Glossary - Cellbiology

Blotting: (Blot Analysis) Widely used biochemical technique for detecting the presence of specific macromolecules (proteins, mRNAs, or DNA sequences) in a mixture. A sample first is separated on an agarose or polyacrylamide gel usually under denaturing conditions; the separated components are transferred (blotting) to a nitrocellulose sheet, which is exposed to a radiolabeled molecule that specifically binds to the macromolecule of interest, and then subjected to autoradiography.

Northern B.: mRNAs are detected with a complementary DNA;

Southern B.: DNA restriction fragments are detected with complementary nucleotide sequences; **Western** B.: Proteins are detected by specific antibodies.

Cell: The fundamental unit of living organisms. Cells are bounded by a lipid-containing <u>plasma membrane</u>, containing the central <u>nucleus</u>, and the <u>cytoplasm</u>. Cells are generally capable of independent reproduction. More complex cells like Eukaryotes have various compartments (organelles) where special tasks essential for the survival of the cell take place.

Cytoplasm: Viscous contents of a cell that are contained within the plasma membrane but, in eukaryotic cells, outside the nucleus. The part of the cytoplasm <u>not</u> contained in any organelle is called the Cytosol. **Cytoskeleton:** (Gk.) Three dimensional network of fibrous elements, allowing precisely regulated movements of cell parts, transport organelles, and help to maintain a cell's shape.

- Actin filament: (Microfilaments) Ubiquitous *eukaryotic* cytoskeletal proteins (one end is attached to the cell-cortex) of two "twisted" actin monomers; are important in the structural support and movement of cells. Each actin filament (F-actin) consists of two strands of globular subunits (G-Actin) wrapped around each other to form a polarized unit (high ionic cytoplasm lead to the formation of AF, whereas low ion-concentration disassembles AF). Actin filaments within all cell types bind to a variety of accessory proteins, including those that prevent or enhance lengthening, those that break filaments, and those that bind filaments together and to other structures, particularly membranes (membrane associated proteins, MAPs). In the smooth muscle cells, actin filaments are part of contractile machinery that includes other proteins such as myosin, tropomyosin, and calmodulin (a calcium binding enzyme regulator) and also play an essential role in cytokinesis of animal cells (division of cytoplasm during telophase in mitosis, causing separation of the cell into two daughter cells) and accounts for the amoebic motion of protists.
- **Intermediate filament**: Cytoskeletal fibers (10nm in diameter) formed by polymerization into a node-chain-node polymer of several classes of cell-specific-subunit proteins, mostly <u>keratin</u> (fibrous, insoluble, and relatively stable). They help to maintain cell shape, provide the reinforcement of epithelial cells by attaching to <u>spot</u>- and <u>hemi</u>-desmosomes; from the major structural proteins of skin, nails, feathers and hair; form the scaffold that holds the Z disks and myofibrils in place in muscle; and generally function as important structural determinants in many animal cells.
- Tubular filament: (Microtubules) A family of rapidly dis/assembling *eukaryotic* cytoskeletal proteins consisting of three highly conserved GTP-binding proteins. Dimers of α- and β-tubulin monomers, line up in longitudinal rows (protofilaments) polymerize into a "spirally" coiled microtubular chain which are necessary for movements of flagella and intercellular vesicles. TF have a slow growing end designated as (-), and a fast growing end (+) which is usually farthest away from the cell center. TF move colored pigment granules around the skin cells of reptiles and fish. A third class of tubular monomers, γ tubulin organizes the spindle apparatus that separate the chromosomes during cell division (mitosis) colchicine, prevents the buildup of TF (see also cell organelles, centrioles).
- **Microtubule-Associated Protein** (MAPs): A protein that binds to microtubules in a constant ratio and determines the unique properties of different types of microtubules. Numerous MAPs have been identified including the motor proteins **Dynein** (<u>retrograde</u> direction towards the cell center) and **Kinesin** (<u>anterograde</u> direction away from the cell center).

Cytosol: The part of the cytoplasm not contained in any organelle is called the Cytosol (up to 70% of the fluid portion). It does contain however the cytoskeleton, lipid droplets, and glycogen granula (storage site). The Cytosol is also the <u>site of glycolysis</u> and <u>gluconeogenesis</u>, excretion cycle, and fatty acid cycle. The Cytosol also possesses two distinct phases - <u>gel-phase</u> (high viscosity at the cell membrane) and sol-phase (more liquid within).

Cell Cycle: Set of events that occur during the division of mitotic cells - periodically cycling between mitosis (M phase) and Interphase. **Interphase** can be subdivided in order into: G1, S and G2 phase. DNA synthesis occurs during S-Phase.

 G_1 -phase: Gap-1, preceding S-phase (haploid); Under certain conditions, cells exit the cell cycle during G_1 and remain G_0 state as non-growing, non-dividing cells. Appropriate stimulation of such cells induces them to return to G_1 and resume growth and division.

S-phase: the phase in which DNA synthesis occurs (doubling of DNA) of decondensed DNA strands (euchromatin).

G₂-phase: after DNA synthesis preceding M-phase (diploid).

M-phase: the mitotic phase where cell division takes place. Within the nucleus, densely packed, condensed chromosomes (**Heterochromatin** and associated proteins, like histone cores) become visible. Mitosis produces two daughter nuclei identical to the parent nucleus; (di-, polyploid) (see chromosome packing)

Prophase: (Gk. *Pro*, early; *phasis*, form) Early stage of <u>n</u>uclear <u>d</u>ivision; nucleus disappears, mitotic spindle forms, chromosome condense and become visible.

