Faculty Science

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B.Sc II Paper-II(Cytology, Genetics, Evolution& Ecology)

Unit-I Topic- Chromosome structure, nucleosome and solenoid model

E. Strasburger in 1875 discovered thread like structures which appeared during cell division. In eukaryotes, the DNA in the nucleus is divided between a set of different chromosomes. The human genome approximately 3.2 x 10^9 nucleotides are distributed over 24 different chromosomes. **Each chromosome consists of a single, enormously long linear DNA molecule associated with proteins that fold and pack the fine DNA thread into a more compact structure.** The complex of DNA and protein is called chromatin (from the Greek chroma, "color," because of its staining properties) by **Walther Flemming** in 1878. The present name **chromosome** was coined by **Waldeyer** in 1888**.** A human cell contains 3 X 10^9 bp per haploid set of chromosomes. The average thickness of each base pair is 3.4 A˚. If the DNA molecules in a haploid set of chromosomes were laid out end to end, the total length of DNA would be 10^{10}A˚, or 1 m. For a diploid cell (as human cells typically are), this length is doubled to 2 m. The diameter of a typical human cell nucleus is only 10– 15 µm, it is obvious that the DNA must be compacted by many orders of magnitude to fit in such a small space. Chromosomes are also associated with many proteins and RNA molecules required for the processes of gene expression, DNA replication, and DNA repair. Bacteria carry their genes on a single, circular DNA molecule. This DNA is associated with proteins that package and condense the DNA, but they are different from the histone proteins. The bacterial chromosome does not have the same structure as eukaryotic chromosomes. With the exception of the germ cells and a few highly specialized cell types that cannot multiply and lack DNA altogether (for example, red blood cells), each human cell contains two copies of each chromosome, one inherited from the mother and one from the father. The maternal and paternal chromosomes of a pair are called homologous chromosomes (homologs). The only nonhomologous chromosome pairs are the sex chromosomes in males, where a Y chromosome is inherited from the father and an X chromosome from the mother.

DNA hybridization is a technique in which a labeled nucleic acid strand serves as a "probe" that localizes a complementary strand. This technique can be used to distinguish these human chromosomes by "painting" each one a different color. Chromosome painting is typically done at mitosis, when chromosomes are especially compacted and easy to visualize. Another more traditional way to distinguish one chromosome from another is to stain them with dyes that reveal a striking pattern of bands along each mitotic chromosome. The pattern of bands on each type of chromosome is unique, and it is these patterns that initially allowed each human chromosome to be identified and numbered. The number, sizes and shape of metaphase chromosomes constitute **the karyotype**, which is distinctive for each species. The display of the 46 human chromosomes at mitosis is called the human karyotype. The diagrammatic representation of a karyotype or morphological characteristics of the chromosomes of a species is known as **idiogram**. **A gene is a nucleotide sequence in a DNA molecule that acts as a functional unit for the production of a protein, a structural RNA, or a catalytic or regulatory RNA molecule.** In eukaryotes, protein-coding genes are usually composed of a string of alternating introns and exons associated with regulatory regions of DNA. A chromosome is formed from a single, enormously long DNA molecule that contains a linear array of many genes.

Chromosome number: There are normally two copies of each chromosome present in every somatic cell. The number of chromosomes (N) in such a cell is known as its haploid number, and the total number of chromosomes (2N) is its diploid number. The suffix 'ploid' refers to chromosome 'sets'. The haploid set of the chromosome is also known as the genome. Structurally, eukaryotes possess large linear chromosomes unlike prokaryotes which have circular chromosomes. In Eukaryotes other than the nucleus chromosomes are also present in mitochondria and chloroplast. The number of chromosomes in each somatic cell is same for all members of a given species. The organism with lowest number of chromosomes is the nematode, *Ascaris megalocephalusunivalens* which has only two chromosomes in the somatic cells (2n=2). In the radiolarian protozoan *Aulacantha* diploid number of chromosomes (2N) is 1600. In plants chromosome number varies from **2N=4 as in *Haplopappus gracilis*** (a flowering plant of family Asteraceae) to **2N=> 1200 as in *Ophioglossum reticulatum*** (a fern).

