

Cellular Organization: Nucleus

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For PG Classes (Sem. IV- Cytology)



General Introduction

Nucleus first discovered by Robert Brown (1833) in the cells of an 'Orchid' and named it as Nucleus.

It is spherical body in the cells also termed as Karyon. Strasburger (1873) suggested that nuclei arise from pre-existing nuclei. It is controlling centre of cell as it contains chromosomes and genes proved by J. Hammerling (1934) by their grafting experiments on two species of green alga *Acetabularia* (*A. mediterranea* and *A. crenulata*). Usually there is a single nucleus per cell. There may be binucleate (e.g. *Paramecium*) and polynucleate condition also. Polynucleate condition may be because of fusion of a number of cells (Coconut endosperm) or by free nuclear divisions without cytokinesis i.e. coenocytic which is commonly found in plants. In some cells, nucleus is present only at early stage but degenerates at maturity (e.g., sieve tubes of plants, mammalian erythrocytes) due to this reason these cells have a short lifespan.

In Prokaryotes (e.g. bacteria, blue-green algae (Cyanobacteria) and mycoplasma etc. , definite nucleus is absent while incipient nucleus or prokaryon is present (double nuclear membrane is absent).

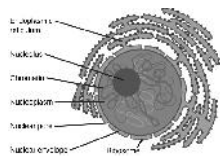


Figure: Nucleus in a eukaryotic cell

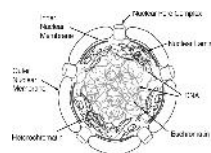
Nucleus is made up of the following Parts

1. Nuclear Membrane
2. Nucleoplasm
3. Nucleolus
4. Chromosomes

Shape of nucleus is variable (spherical, oval, elongated or flattened). The branched nuclei are sometimes present in glandular cells. In some cells nucleus is irregular (e.g. in some leukocytes).

Size of the nucleus is variable (5-30 μ). Size is larger in metabolically active cell as compared to metabolically inactive cell.

Chemically the nucleus consist of proteins (about 70%), phospholipids (about 5%) DNA (about 10%), RNA (about 2-3%).



1. Nuclear membrane (Nuclear envelope or Karyotheca) Enclosing nucleoplasm, double membranal structure and space between these two membranes (outer and inner) is called as 'perinuclear space' of 15 nm or more. The outer nuclear membrane is continuous with the membrane of Endoplasmic Reticulum has attached ribosomes on it. Nuclear pores (about 10 to 100 nm size) on nuclear membrane can pass the molecules or particles of 20-25 nm like ribosome. Nuclear pores are plugged with distinct fibrous material 'annulus' (only in animal cells) Nuclear pore complexes are multiprotein aqueous channels that penetrate the nuclear envelope connecting the nucleus and the cytoplasm.

Below the nuclear envelop present a filamentous network of protein called nuclear lamina, which attach chromosomes to nuclear envelop and to help in the growth of nuclear envelop

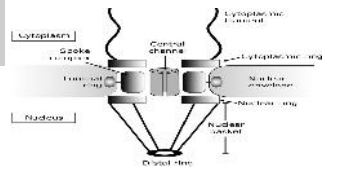
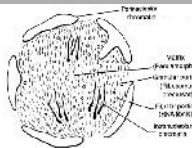


Fig. Nuclear pore complex in nucleus of an animal cell

Nucleolus is made up of four parts:

1. Granular part is made up of ribonucleoprotein granules (r-RNA and proteins are present in ratio 2:1) having a diameter of 150 Å to 200 Å. These granules are also called nucleolar ribosomes
2. Fibrillar part is made up of fibrils of ribonucleoproteins called nucleonema having a diameter of 50 Å.
3. Amorphous matrix or pars amorpha: This is a homogeneous substance having protein granules and fibrils.
4. Chromatin: It is feulgenpositive and is of two types: i. Perinucleolar chromatin which covers the nucleolus and internucleolar chromatin is present inside the nucleolus. ii. Intranucleolar chromatin is present inside the nucleolus.



2. Nucleoplasm: The nucleoplasm has a complex chemical composition inside the nuclear envelope, it is composed mainly of the nuclear proteins but it also contains other inorganic and organic substances such as nucleic acids, proteins (histone and non-histone), enzymes, lipids and minerals. The common nucleic acids of the nucleoplasm are the DNA and RNA, both near curve in the natural molecular state or in the form of the monomer nucleotides. Nucleoplasm is the site to perform various enzymatic activities and the synthesis of nucleic acids (DNA and RNA) and ribosomal subunits. Chromatin materials and nucleolus are present in the nucleoplasm.

3. Nucleolus: It was discovered by Felice Fontana (1781) and termed as nucleolus by Donald Bowman (1840). In the nucleoplasm, it is spherical, dense, colloidal body remains attached with nucleolar organizing chromosomes, visible during the interphase of the cell division under the microscope. The nucleolus is a dynamic structure that assembles around the clusters of rRNA gene repeats during late telophase, persists throughout interphase and then disassembles as cells enter mitosis.