Metaphase: (L. *meta*, half) Intermediate stage o.n.d.; chromosomes align along the equatorial plane. **Anaphase**: (Gk. *ana*, away) Spindle separates centromere, pulling chromatids apart to the opposite poles of the cell.

Telophase: (Gk,. *Telo*, late) Late stage; spindle dissolves, nuclear envelope reappears daughter nuclei re-form (segregation and cytokinesis).

MPF (mitosis promoting factor,) A heterodynamic protein that triggers entrance of a cell into the M-phase by inducing chromatin condensation and nuclear envelope breakdown; originally Maturing PF, since it supports maturation of G2-arrested frog oocytes into mature eggs. It consists of two subunits (an A or B type cyclin and a cyclin dependent protein kinase) which together express the MPF-characteristics.

Cell Membrane: (Cell Wall) A specialized, rigid extracellular matrix that lies next to the plasma membrane, protecting a cell and maintaining its shape. It is prominent in most fungi, plants (both cellulosic) and prokaryotes, but is not present in most metazoan (multicellular animals). (See also membranes) CM in prokaryotes: Bacteria have a rigid layer of cell walls, thin sheets composed of N-acetylglucosamine, N-acetylmuramic acid, and a few amino acids. Also called Murein. Staining reveals two classes of bacteria:

- Gram-Negative: A prokaryotic cell whose cell wall contains relatively <u>little peptoglycan</u> but contains
 an outer membrane composed of <u>lipopolysaccharide</u> (<u>LPS</u>), lipoprotein, and other complex
 macromolecules.
- **Gram-Positive**: A prokaryotic cell whose cell wall consists <u>chiefly</u> of <u>peptidoglycan</u> and lacks the outer membrane of Gram-negative cells.

CM. Junctions: Specialization regions of the cell surface through which cells are joined to each other:

• **Desmosomes**: (providing mechanical stability, 30nm apart) consisting of dense protein plaques connected to intermediate filaments that mediate adhesion between adjacent cells and between cells and the extracellular matrix.

Adherens junctions: Primarily in <u>epithelial</u> cells, form a belt of <u>cell-cell</u> adhesion just under the tight junctions.

Hemidesmosome: Similar in structure to spot Desmosomes, anchor the plasma membrane to regions of the extracellular matrix (a usually insoluble network consisting of glycos-amino-glycans, collagen, and various adhesive proteins like laminin or fibronectin, which are secreted by the animal cells. It provides structural support in tissues and affects the development and biochemical functions of cells) **Spot Desmosomes**: In all epithelial cells and many other tissues, such as smooth muscles. They are button-like points of contact between cells, often thought as spot-welds between adjacent plasma membranes.

- **Gap Junctions**: (communicative) <u>Protein-lined channels</u> (3 nm thick) between adjacent cells that allow passage of <u>ions</u> (electrical connection in nerves mediated by the *flip-flop* characteristics of ion concentration) and small molecules between the cells.
- **Plasmodesma**: (communicative) Large bridges of cytoplasm that connect plant cells and allow rapid exchange of materials between them.
- **Tight Junctions**: (communicative) <u>Ribbon</u>-like bands connecting adjacent epithelial cells that prevent leakage of fluid across the cell layer.

Cell Organelles: In eukaryotic cells, a complex cytoplasmic structure with a characteristic shape that performs one or more specialized functions.

Centrioles: Rod-shaped organelles that organize certain cytoskeletal fibers and are instrumental in the movement of cellular parts during cell division (mitosis and meiosis).

Mitochondrion (pl. Mitochondria): A microbody that provides cells with energy in form of ATP-molecules by breaking down certain C-containing molecules (glucose) into water and CO₂ (needs O₂). They are defined by two limiting membranes. The <u>inner membrane</u> forms folds or invaginations called **cristae** (increases surface area to extend the energetic output and to prevent the electrons of the electron transport chain to reconvert into the energetically lower state), which project into the interior of the organelle. Cristae may be shelflike or tubular, and in the steroid-secreting cell. Sizes and shapes of M vary considerably within one cell, mitochondria move, change shape, divide and fuse; Mitochondria do have their own genome. This DNA encodes the cytochrome (e-transport chain), rRNA, tRNA.

- The following reactions take place within a mitochondrion:
- Aerobic Metabolism: Foodstuff molecules are oxidized completely to CO₂, and H₂O by molecular O₂.
- Anaerobic M.: Foodstuff, molecules are oxidized incompletely to lactic acid.
- Oxidative phosphorylation: The substrates needed are Pyruvate, fatty acids, ADP, and P_i. They are transported to the matrix from the Cytosol by transports; O₂ diffuses into the matrix. A shuttle system provides free electrons from cytosolic NADH to generate mitochondrial NADH. Fatty acids, and Pyruvate are needed to keep the KREBS-cycle running which provides the mediators for the electron transport chain. ATP is transported to the Cytosol in exchange for ADP and P_i, CO₂ diffuses out from the mitochondrial matrix into the Cytosol across the mitochondrial membranes:

$$NADH + H^{+} + 3ADP + 3P_{i} + \frac{1}{2}O_{2} \rightarrow NAD^{+} + 4H_{2}O + 3ATP$$

• ATP-synthase: (the F₀F₁ ATPase complex) The F₀ portion is an integral membrane protein; the F₁ portion contains three copies of α-, and three β-subunits and is bound to F₀ via subunits γ, δ, and ε. The synthesis of ATP from ADP and P_i occurs spontaneously at the catalytic site on a β-subunit of the F₁, due to tight binding of ATP to this site. Proton movement through F₀, driven by the proton-motive force, promotes the catalytic synthesis of ATP by causing the bound ATP to be released; this frees up the site for the binding of ADP and P_i, which, in turn, spontaneously combine to form another tightly bound ATP; the entire process is osmotically coupled.