2N = Number of Chromosomes in somatic cells

N = Number of Chromosomes in gametic cells

X = The basic chromosome number of an organism

e.g. In human 2N= 2X= 46(44 Autosomes + 2 Sex chromosome)

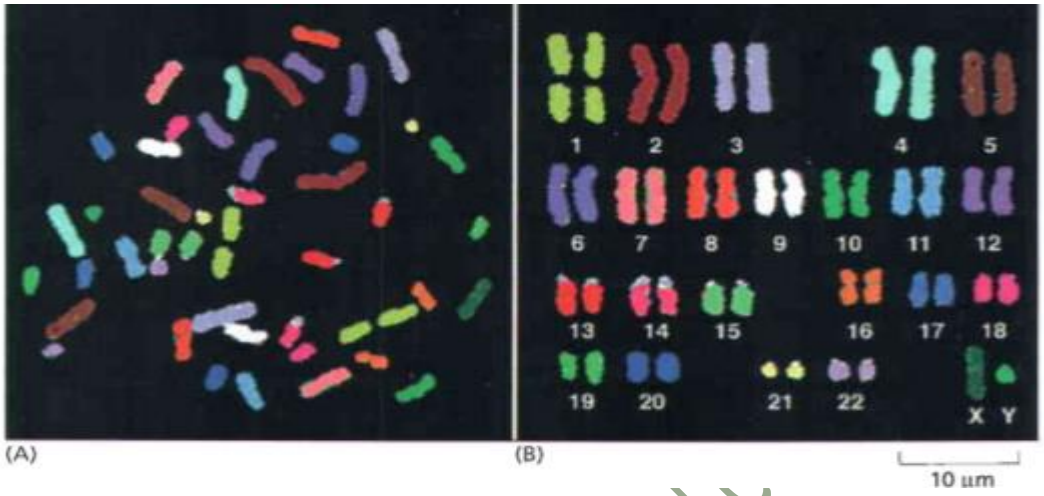
Male gamete: N = 22 + Y or N = 22 + X , Female gamete: N = 22 + X

In *Triticum aestivum* (Hexaploid Wheat) : 2N =6X = 42

N = 21(Haploid chromosome number), X = 7 (Basic chromosome number)

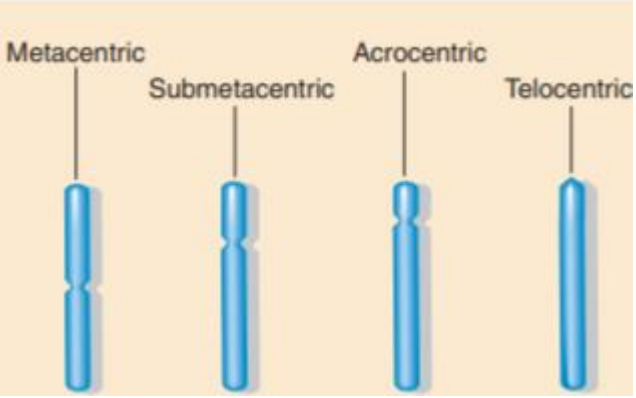
Morphology:

Size: The size of chromosome is normally measured at mitotic metaphase. The most mitotic chromosome falls in the range of 3µm in Drosophila to 5µm in human and 8-12µm in maize. The monocots contain large sized chromosomes as compared to dicots. Organisms with less number of chromosomes contain comparatively large sized chromosomes. The chromosomes in set vary in size.