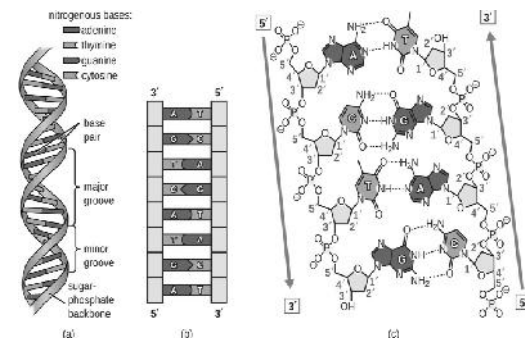


Figure: DNA structure and composition

4. Chromatin threads or Chromosomes

During interphase, chromatin threads are present in the form of a network called chromatin reticulum. At the time of cell division, these thread like structures of chromatin reticulum become visible as independent structures.

E. Strasburger(1875) discovered these distinct structures during cell division.

Wilhelm von Waldeyer (1888) gave the term chromosomes to them because they get stained with basic dyes like basic fuchsin and aceto-carmin. In all eukaryotes, the nucleus contain definite number of chromosomes having definite size and shape.

In all eukaryotes, the nucleus contain definite number of chromosomes having definite size and shape. Chromosome number is variable, highest number of chromosome is in Pteridophyte ($2n=1262$, in *Ophioglossum*). While least chromosome number is in plants ($2n=4$ ($n=2$) in *Haploappus gracilis* (Asteraceae) and in a protozoan, radiolarian, Aulacantha, the diploid number is $2n=1600$.

Where: 'n' represents the gametic or haploid chromosome number.

'2n' is the diploid or somatic number.

'x' is the basic chromosome number or primitive ancestral number in polyploids, e.g. in wheat (hexaploid), $2n=6x=42$, where $n=21$ (genetic haploid number) and $x=7$ (basic chromosome number).

Structure or morphology of Chromosome

Each chromosome consists of two coiled filaments throughout its length, called chromatids by **Vejdovsky**. Chromatids have bead-like structures chromatomers, which are to be the gene-bearing portions of chromosomes.

Chromatid is a half chromosome or daughter chromosome. The two daughter chromatids are connected at the **centromere** or **primary constriction**. Structure of chromosome is clearly visible at metaphase.

The chromosomes are embedded in the non-genetic substance known as matrix which is bounded by a sheath called pellicle. Matrix and pellicle both are **achromatic** and present only at metaphase when nucleolus is disappear.

Besides primary constriction (centromere), in some chromosomes, there is present secondary constriction also and due to this a small portion is pinched off from chromosome body. This portion is known as satellite or trabant. The chromosomes having satellite or trabant are called SAT-chromosomes (SAT- Sine Acid Thymonucleinic, having thymonucleic acid. SAT-chromosomes are used as marker chromosomes. Sometimes two satellites are present in a chromosome these chromosomes are called Tandem SAT-chromosomes. The ends of the chromosomes are called telomere (which do not unite with any other structure).

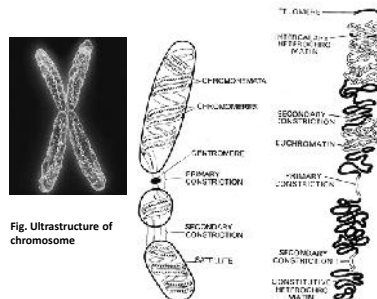


Fig. Ultrastructure of chromosome

Heterochromatin or Euchromatin
These terms were given by Emil Hertz (1928). It was observed that when chromosomes are stained with basic dyes like acetocarmine or feulgen (basic fuchsin), then two types of regions are distinguished. i. Heterochromatic region (genetically inactive region) and ii. Euchromatin region (active region, rich in DNA).

Shape of chromosome can be determined at 'anaphase'

Depending upon the position of centromere or Kinetochore (a point where Spindle fibres attach during cell division) the shape of the chromosome may be:

1. **Telocentric** (the centromere present at one end, the shape of chromosome is rod like)
2. **Acentric** (the centromere is present at terminal position, slightly away from end, one chromosome has a long and another a very short arm)
3. **Sub-metacentric** (centromere is located near the centre, so the chromosome has one long and another slightly shorter arm, the chromosome may be 'L' shaped.
4. **Metacentric** (centromere is present in centre of the chromosome, hence both the arms are equal, 'V' shaped chromosomes).
5. **Acentric** (when centromere is absent)

The pair of **chromosomes** that regulate the somatic characters of the body are known as **autosomes**, whereas the pair of **chromosomes** that determines the sex of an organism, as they regulate the sex-linked traits are known as **sex chromosomes** or **allosomes**

On the basis of sex and other body characters chromosomes in eukaryotes are :

1. **Autosomes** - present in somatic body and
2. **Sex chromosomes** (Heterosomes or Idiosomes)- These chromosomes concerned with sex (are 'X' and 'Y' type, for determination of sex.