Nucleus: The membrane-bound region in Eukaryotes that contains the cell's DNA. To accomplish the tasks of <u>transcription</u> (the formation of RNA from a single strand of DNA molecule, where the flow of information is diagrammed as DNA \rightarrow RNA \rightarrow Protein) and <u>replication</u> (cell division), certain preconditions have to be met:

- **Spriting**, the <u>decondensation</u> of chromatin threads after a completed cell division to make RNA polymerase possible (euchromatin); redundant, double or triple encoded genes can remain condensed (Heterochromatin), hence remain inactive; Heterochromatin can be either *facultative* (a cell can express just those gene necessary for its tasks, i.e. nerve, liver cell etc. or *constructive* as seen in <u>Barrbodies</u> depending upon the homo/heterozygotic state of the all over organism.

 In prokaryotes, <u>plasmids</u> play a role in determining the stage of cellular reproduction S1, S2, I-phase.
- **Nucleus plasma relation**, the larger the plasmatic phase the more likely mitosis is going to take place see also cell surface to volume ratio.
- Nuclear Envelope: Double-membrane structure surrounding the nucleus separating the nucleoplasm from the cytoplasm. The outer membrane is <u>continuos with the endoplasmic reticulum</u> and the two membranes are perforated by <u>nuclear pores</u> (granulated pores). The inner layer made of a fibrous network is composed of coiled <u>lamin</u> filaments and holds the envelop together similar to tubulin structure (during mitosis, at late prophase, both inner and outer layer of the envelope are dissolved to be rebuilt after telophase). Another key function of the NE protects the enclosed DNA from being broken apart by movements of the cytoplasm.
- Nuclear Pore: Double-iris diaphragm gates, which serve as pathways for traffic between the nucleus and the cytoplasm. This gate allows passive diffusion even though when closed (does not shut completely), but requires energy to open entirely, mediating the transport of larger molecules.
- **Nucleolus**: Dark-staining region within the nucleus of eukaryotic cells where ribosomal RNA is synthesized on many copies of DNA. Fibrillar and granular regions within the nucleolus bring about the various types of rRNA.
- **Nucleoplasm**: Provides the energy needed for those processes via glycolysis and is the site of <u>ribosome synthesis</u>. The <u>nuclear organizer</u> is the region of DNA on one or more chromosomes that direct the synthesis of ribosomal RNA. nucleoplasm consists in two distinct phases: <u>nucloesole</u> (enzymes, proteins, with limited glycolytic reactions) and the <u>nucleogel</u> (???).

Peroxisome: Small <u>microbody</u> in eukaryotic cells whose function include <u>degradation</u> of fatty acids and amino acids that forms as a by-product of a cell's metabolic functions, hence are most abundant in liver cells. POx splits hydrogen peroxide, which is converted to water and oxygen by oxidases (catalase):

$$H_2O_2 \rightarrow (catalase) \rightarrow H_2O + \frac{1}{2}O_2$$

Plastid: (Gk. plastid, formed molded) Plant-organelle in the cells of certain groups of Eukaryotes that is the site of such activities as food manufacture and storage; plastids are bounded by two membranes. Chloroplasts do have their own DNA which encodes for their chlorophyll.

- **Chloroplast**: : (Gk. chloros, green) A plastid in which <u>chlorophylls</u> are contained; the site of photosynthesis; it contains grana (stacks of thylakoids), starch grains and tiny lipid droplets.
- Cyclic Photophosphorylation: A reaction in which phosphate is added to a compound; e.g.: the formation of ATP from ADP, NADPH from NADP⁺ and inorganic phosphate (instead of NADH in

animal cells). PSII:
$$ADP + P_i \rightarrow (E_{light} = h \cdot f) \rightarrow ATP$$

PSI:
$$2NADP^+ + 2H^+ \rightarrow (E_{light} = h \cdot f) \rightarrow 2NADPH$$

Ribosome: A complex comprising several different rRNA molecules and more than 50 proteins, organized into a large subunit and small subunits, involved in protein synthesis; R may lie freely in the cell or are attached to the membranes of the endoplasmic reticulum. **Polysomes**, heaps of transcribing ribosomes working along one mRNA strand for protein synthesis. Prokaryotic ribosomes are slightly lighter than eukaryotic ones and cannot take over task of cells of the other kingdom (except for a few special cases).

Vacuole: (L vaccus, empty) A space or a cavity within the cytoplasm filled with a watery fluid, the cell sap; part of the lysosomal compartment of the cell.

Cell Surface to Volume Ratio: The ratio of the surface area of a cell to its volume; imposes limits on cell size because as the cell's linear dimensions grow, its surface area increases less than its volume, thus hindering diffusion of essential materials into and metabolic waste products out of the cell.

Cell Theory: The biological doctrine stating that <u>all living things are composed of cells</u>; cells are the <u>basic units within organisms</u>, and the <u>chemical reactions of life occur within cells</u>; all cells <u>arise from dividing preexisting cells</u>.

Compartments within an eukaryotic cell may have arisen by the following mechanisms:

Endosymbiont Theory: Both chloroplasts and mitochondria are thought to be prokaryotic derivatives which made their way into the host cells with time. Since plants do have a cellulose cell wall, hence cannot undergo phagocytosis, (it is still an argument awaiting final solution).