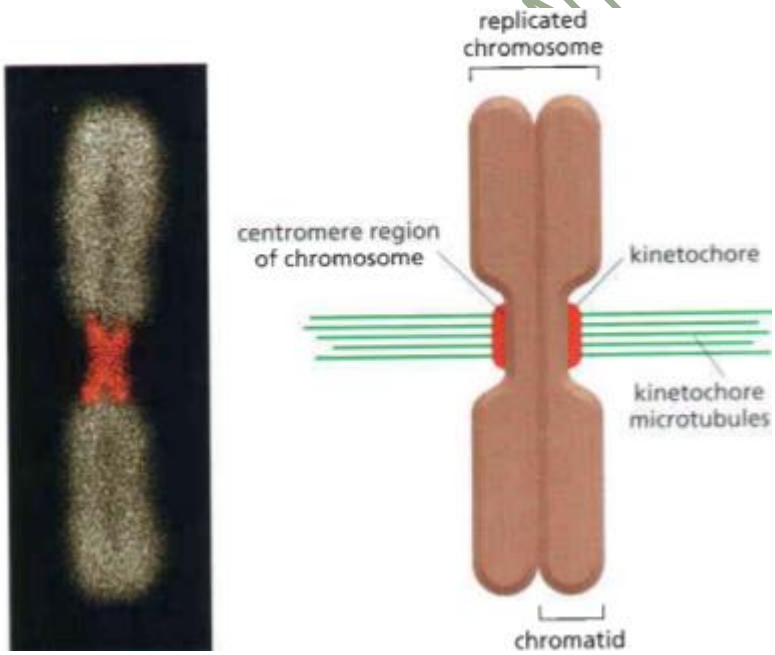
The complete set of human chromosomes

Shape: The shape of the chromosome changes from phase to phase in the continuous process of cell growth and cell division. During the resting/interphase stage of the cell, the chromosomes occur in the form of thin, coiled, elastic and contractile, thread like stainable structures, the chromatin threads. In the metaphase and the anaphase, the chromosome becomes thick and filamentous. Each chromosome contains a primary constriction known as centromere. The centromere divides the chromosome into two parts and each part is called chromosome arm. The position of centromere varies from chromosome to chromosome providing it a different shape. A metacentric chromosome (V shaped) has the centromere at about the center, so the chromosome appears to have two approximately equal arms. Submetacentric chromosomes (L or J shaped) have one arm longer than the other, acrocentric chromosomes have one arm with a stalk and often with a "bulb" (called a satellite) on it, and telocentric chromosomes (Rod shaped) have only one arm, because the centromere is at the end.



Types of chromosomes on the basis of position of centromere

Structure of Chromosome: A chromosome at mitotic metaphase consists of two symmetrical structures called chromatids. Each chromatid contains a single DNA molecule and both chromatids are attached to each other by centromere (or primary constriction) and become separated at the beginning of anaphase. The chromomeres are bead like accumulations of chromatin material that are sometimes visible along interphase chromosomes. The chromomere bearing chromatin has an appearance of a necklace in which several beads occur on a string. Chromomeres are regions of tightly folded DNA and become especially prominent in polytene chromosomes.



A Metaphase Chromosome

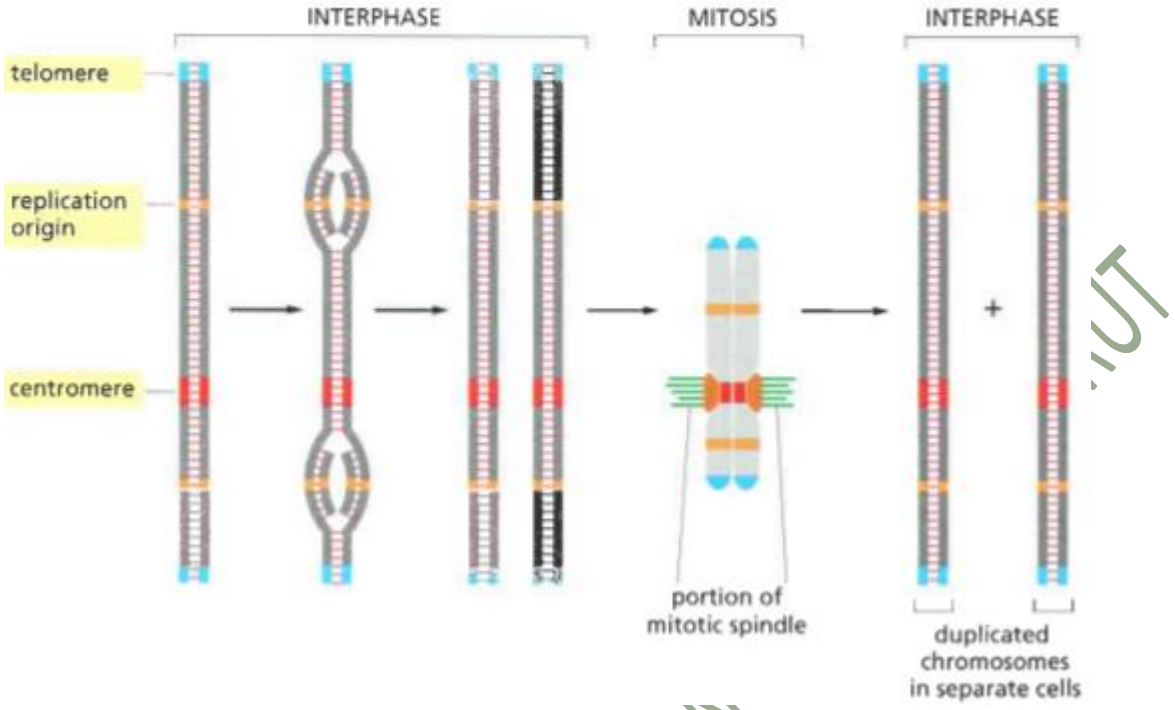
Besides the primary constrictions or centromeres, chromosomes also consist of secondary constriction at any point of the chromosome and are constant in their position and extent. These constrictions are helpful in identifying particular chromosomes in a set. Chromosomes also contain nucleolar organizers which are