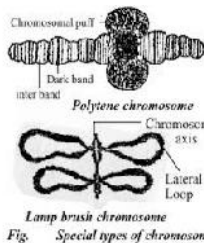
For example, humans have a **diploid genome** that usually contains 22 pairs of autosomes and one **allosome** pair (46 chromosomes total)

B-chromosomes

These are a type of supernumerary chromosomes, that may or may not be present in an organisms as extra chromosomes over and above standard chromosome complement (chromosomes of standard complement are called **A-chromosomes**). These are not found in all individuals of a species and in all cells of an individual. These are generally smaller in size as compared to A-chromosomes and are not homologous with any of the A-chromosomes. They are genetically inert, show non-Mendelian inheritance. Most effect of these chromosomes is on seed and pollen fertility.

Giant chromosomes

- Found in certain tissues e.g., salivary glands of larvae, gut epithelium, Malpighian tubules and some fat bodies, of some **Diptera** (*Drosophila*, *Scara*, *Rhynoscara*).
- These chromosomes are very long and thick (upto 200 times their size during mitotic metaphase in the case of *Drosophila*)
- Hence they are known as **Giant chromosomes**.



Polytene chromosomes or Salivary gland chromosomes: are large chromosomes which have thousands of DNA strands. They provide a high level of function in certain tissues such as salivary glands. **Polytene chromosomes** were first reported by E.G. Babbiani in 1881 in salivary gland cells of insects of Diptera. The size of these chromosomes increases upto 200 times or more than normal. There occurs polyteny i.e. number of chromatids or fibrils increases upto 2000 or more per chromosome. A characteristic feature of these chromosome is that somatic pairing occurs in them and hence their number appears half of normal somatic cells. These chromosomes show distinct **dark and light bands** i.e. chromatic (dark) and achromatic (light) regions. They form puffs or loops (in region of dark bands). Which are called **Babiani puffs** or **Babiani rings** where synthesis of m-RNA occurs.

Lampbrush chromosomes (LBCs) are transcriptionally active chromosomes (length 800-1000 μ) found in the germinal vesicle (GV) of large oocytes of many vertebrate and invertebrate animals and also in the giant single-celled alga *Acetabularia*. Also present in spermatocyte nuclei of fruitfly (*Drosophila*) They were first described by Walther Flemming in 1882. These chromosomes are having a main axis made of DNA on which are present chromatomers and from each chromatomer, 1-9 loops arise in pairs. The loop axis is again made of DNA which is surrounded by a matrix on both side made of RNA and proteins. Due to pair of loops look like a lampbrush hence called Lampbrush Chromosomes.

Ultrastructure:

Chromosomes are made up of very fine fibrils (2 nm-4nm in thickness). As the diameter of DNA molecule is also 2nm, so it is considered that a single fibril is a DNA molecule. It is also seen that chromosome is about a hundred times thicker than the DNA whereas the length of DNA in chromosome is several hundred times that of the length of chromosome. So it is considered that long DNA molecule is present in folded manner which forms a famous model of chromosome structure called 'Famous Fibre Model', given E.J. Dupraw (1965). The most accepted model of chromosome or chromatin structure is the 'nucleosome model', proposed by Kornberg and Thomas (1974).

Nucleosome is a basic unit of DNA packaging in eukaryotes, consisting of a segment of DNA wound in sequence around eight histone protein cores. This structure is often compared to thread wrapped around a spool. Nucleosomes form the fundamental repeating units of eukaryotic chromatin, which is used to pack the large eukaryotic genomes into the nucleus while still ensuring appropriate access to it (in mammalian cells approximately 2 m of linear DNA have to be packed into a nucleus of roughly 10 μm diameter). Nucleosomes are folded through a series of successively higher order structures to eventually form a chromosome; this both compacts DNA and creates an added layer of regulatory control, which ensures correct gene expression. Nucleosomes are thought to carry epigenetically inherited information in the form of covalent modifications of their core histones

- The name nucleosome was given by P. Outter (1975) for these repeating units of chromatin which are present as beads on string.
- The diameter of nucleosome is 12.5 nm.
- The adjacent nucleosomes are joined by inter-nucleosomal DNA or linker DNA and hence can be separated from chromosomes by being treated chromatin with nuclease enzyme.
- According to A. Klug (Nobel Prize winner, 1982), the chain of nucleosomes is further folded or coiled to form a 'solenoid' (having six nucleosomes per turn).

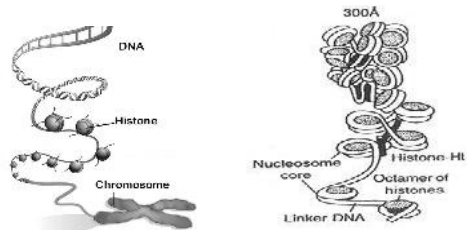


Fig. 45.7. Solenoid structure

The model was proposed on the basis of X-ray diffraction, electron microscopy, nuclease digestion and chemical cross-linking

Nucleosome, i.e. subunit of chromatin is an octamer of histones (2 molecules each of H_2A , H_2B , H_3 and H_4) with DNA of 200 base pairs i.e. the nucleosome is made of core of eight molecules of histones wrapped by double helical DNA with $1\frac{1}{4}$ turns making a repeating unit. Further, this unit is having a single molecule of H_1 histone at the ends.

THANKS