Phase Theory: (after Schnepf) (1) Similar structures are more likely to combine than dislike structures; e.g.: fungal hyphae (plasmogamy, followed by karyogamy); (2) Similar structures always are separated by a double membrane (bilayer).

Cellular Strategists:

K-Strategists: Eukaryotes may be unicellular, colonial, or multicellular, hence complexity dominates within the organism, which is subdivided into organs each responsible for specific tasks.

- Animal C.: Cells of metazoans packed together to form tissues, organs and entire organism.
 Germ C.: Sperm cell or ovum containing the haploid gametes.
 Somatic C.: A cell that is not destined to become a gamete; a *body cell*, whose genes will not be passed on to future generations.
- **Plant** C.: Eukaryotes of the plant kingdom consist of a cell wall (cellulose) and a protoplast. **R-Strategists**: Single-celled organisms focusing mainly in reproduction, as seen in bacteria which can reproduce within minutes.
- Bacterial C.: All prokaryotes unicellular, single celled organism, excluding Archaea.

Chromosome: (G. chroma, color; soma, body) A linear end to end arrangement of genes and other DNA, sometimes with associated protein and RNA, found in Eukaryota.

Euchromatin: Decondensed DNA strands, allowing RNA polymerase.

Heterochromatin: Densely staining condensed chromosomal regions, believed to be for the most part genetically inert (found in salivary glands of *Drosophila sp.* near the chromocenter).

Lampenbürsten C.: Extremely <u>decondensed</u> DNA (euchromatin) where transcription of mRNA takes place; resulting electrostatic charges repel RNA strands - giving it a lamp-shade appearance.

Polytene C.: A giant chromosome produced by an endomitotic process in which the multiple DNA sets remain bound in a haploid number of chromosomes; also known as DNA-amplification.

Puff: A localized synthesis of RNA occurring at specific sites on giant chromosomes of *Drosophila sp*. Chromosomal **Mutation**: (L. mutare, to change) A <u>permanent</u> change in chemical structure, organization, or amount of DNA; produces a gene or a chromosome set differing from the wild type, resulting in a faulty protein (loss or gain of function); although most mutations are lethal, a few of them do indeed have a potential survivability and are consequently favored or suppressed in evolution due to selective mechanisms (evolution in real time).

M. at **Genome level**: Altering the chromosomal number (detectable with microscopic analysis).

Dis / Appearance: Paired chromosomes fail to segregate properly in meiosis (early anaphase); number of chromosomal sets altered - monosomy (2n-1); disomy (n+1); trisomy-21/18/13-, XXX, XXY. (2n+1); tetraploidity. Aneuploidy can lead to malignant tissue growth as well.

Chromosome **Packing**: In Eukaryonts;

Histone: A type of basic protein that forms the unit around which DNA is coiled in the nucleosomes of eukaryotic chromosomes, allowing extreme long DNA molecules to be packed into a cell nucleus. **h1** (stabilizing solenoid, in between every nucleosome) **h2**, **h2a**, **h2b**, **h3**, **h4** (form the octameric core).

Nucleosome: The <u>basic unit</u> of eukaryotic chromosome structure; a ball of eight histone molecules wrapped around by two coils of DNA; it is the main protagonist in packing the DNA strand; can easily be disturbed by UV-exposure (easily absorbs wavelengths of about 260 [nm])

Scaffold: The central framework of a chromosome to which the DNA solenoid is attached as loops; composed largely of topoisomerase.

Solenoid Structure: The packed arrangement of DNA in eukaryotic nuclear chromosomes produced by coiling the continuos string of nucleosomes.

Supercoil: A closed double stranded DNA molecule that is twisted on itself in prokaryotes.

Chromosome Structures: Regarding to their all over structure chromosomes are classified as:

- Acrocentric C.: A chromosome having the centromere located slightly nearer one end than the other.
- **Metacentric** C.: A chromosome having its centromere in the middle.
- **Pointiform** C.: A chromosome in which one allele is present only as a point-like appandage.
- **Submetacentric** C.: A chromosome in which one allele is shorter than the other.

Centromer: A kinetochore; the constricted region (CEN) of a nuclear chromosome, to which the spindle fibers (microtubuli) attach during division.

DNA-Amplification: In a polytene chromosome (e.g.: salivary glands of *Drosophila sp*, is replicated in parallel many times; or ciliates replicate genome up to 100 fold, which allows them to shuffle the genetic material to have equally amitotically or nonmitotically divided micronuclei).

Okazaki Fragments: Each of the short discontinued segments in the 3'-5'-direction of the <u>lagging</u> strand made by DNA polymerase-III - about 1500 bases in eu-, 150 bases in prokaryota.

Primer: A short RNA nucleic chain (polynucleotide) required to recognize the origin in DNA replication for DNA polymerase, where the 1st nucleotide is attached.

Telomer: The tip or end of a chromosome. <u>Telomerase</u> makes sure that the <u>lagging</u> strand will <u>always</u> be completed correctly without shortening replicated DNA, even though located at terminal ends (shortening of telomere could be the key to aging).

Cilium: (L. eyelash) A short, centrioles-based, hairline organelle. Rows of cilia propel certain protists. Cilia also aid the movement of substances across ephitelial surfaces of substances across ephitelial surfaces of animal cells - see flagellum.

