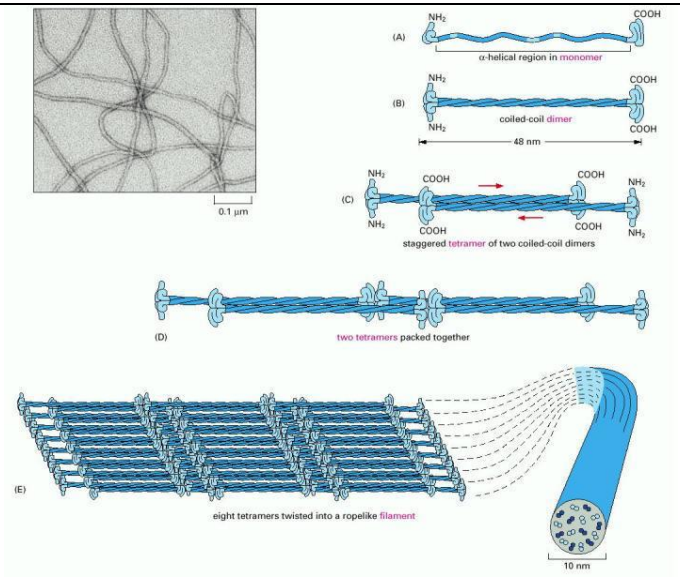
Explain to students what is tubulin

Project the video on assembly and disassembly of microtubules.

Explain the polarity of microtubules (+) and (-) end.

Cilia and Flagella In cilia and flagella, the microtubules are organized as a circle of nine double microtubules on the outside and two microtubules in the center in a structure called axoneme. **Centrosome** The centrosome is a region near the nucleus of animal cells that functions as a microtubule-organizing center. It contains a pair of centrioles, two structures that lie perpendicular to each other. Each centriole is a cylinder of nine triplets of microtubules. The centrosome replicates itself before a cell divides, and the centrioles play a role in pulling the duplicated chromosomes to opposite ends during cell division.	 **Figure 1.3. Eukaryotic cilium diagram**. Modification of work by Dartmouth Electron Microscope Facility, Dartmouth College; scale-bar data from Matt Russell. Lower panel, modification of "Eukaryotic cilium diagram," by Mariana Ruiz Villareal, November 25,2007 . [Image], accessed on February 14,2019 https://commons.wikimedia.org/wiki/File:Eukaryotic_cilium_diagram_en.svg.	10 minutes	Notes to tutor Explain to students what is a cilia and a flagella. Provide examples that flagella are motile structure of sperm while cilia are lining the cells of the respiratory tract.

IN CLASS ACTIVITY: MICROTUBULE DYNAMISM			
Purpose:	To demonstrate the importance of dynamism of microtubules	10 minutes for discussion in group 10 minutes for feedback	Students will use the previous group for this activity.
Over to you:	A microtubule can oscillate between growing and shortening phases. This complex dynamic behavior allows the cell to quickly assemble or disassemble microtubule structures. Read the following questions from Alberts Bruce (Alberts, 2010) Essential cell biology and answer them.		
Activity:	**Question 1:** The drug Taxol, extracted from the bark of yew trees, has an opposite effect to the drug colchicine, an alkaloid from autumn crocus. Taxol binds tightly to microtubules and stabilizes them; when added to cells, it causes much of the free tubulin to assemble into microtubules. In contrast, colchicine prevents microtubule formation. Taxol is just as pernicious to dividing cells as colchicine, and both are used as anticancer drugs. Based on your knowledge of microtubule dynamics, suggest why both drugs are toxic to dividing cells despite their opposite actions. **Question 2:** When cells enter mitosis, their existing array of cytoplasmic microtubules has to be rapidly broken down and replaced with the mitotic spindle that forms to pull the chromosomes into the daughter cells. The enzyme katanin, named after Japanese samurai swords, is activated during the onset of mitosis, and chops microtubules into short pieces. What do you suppose is the fate of the microtubule fragments created by katanin? Explain your answer.		

Intermediate filaments These components of the cytoskeleton, with a diameter of 10nm, are intermediate in thickness between microtubules and microfilaments. These rope-like filaments function primarily in structural roles. Their function is to maintain the cell shape and anchoring organelles. They constitute a population of heterogeneous filaments that includes keratin, vimentin and neurofilaments. **Figure 1.3. Assembly of filaments**, by Alberts B, Molecular Biology of the Cell. 4th ed. New York: Garland Publishing; c2002.Figure 16-16.Accessed on February 15,2019. https://commons.wikimedia.org/wiki/File:Assembly_of_filament.JPG.		30 minutes	Note to tutors Emphasize that intermediate filaments have no role in cell movement. Their function is purely structural. Keratin is the main material that strengthens hair and nails and the epidermis of the skin. Vimentin : provides structural stability for many kinds of cells Neurofilaments are the intermediate filaments of nerve cells.

Microfilaments

They are the thinnest class of the cytoskeletal fibers, are solid rods of the globular protein actin and function in moving cellular components, for example, during cell division.

They also maintain the structure of microvilli, the extensive folding of the plasma membrane found in cells dedicated to absorption. These components are also common in muscle cells and are responsible for muscle cell contraction. They are involved in moving cellular components, for example, during cell division.

They also support microvilli as well and maintain their shape. Actin filaments are also present in muscle cells and are responsible for muscle cell contraction.

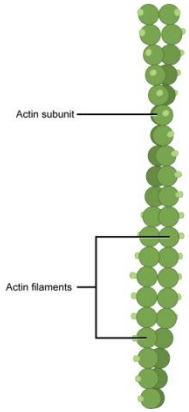


Figure 1.4. Microfilaments are made of two intertwined strands of actin. By OpenStax Biology 2nd Edition, Feb 13, 2019, accessed on February 20th, 2019. https://cnx.org/contents/jVCgr5SL@15.3:MrwSH721@10/4-5-The-Cytoskeleton

LABORATORY PRACTICALS : CELLULAR FRACTIONATION			
Purpose:	To separate cellular organelles by a technique called **differential centrifugation** The process of breaking open cells in an isotonic buffer is **homogenization** and the subsequent isolation of organelles is **cellular fractionation**.	120 minutes	Before this laboratory exercise takes place, make sure materials specified in the Standard Operating Procedures (SOPs) are available and the laboratory prepared. In groups of 2 per table, students will use one light microscope. Ask students: To separate cellular organelles by differential centrifugation At the end of manipulation, each group must write down a short report.
Over to you:	In this experiment, cellular fractionation of a homogenized goat or beef liver will be accomplished via a technique called **differential centrifugation.**		
Activity:	This experiment will be done according the lab manual.		

Anticipated time required for Unit 1.3 activities :

Theory : 2h all activities + 2h self-learning

Practical : 2h

Reference

Alberts, B. (2010). *Essential cell biology* (3rd Ed). New York; London: Garland Science.