certain secondary constrictions that contain the genes coding for 5.8S, 18S and 28S ribosomal RNA and induce the formation of nucleoli. Sometimes the chromosomes bear round, elongated or knob like appendages known as satellites. The satellite remains connected with the rest of the chromosomes by a thin chromatin filament. The important DNA elements in eukaryotic chromosomes include origins of replication, centromere and telomeres are required for chromosome maintenance. Each eukaryotic chromosome consists of two telomeres, one centromere, and many origins of replication. **Origins of replication** are located throughout the length of each chromosome. These are the sites at which the DNA replication machinery assembles and replication is initiated. They are typically found some 30–40 kb apart throughout the length of each eukaryotic chromosome. Prokaryotic chromosomes typically have only a single site of replication initiation. In general, origins of replication are found in the non-coding regions.

Centromere or primary constriction is required for the correct segregation of the chromosome after DNA replication. The two copies of each replicated chromosome are called sister chromosomes, and during cell division they must be separated with one copy going to each of the two daughter cells. Centromere in a chromosome contain specific DNA sequences with special proteins bound to them, forming a disc shaped structure, called **kinetochore**. The kinetochore assembles at each centromere DNA (CEN DNA), and before chromosome segregation, the kinetochore binds to protein filaments called microtubules that eventually pull the sister chromosomes away from each other and into the two daughter cells. The chromosomes of most organisms contain only one centromere and are known as monocentric chromosomes. Some species have diffused centromeres, with microtubules attached along the length of the chromosomes and are termed holocentric chromosomes (e.g. in nematode Ascaris). Centromeres vary greatly in size. In the majority of eukaryotes, centromeres are 40 kb and are composed of largely repetitive DNA sequences.

Telomeres are located at the two ends of a linear chromosome. Like origins of replication and centromeres, telomeres are bound by a number of proteins. In this case, the proteins perform two important functions. First, telomeric proteins distinguish the natural ends of the chromosome from sites of chromosome breakage and other DNA breaks in the cell. Ordinarily, DNA ends are sites of frequent recombination and DNA degradation. The proteins that assemble at telomeres form a structure that is resistant to both of these events. Second, telomeres act as specialized origins of replication that allow the cell to replicate the ends of the chromosomes. Telomeres facilitate end replication through an unusual DNA polymerase called telomerase. Most telomeres have a simple

repeating sequence that varies from organism to organism. This repeat is typically composed of a short TG-rich repeat