Clone: (Gk. klon, twig) A population of cells or individuals derived by <u>asexual</u> division from a single cell or individual; one of the members of such a population, which are genetically identical to its parent. From a genotype point of view every cell within an organism is a clone of the originally fertilized zygote, whereas from a phenotypic point of few it is not, since different genes are activated resulting in a vast variety of cell types.

Cytomembrane System: <u>Intracellular membrane</u> organelles such as endoplasmic reticulum, Golgi apparatus, Lysosomes and vesicles.

Endoplasmatic Reticulum: Network of interconnected membranous structures within the cytoplasm of eukaryotic cells. Resulting products of the ER are transported by vesicles to the Golgi complex.

- Rough ER: It is associated with the ribosomes and functions in the synthesis and processing of membrane- and secretory proteins (food digesting enzymes) for various organelles such as lysosomes.
- **Smooth** ER: Consists of a ribosome-free network of fine tubules; detoxify lipid-soluble substances as well as synthesize lipids (cholesterol, steroids, modify sex-hormones etc.).

Golgi Complex: Proteins synthesized on the rough ER are sequestered in transport vesicles and travel to the GC, where they are structurally modified and sorted according to their destination. In addition to directing proteins to secretory vesicles, lysosomes, or particular membranes, the Golgi in polarized epithelial cells carry out a more refined sorting to apical versus basolateral plasma membrane domains. Each Golgi apparatus is made up of **cisternae** and related vesicles and is easily recognized on electron micrographs by its unique crescent form with a <u>convex</u> (**cis**, **entry**) and a <u>concave</u> (**trans**, **exit**) surface. The Golgi functions in a highly ordered sequential manner with separate biochemical events occurring initially in the *cis*-, then in the *medial*-, and finally in the *trans*- cisternae.

Lysosomes: Spherical vesicles that contain powerful digestive enzymes. In their simplest form as primary lysosomes, they are homogeneous, dense, membrane-bound organelles packed with acid hydrolase capable of breaking down polymers of all types, halt infections, and sometimes trigger autolysis (as a suicide bag) when the cell ages or damages. The low pH required for the hydrolase activities (<pH 5) is maintained by a membrane ATP-dependent hydrogen ion pump. A major function of lysosomes is to processes and recycle worn-out material entering from the extracellular environment before it is released into the cytoplasm.

Vesicle: A small membranous container in a cell.

- **Endocytic** V.: Bud off the plasma membrane and carry the engulfed liquid or solid materials to lysosomes, where they are digested and recycled.
- Exocytic V.: Transports material to the cell surface; lipids for inclusion in plasma membrane, and proteins for secretion.
- Transport V.: Shuttles lipids, proteins, and other materials to and from various organelles.

Cytosis: (Gk. kytos, bladder) Feeding at cellular level.

Endo-C.: (Gk. *endon* within) The take-up of material from outside by invaginations of the cell membrane to form vesicles. Receptor mediated EC, is brought about by specific **ligands** needed to activate it; e.g.: cholesterol uptake from the blood is mediated by LDL-receptors.

- Pino-C.: (Gk. pinos, to drink) The uptake by cells of droplets of solution by endocytosis.
- **Phago-**C.: (Gk. *phagein*, to eat) The engulfing by endocytosis of microorganisms, other cells, and foreign particles by a cell such as a white blood cell or an amoebae.

Exo-C.: (Gk. *ex*, out) Molecules synthesized within the cell are released in the external environment. **Euploidity**: A cell having any number of complete chromosome sets, an individual composed of such cells; euploidity includes diploid, tetraploid, polyploid organisms.

Flagellum: (L. *flagellare*, to whip) Long whiplike organelle protruding from the surface of the cell that either propels the cell (locomotion), or moves fluids past the cell serving as a feeding apparatus.

Eukaryotic flagellum: Movement of the thick microtubular protein chain (9+2 structure) is brought about by coordinated sliding of the several microtubules present in each flagellum.

The core of each flagellum contains the **axoneme**, a central pair of microtubules, surrounded by 9 peripheral pairs of microtubuli. Each doublet consists of one complete tubule (subfiber A) attached to an incomplete tubule (subfiber B). **Dynein** side-arms project from the A-tubule of each doublet. Whiplike movements are initiated by chemicals (neurotransmitters, hormones). Ciliar or flagellar movement involves the controlled attachment and detachment of the **Dynein** side arms to the facing B tubule such as that each doublet "walks" along an adjacent doublet. This process requires ATP (Dynein hydrolyzes ATP to ADP + P)

Prokaryotic flagellum: (Bacterial flagellum) A thin (< 20nm), filamentous organ of motility that functions by rotation. It protrudes out across the cell-membrane, sustained by special protein-bearings. The constantly rotating flagellar motor is driven by the permanent conversion of ATP to ADP + P. In the absence of chemical attractant the cell swims randomly in runs, changing direction during tumbles. In the presence of an attractant runs become biased, and the cell moves up the gradient of the attractant (ligand-mediated transducer proteins along the lateral side of the bacterium allow gradient detection; a self-limited saturation limit prevents over-stimulation of directed swimming).

Fractionation: Organelle sorting of cellular substances physically (mechanically, ultrasonic, etc.) or chemically separated into their individual components by homogenizers yielding a cellular suspension.

Cell Fractionation: The different sedimentation rates of various cellular components make it possible to separate them partially by centrifugation. Each soluble fraction can be further separated by density-gradient centrifugation, by gradually increasing rotation to as much as 300 E³ g.

