

**Molecular Cell Biology**

**Prof. D. Karunakaran**

**Department of Biotechnology**

**Indian Institute of Technology Madras**

**Module 2**

**Nuclear Structure, Chromatin Packing and Transport, Transport of  
Biomolecules**

**Lecture 2**

**Chromatin Packing**

### **DNA content is same in all cells of an organism**

- DNA is in a complex with proteins such as histones and non-histone chromosomal proteins – together called chromatin.
- The genetic material is same in all the cells in a multicellular organism and that means the genetic material is not lost during differentiation.
- The diploid cells thus contain the same amount of DNA.
- Gametes are haploid and thus contain only half the amount.
- Exceptions are there –some of the liver cells are polyploid having higher DNA content.
- Since the DNA content of cells in an individual/species is constant, it is called C-value.

### **Chromosomes are tightly packed**

- If all 46 chromosomes in a human cell are arranged end-to end, they would stretch up to 2 meters.
- However, the nucleus is only about 6 mm in diameter.
- Hence it is clear that the chromosomes are tightly packed in a small space.
- This is achieved by specialized proteins that bind to and fold the DNA, generating a series of coils and loops that provide higher levels of organization

### **C- Value paradox**

- Eukaryotes have more DNA content than prokaryotes.
- Salamander *Amphiuma* has very high DNA content 94pg.
- Humans have about 3 pg DNA per haploid genome or  $3 \times 10^9$  base pairs.
- This means we can theoretically have 3 million proteins if all of these base pairs are functional.
- Then salamander will have 30 times more no. of proteins and that is not the case.
- This is sometimes called c-value paradox.
- Thus most of the eukaryotic genomes are of non-coding nature.
- DNA content is proportional to the size of the chromosome.