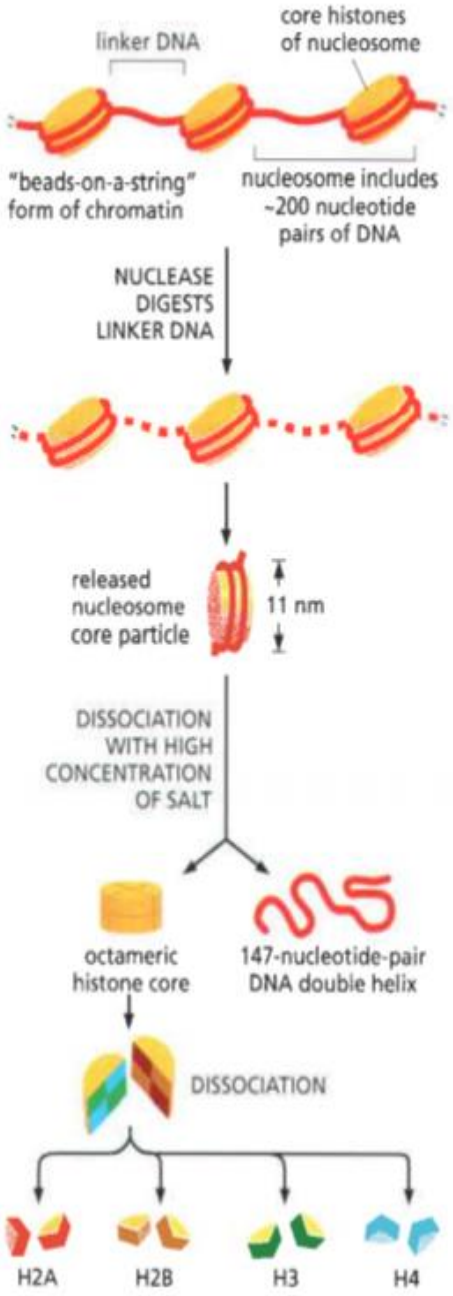


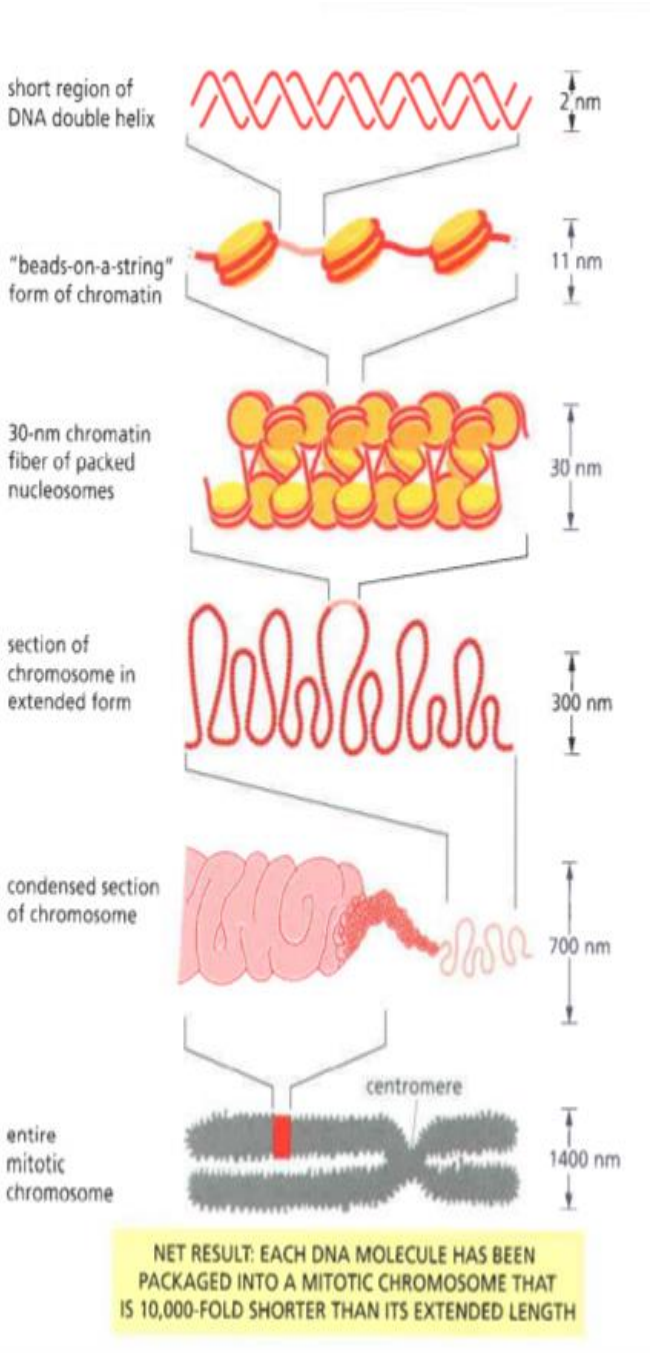
The three DNA sequences required to produce a eukaryotic chromosome

Chromatin: Chromatin consists of DNA, RNA and protein. During interphase the highly condensed and darkly stained chromatin called as **heterochromatin**. The chromatin which is lightly stained and which remains extended was called **euchromatin (see topic-Nucleus)**. The proteins of chromatin are of two types: histones and non-histones. Histones are basic proteins as they are rich in arginine and lysine (basic amino acids). At physiological pH they are cationic and can interact with anionic nucleic acids. They form a highly condensed structure. The histones are of five types called H1, H2A H2B, H3, and H4-which are very similar among different species of eukaryotes and have been highly conserved during evolution. H1 is the least conserved among all and is also loosely bound with DNA. Non-histones: In addition to histones the chromatin comprise of many different types of non-histone proteins, which are involved in DNA replication and gene expression. They display more diversity or are not conserved. They may also differ between different tissues of same organism. Roger Kornberg in 1974 described the basic structural unit of chromatin known as **the nucleosome**.