Fluorescence Activated Cell Sorter (FACS): A concentrated suspension of cells is allowed to react with a fluorescent antibody or dye that binds to a particle of molecule such as DNA. The suspension is mixed with a buffer, the cells are passed single file through a laser light beam, and the fluorescent light emitted by each cell is measured. The light scattered by each cell can then be measured simultaneously; from this the size and shape of the cell can be determined. The suspension is then forced through a nozzle, which forms tiny droplets containing at most a single cell. At the time of formation, each droplet is given an electric charge proportional to the amount of fluorescence. Droplets with no or different charges are separated by an electric field and collected.

Gene Dose: The number of copies of a particular gene present in the genome, the entire complement of genetic material in a chromosome set; it is usually precisely determined, since an upset balance can disrupt certain enzymatic pathways - tetraploidity and mongolism; gene dose in homozygote organism is controlled by muting redundant copies.

Micelle: A microscopic particle made from an aggregation of amphipathic molecules in solution.

Microscopy: Device to magnify microscopic samples, usually not visible for the naked eye. The specimen is usually mounted on a transparent glass slide and positioned on the movable specimen stage of the microscope. Light from a bright source is focused by the collector and condenser lenses onto the specimen. The objective lens picks up the light transmitted by the specimen and focuses it on the focal plane of the objective lens, creating a magnified image of the specimen.

Resolution to light microscopy is limited to about 0.2 [μm], whereas in electron microscopy to 5 [pm] Confocal Laser M.: Usually an inverse microscope which operated by monochromatic light source. This laser mediated scanning technique uses fluorescenting substances to stimulate a particular layer of the specimen to release secondary light emissions. With the help of integrated computing systems a three dimensional image can be created. CFM increases resolution and magnification of up to 7000 fold.

Electron M.: A beam of electrons emanating from a heated tungsten filament is focused onto the specimen plane by the magnetic condenser lens. The entire column, from the electron generator to the screen, is maintained at very high vacuum.

To make specimen useable for EM, the sample has to undergo some fundamental processes. A filament of heavy metal, such as platinum, is heated under vacuum so that the metal evaporates and some of it falls over the sample grid in a very thin film. The biological material is then dissolved by acid, so that the observer views only the metal replica of the sample (artifact).

- Scanning EM: The electron beam scans the sample in periodic cycles over and over again. Electrons scattered from the specimen (entire organic structures can be used without slicing) are collected and amplified electronically to be displayed synchronously with the scanning beam on a visualizer yielding a 3D-image.
- **Transmission** EM: The electrons passing through the ultra-thin specimen are focused by a series of objective and projector lenses to form a magnified image of the specimen on a fluorescent viewing screen or a piece of photographic film.

The use of antibodies to detect sub-cellular location of a specific protein: A slice of tissue is fixed with glutaraldehyde and sectioned. An IgG specific protein antibiody is then added to the section. When complexed with gold-particles (protein covered Au-vesicle, 4nm in diameter), the stem of the IgG binds tightly to the protein-coating of the particle. Each gold-particle makes the resulting immune complex visible in the electron microscope when this immune complex attaches to the specific protein.

- Freeze-fractured: Quickly frozen tissue (exposed to liquid nitrogen, -196°C) immobilizes cell components. Once frozen, the cell is fractured with a sharp blow from a cold knife.
- Deep-etching: The specimen is placed in vacuum to remove ice-crystals (sublimation).
- A carbon (vertically) followed by a heavy metal (this one from an angle other than 90°) monolayer are added to the specimen, while the organic support layer is itched away in acid.

Fluorescence M.: (Epi-Fluorescence M - light exposition from top) Light from a multiwavlength or single-wavelength source moves through an excited filter, which allows only the desired wavlength of exciting radiation to pass. This radiation is reflected by the dichromatic filter (between ocular and objective) and focused by the objective lens; most of it passes through the dichromatic filter and is not reflected. A final barrier filter blocks any residual light with the frequency of the exciting radiation, so that only socondary emission from the flourescence particles are sent back to bee seen in the ocular. **Interference Contrast** M.: The light source is split up into reference- and object-beam with the help of Wollaston prisms (double-refracting optical device). The object beam, while passing through the object generates a slight shift in phase (run-time difference), that, when the two beams merge again (in a 2nd

Wollaston prism) interference effects occur, yielding a relief-like specimen. Polarizers help to optimize the image.

Phase Contrast M.: Incident light passes through an annular diaphragm, which focuses a circular annulus (ring) of light on the sample. Light that passes unobstructed through the specimen is focused by the objective lens onto the thick gray ring of the phase plate, which absorbs some of the direct light and alters its phase by ½ of a wavelength. If a specimen refracts or diffracts the light, the phase of some light waves is altered and the light waves are redirected through the thin, clear region of the phase plate. The refracted and unrefracted light are recombined at the image plan to form the image.

Plasma Membrane (zoology) or Plasmalemma (botany) or simply Cell Cortex: Biological envelope (bilayer) that surrounds all cells, plastid, mitochondrion etc.; consists of a single phospholipid bilayer (generally 40% lipid and 60% protein share); typically with a hydrophobic (fatty acid-CH2-tail) and a hydrophilic (organic-phosphate-glycerol group) head. The PM separates electric charges and actively transporting ions via membrane- or ion pumps, regulating the constant flow of materials into / out of the cell, allowing water ,ions, and certain organic molecules to pass through, while allowing toxic or useless by-products of cellular metabolism to exit the cell. All membrane phospholipids are amphipathic, having both hydrophobic and hydrophilic portions and has very flexible and dynamic characteristics (see Fluid-mosaic model).