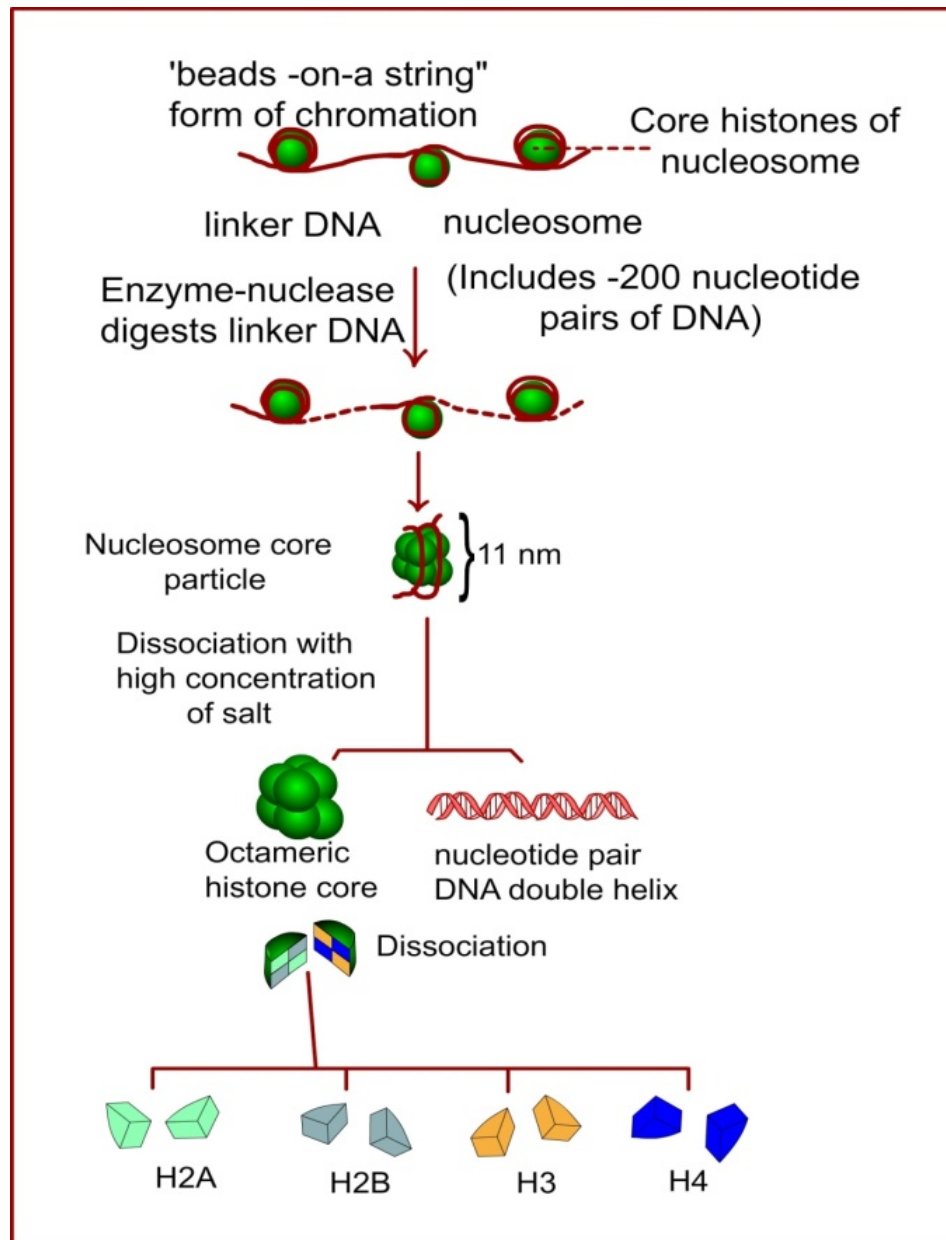
## **Chromatin and nucleosome**

- Chromatin is generally present as a string of 10nm beads.
- However, along with chromatin the shape will be like a fiber.
- The repeating units of this structure are called nucleosome.
- Roger Kornberg in 1974 proposed that the histones are arranged as octamers.
- The histones are H2A, H2B, H3 and H4 and they are similar in many species.
- Histone H1 is however, not conserved among the species and is tissue specific in its expression.
- Nucleosome is 11 nm in diameter and 5.7 nm in height.
- Prokaryotes do not have basic proteins to neutralize the acidic phosphates of DNA but have polyamines.
- Chromatin condensation and decondensation occurs in eukaryotes during cell proliferation but this does not occur in prokaryotes.

## **Nucleosome core particle**

- If chromatin is unfolded partially, under the electron microscope we can see it as a series of "beads on a string".
- The string is DNA, and each bead is a "nucleosome core particle" that consists of DNA wound around a protein core formed from histones.
- Unfolded chromatins by digestion with nucleases break down DNA by cutting between the nucleosomes.
- After digestion for a short period, the exposed DNA between the nucleosome core particles, the linker DNA, is degraded.
- Each individual nucleosome core particle consists of a complex of eight histone proteins-two molecules each of histones H2A, H2B, H3, and H4-and double-stranded DNA that is 147 nucleotide pairs long.
- The histone octamer forms a protein core around which the double-stranded DNA is wound.

## Nucleosome structure



## Histones

- All four of the histones that make up the core of the nucleosome are relatively small proteins (102-135 amino acids), and they share a structural motif, known as the histone fold, formed from three alpha helices connected by two loops.

- In assembling a nucleosome, the histone folds first bind to each other to form H3-H4 and H2A-H2B dimers, and the H3-H4 dimers combine to form tetramers.
- An H3-H4 tetramer then further combines with two H2A-H2B dimers to form the compact octamer core, around which the DNA is wound.
- 142 hydrogen bonds are formed between DNA and the histone core in each nucleosome.
- Nearly half of these bonds form between the amino acid backbone of the histones and the phosphodiester backbone of the DNA.

### **Histone tails**

- Numerous hydrophobic interactions and salt linkages also hold DNA and protein together in the nucleosome.
- For example, more than one-fifth of the amino acids in each of the core histones are either lysine or arginine (two amino acids with basic side chains), and their positive charges can effectively neutralize the negatively charged DNA backbone.
- In addition to its histone fold, each of the core histones has an N-terminal amino acid "tail", which extends out from the DNA-histone core.
- DNA in an isolated nucleosome unwraps from each end at rate of about 4 times per second, remaining exposed for 10 to 50 milliseconds before the partially unwrapped structure recloses.
- In this way most of the DNA in an isolated nucleosome is made available for binding with other proteins

### **Chromatin remodeling**

- In cooperation with histone chaperones, some remodeling complexes are able to remove either all or part of the nucleosome core from a nucleosome.
- This is done by catalyzing either an exchange of its H2A-H2B histones, or the complete removal of the octameric core from the DNA.
- The nucleosome to nucleosome linkages are formed by histone tails, mainly by the H4 tail.