Osmotic effects (in plants by the vacuole, in erythrocytes by the cytoplasm) do prove that these membranes are permeable (either semipermeable or selective). Osmotic lysis does not necessarily lead to cell death, instead re-sealing of lytic spots can guarantee that cell will be able to withstand short term alterations

Membrane, Components of:

- **Peripheral** proteins superficially floating (either on the inside or outside of the plasma membrane) proteins and are not involved in ion transport; usually receptors or parts of an amplificatory cascade.
- Intrinsic proteins are <u>integrally</u> placed proteins that span the membrane (connecting the extra- with the intracellular "world"), form selective filters, thus able to conduct ions or molecules across it and are mostly used as membrane pumps mechanism that actively transport nutrients into cellular products and waste against a gradient.
- **Lipid** content influences fluidity of the membrane. Especially <u>cholesterol</u> interacts with adjacent phospholipids in the membrane, partially immobilizing fatty acid chains, rendering the membrane less fluid but mechanically stronger; <u>cardiolipin</u>, typical phospholipid found in mitochondria of the heart.

Membrane Permeability: The ease with which substances can pass through a membrane. Gaseous molecules easily diffuse passively across the membrane (michaelis curve), whereas larger molecules need to bee actively transported.

Mathematically expressed as the net flux over time [mol] x, distance [m] across a membrane: c, concentration [mol] $F = D \cdot dc/dx$ D, diff constant [?]

Membrane Skeleton: A network of cytoskeletal proteins (<u>spectrin tetramer</u> - see also cytoskeleton) and their interactions with integral proteins constitute for the mechanical stability of the membrane. **Fluid Mosaic Model**: Current model of cell membrane structure in which proteins (both intrinsic and peripheral) are embedded in the lipid bilayer and can move freely through the fluid-like lipid structure.

Transmembral Proteins: Integral proteins that either consume ATP by **pumping** molecules or ions actively against the concentration gradient, or yield energy by allowing ions or molecules to tunnel through the protein channel to generate ATP (converting KE into PE).

In general two neutral molecules fuse to give rise to two ions with opposite charges, which are far more easily to transport than a non-charged molecule.

Type of transport proteins:

Antiporter: A specific membrane-bound protein capable of cotransport processes in which the movement of a molecule or ion across a membrane against a concentration gradient is driven by the movement in the <u>opposite</u> direction of a second ion down its concentration gradient; e.g.:
 Bohr effect - anion transport across erythrocyte membrane: H₂O + CO₂ ↔ H⁺ + HCO₃⁻
 An anion-Antiporter catalyzes the reversible exchange of Cl⁻ and HCO₃⁻ ions across the membrane and works in conjunction with carbonic anhydrase. In systemic capillaries, the overall reaction causes HCO₃⁻ to be released from the cell, which is essential for CO₂ transport from the tissues to the lungs. In the lungs, the overall reaction is reversed.

 ${\bf Na^+-K^+}$ **Pump**: Membrane mechanism responsible for active extrusion of ${\bf Na^+}$ from the cell at the expense of metabolic energy; there is a 3:2 exchange of intra- ${\bf Na^+}$ for extracellular ${\bf K^+}$. The enzyme possesses a high affinity for ${\bf Na^+}$ -ions on the cytosolic side, which under the influence of ATP, causes a conformational change mediating $3{\bf Na^+}$ -ions toward the exterior side of the membrane. This transport causes ATP to convert to ADP + P, where P remains attached to the enzyme; the dissociation of the $3{\bf Na^+}$ -ions at the exterior face, immediately inhibiting bonding of ${\bf Na^+}$ to the emptied locations but facilitating the attachment of $2{\bf K^+}$ -ions at neighboring binding sites. Once $2{\bf K^+}$ -ions have attached, hydrolysis of the P-atom occurs causing another conformational change of the enzyme, resulting in the transportation of the ${\bf K^+}$ -ions across the membrane into the Cytosol, allowing another $3{\bf Na^+}$ -ions to attach at the cytosolic face.

Acidification of stomach lumen by parietal cells in the gastric lining: The apical membrane of parietal cells contains a H^+/K^+ ATPase as well as Cl^- and K^+ channel proteins. The basolateral membrane contains an anion-antiporter that exchange HCO_3^- for Cl^- ions into the cell. The water inside the cell is split to yield H^+ and OH^- , where the anion combines with the free internal CO_2 to form HCO_3^- . The H^+/K^+ ATPase transports the H^+ out into the gastric lumen in exchange for the potassium to be pumped in. The build up of H^+ causes Cl^- ions to follow passively by diffusion from the cell to form the hydrochloric acid medium needed in the stomach. K^+ uniporters pump the potassium ions back into the lumen to keep the cell's membrane potential constant.

- Symporter: A specific membrane-bound protein capable of <u>cotransport</u> in which two different molecules or ions move across a membrane in the <u>same</u> direction; e.g.:

 Glucose transport from <u>intestinal lumen</u> into the blood: Entry of glucose from the intestinal lumen into the epithelial cell is catalyzed by a Na⁺-glucose symport protein located in the apical surface membrane. The Na⁺/K⁺ ATPase in the basolateral surface membrane generates the Na⁺ gradient, which provides the energy for glucose uptake by pumping out the Na⁺ ions entering the cell by a Na⁺-glucose Symporter. Glucose leaves the cell via a facilitated-diffusion Uniporter in the basolateral membrane.
- **Uniporter**: A specific membrane-bound protein that transport a <u>single</u> type of molecule down its concentration gradient; e.g.:

Stomatal opening and closure: opening of stretch-activated (mechanically triggered) K^+ and Cl^- channels in the plasma membrane of the guard cell is followed by an influx of K^+ and Cl^- . The increase in cytosolic KCl triggers the osmotic influx of water, causing the cell to bulge and opening the stomatal pore.

 Ca^{2+} pump: Ca^{2+} -binding sites are on the cytosolic and exoplasmic faces respectively. An ordered sequence of steps is essential for coupling ATP hydrolysis and the transport of Ca^{2+} -ions across the membrane. The cytosolic side possesses a high affinity for Ca^{2+} -ions; once 2 ions have attached, ATP can be converted to ADP, where P remains bonded to the enzyme. Along with the conformational change, the two Ca^{2+} -ions are transported across the membrane into the exterior face. Once the ions are liberated, hydrolysis of P occurs, causing a second conformational change which allows the cycle to start again.

Porin: A class of transmembrane (pore-forming) proteins which are capable of forming spontaneous channels through which small medium-sized molecules can flow; the channel remains open until the specific substance has passed the barrier - flip-flop mechanism; e.g.: in the lipo-saccharide layer of gram-negative bacteria. Porin-code is encoded in the attacking organism.

Transport triggering mechanism:

- Voltage activated: Achieved by <u>changing</u> the membrane potential; e.g.: neuromuscular junction.
- Ligand activated: A hormone or transmitter activates channel chemically, requiring a receptor site.
- Mechanically activated: <u>Deformation</u> of the membrane activate channels (support by cytoskeleton).

Transmembral Potential: Different ion-concentration on both sides of the membrane results in a voltage gradient; if ion-pumps would not keep up this balance, leakages would result in a neutralization of the charges; e.g.: saltatory propagation of action potential in nerves.

Action Potential (AP): Pertaining to the independence of response magnitude from the strength of the stimulus; response is "all" if the stimulus achieves threshold and "none" if the stimulus fails to achieve threshold:

Steps in potential propagation:, Ca²⁺-ions influx into a nerve cell will trigger an AP once threshold has been reached. At the site of AP, the membrane resting potential (-60mV) becomes more positive (+40mV) due to the opening of Na⁺-channels. Depolarization spreads passively in both directions along the axon but the Na⁺ channels proximal to the nerve cell are still inactivated (refractory period) and cannot be reopened again. Instead Na⁺ channels distal to the AP site of the nerve cell have not yet experienced voltage change, hence **depolarization** will take place once the threshold-level is exceeded opening those Na⁺-channels. The influx of Na⁺-ions causes the axon-potential to *overshoot* until the repolarisation-level is reached (+40mV). In this moment Na⁺-channels close while K⁺-channels open to allow K⁺-ions to rush into the axon gradually *repolarizing* the potential until hyper-polarization is reached (-70mV). There K⁺-channels close again to permit the potential to rise slightly to the resting potential (-60mV). Continuously operating Na⁺/K⁺-pumps transport Na⁺-ions into the extracellular fluid, and K⁺-ions into the axonal Cytosol.

- **Membrane Resting Potential**: (MRP) The normal unstimulated membrane potential of a cell at rest; can be up to -100mV (average membrane potential at rest: -60mV for K⁺-ions) resulting from an unbalanced Na⁺/K⁺ ratio, where for 2K⁺, 3Na⁺-ions are transported out i.e.: dynamic balance of in/outward flowing ions (see Na⁺/K+ pump).
- **Donnan Equilibrium**: Electrochemical equilibrium that develops when two solutions are separated by a membrane permeable to only some of the ions of the solution.

 $E_{x} = \frac{R \cdot T}{z \cdot F} \cdot \ln \frac{C_{i \cdot fix}}{C_{out}} \quad [V]$ $C_{x}, concentration \quad [mol/1]$ $C_{oldmon} \quad F_{guestion} \quad The equation describing the equilibrium. For foreday constant.$

• Goldman Equation: The equation describing the equilibrium potential for a system in which more than one species of diffusible ions are separated by a semipermeable membrane; if only one species can diffuse across the membrane the equation reduces itself to the Nernst equation

$$E_{ions} = \quad \frac{R \cdot T}{z \cdot F} \cdot ln \quad \frac{P_K[K^+]_{out} + P_{Na}[Na^+]_{out} + P_{Cl}[Cl^-]_{in}}{P_K[K^+]_{in} + P_{Na}[Na^+]_{in} + P_{Cl}[Cl^-]_{out}} \quad [V]$$

• <u>Nernst Equation</u>: Equation for calculating the electrical potential difference across a membrane that will just balance the concentration gradient of an ion.

F, faraday constant	[F]
R, gas constant 8.314	[kJ/mol]
T, temperature	[K]
Ex, potential difference	[V]
[X] ion concentration	[mole/l]

P_x, permeability constant [-]

al z, [C] E_x , potential difference [V] X_{out} , X_{in} , int./ext. ion conct.[mole/l]

$$E_x = -\frac{R \cdot T}{z \cdot F} \cdot ln \; \frac{X_{out}}{X_{in}} = 0.058 \cdot log \; \frac{X_{ou}}{X_{in}} \quad \; [V]$$

Starch: (M.E. sterchen, to stiffen) a complex insoluble carbohydrate, the chief food storage substance of plants; composed of a thousand or more glucose units.

Transforming Principle: RNA from a virus (retrovirus) is transferred to the DNA of its host (bacteriophage) to make the host reproduce the viral DNA; in prokaryota, the transforming principle is achieved by the formation of a transduction pilus where replicated plasmids (short stranded DNA-rings) migrate from the positive (+) to the negative (-)