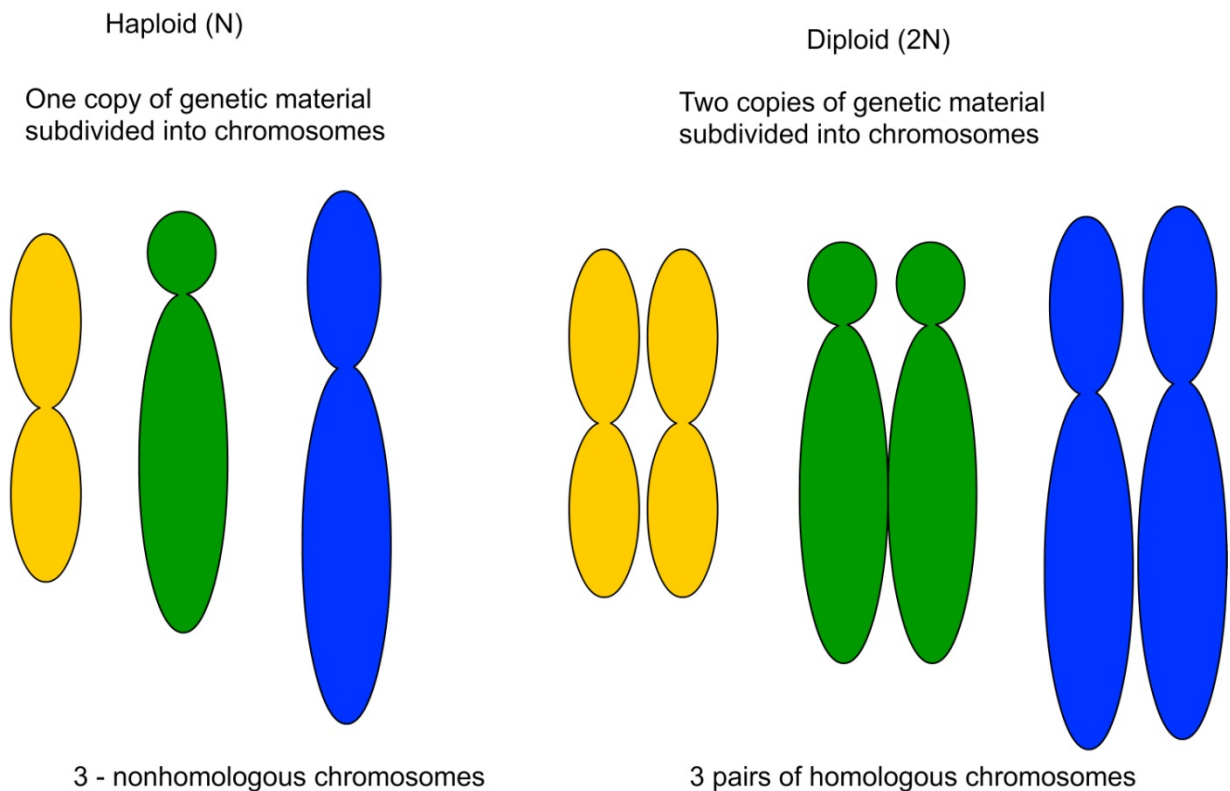
- An additional histone that is known as histone H1 is larger than the individual core histones. This is also important for nucleosome to nucleosome linkages.
- Eukaryotic cells contain a large variety of ATP-dependent chromatin remodeling complexes.
- The subunit in these complexes that hydrolyzes ATP is evolutionarily related to the DNA helicases.
- By using the energy of ATP hydrolysis to move this DNA relative to the core, this subunit changes the structure of a nucleosome temporarily, making the DNA less tightly bound to the histone core.
- Through repeated cycles of ATP hydrolysis, the remodeling complexes can catalyze nucleosome sliding.
- By sliding the nucleosome core along the DNA double helix, nucleosomal DNA is made available to other proteins in the cell.

## **Chromosomes**

- As a 30-nm fiber, the typical human chromosome would still be 0.1 cm in length and able to span the nucleus more than 100 times.
- Clearly, there must be a still higher level of folding, even in interphase chromosomes.
- This higher-order packaging almost certainly involves the folding of the 30-nm fiber into a series of loops and coils.
- This chromatin packing is fluid, frequently changing in response to the needs of the cell.
- The chromosomes from nearly all eukaryotic cells become readily visible by light microscopy during mitosis, when they coil up to form highly condensed structures.
- This condensation reduces the length of a typical interphase chromosome only about tenfold, but it produces a dramatic change in chromosome appearance.
- The two daughter DNA molecules produced by DNA replication during interphase of the cell-division cycle are separately folded to produce two sister chromosomes, or sister chromatids, held together at their centromeres.

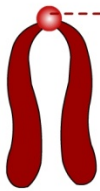
- Chromatid: one of the two copies of a replicated chromosome that is joined at the centromere to the other copy. The two identical chromatids are called sister chromatids.
- Centromere: the chromosomal region that holds sister chromatids together and where the kinetochore forms.
- Kinetochore: the centromeric substructure that binds microtubules and directs chromosome movement in mitosis.
- Gene: a segment of DNA encoding a functional RNA or protein product.

### Chromosomal organization of haploid and diploid organisms

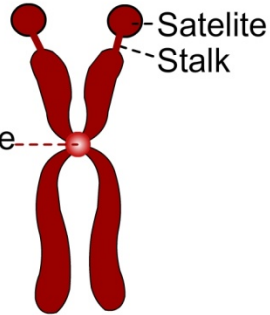


## Types of Chorosomes

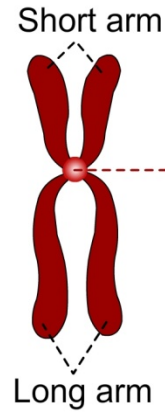
Telocentric



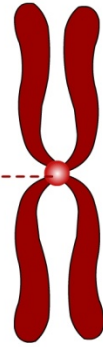
Acrocentric



Submetacentric

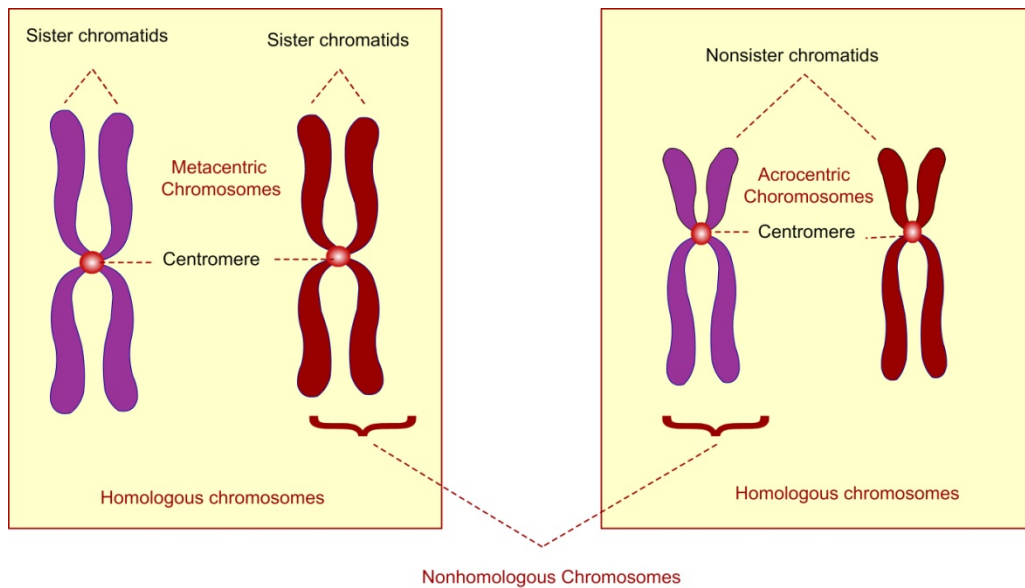


Metacentric



## Metaphase Chromosomes

Chromosomes have duplicated and appear rod like





## Heterochromatin

- As mentioned earlier there are two types of chromatin in the interphase nuclei of many higher eukaryotic cells: a highly condensed form, called heterochromatin, and all the rest, which is less condensed, called euchromatin. Heterochromatin represents an especially compact form of chromatin.
- Heterochromatin is present in many locations along chromosomes but more than ten percent of the genome is highly concentrated in specific regions, most notably at the centromeres and telomeres
- When a gene from euchromatin is experimentally shifted into a region of heterochromatin, it fails to express, and the gene is said to be silenced.
- These differences in gene expression are called **position effects**, since the activity of a gene depends on its position relative to a nearby region of heterochromatin on a chromosome

## Chromosomal loops

- DNA in human chromosomes is organized into loops.
- A loop can contain between 50,000 and 200,000 nucleotide pairs of DNA.
- Highly folded loops of chromatin expand to occupy an increased volume when a gene within them is expressed.
- The position of a gene in the interior of the nucleus changes when it becomes highly expressed.
- A very actively transcribed region often extends out of its chromosome as if in an extended loop

## Minor organelles in the nucleus

- Nucleus is very heterogeneous, with functionally different regions.
- Portions of chromosomes can move to these regions when their gene expression changes.
- Many minor organelles are also present inside the nucleus.
- For instance, spherical structures called Cajal bodies and interchromatin granule clusters are present in many plant and animal cells.

- They contain selected protein and RNA molecules that bind together to create networks that are highly permeable to other protein and RNA molecules in the surrounding nucleoplasm.

### Study Questions

1. What is a centromere?
2. What is meant by c-value paradox?
3. The total number of chromosomes in a human cell are  
a) 30    b) 90    c) 46    d) 26
4. Match the following

Histone	Cell division
Telocentric	Octamers
Mitosis	Cajal bodies
Nucleus	Chromosomes

5. The repeating unit of a chromatin is called -----