The first level of packing- Winding of DNA around a protein core to produce a "bead-like" structure called a nucleosome. This gives a packing ratio of about 6. This structure is invariant in both the euchromatin and heterochromatin of all chromosomes. One nucleosome consists of two copies of each H2A, H2B, H3 and H4 complexed with 200 bps of DNA and outside H1. After partial digestion

with nuclease linker DNA and H1 is released and nucleosome core particle is left. A nucleosome core particle consists of histone octamer and 147 bp of DNA is wrapped around it. A segment of 1.7 turn of B DNA is wrapped around the histone octamer in left-handed superhelix.

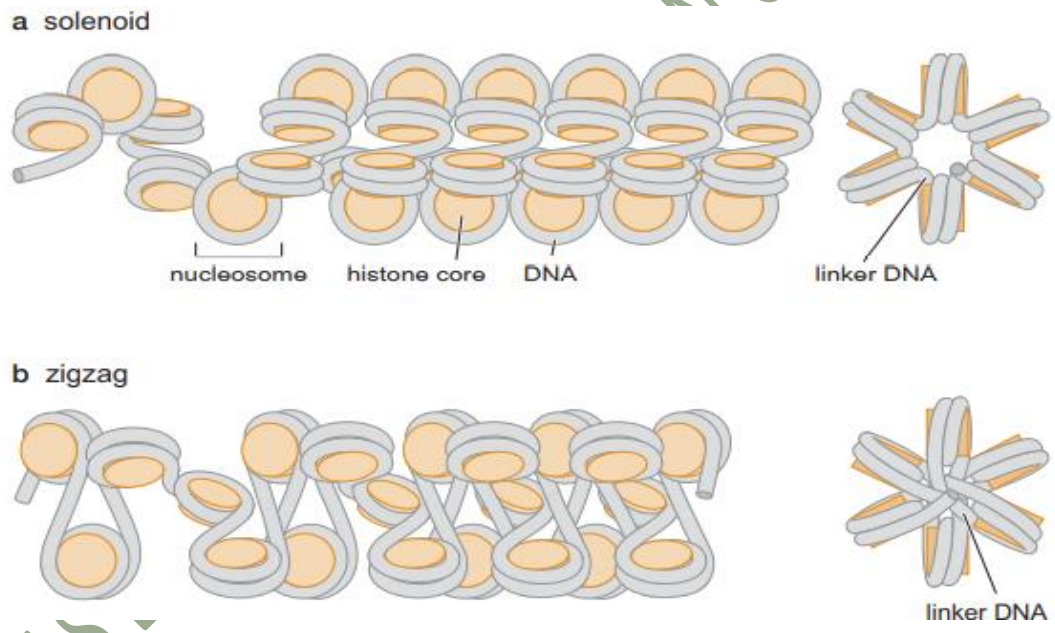




Structural organization of the nucleosome **Chromatin packing**

The second level of packing- Coiling of beads in a helical structure called the 30 nm fiber that is found in both interphase chromatin and mitotic chromosomes. The H1 is important for the stabilization of the 30 nm structure. Binding of H1

stabilizes higher-order chromatin structures. There are two models for the structure of the 30-nm fiber. **In the solenoid model**, the nucleosomal DNA forms a superhelix containing approximately six nucleosomes per turn with a helical pitch of 11 nm. The 30-nm fiber is composed of nucleosome discs stacked on edge in the form of a helix. In this model, the flat surfaces on either face of the histone octamer disc are adjacent to each other, and the DNA surface of the nucleosomes forms the accessible surface of the superhelix. The linker DNA is buried in the center of the superhelix, but it never passes through the axis of the fiber. The linker DNA circles around the central axis as the DNA moves from one nucleosome to the next. An alternative **"zigzag" model** is based on the zigzag pattern of nucleosomes formed upon H1 addition. In this case, the30-nm fiber is a compacted form of these zigzag nucleosome arrays. A recent X-ray structure of the spring-like nature of isolated 30-nm fibers support the zigzag model. Unlike the solenoid model, the zigzag conformation requires the linker DNA to pass through the central axis of the fiber in a relatively straight form. Together, the packaging of DNA into nucleosomes and the 30-nm fiber results in the compaction of the linear length of DNA by 40-fold.



a.Solenoid model, b. Zigzag model

The final level of packaging- The final level of packaging is characterized by the 700 nm structure seen in the metaphase chromosome. The 30-nm fibers are further folded to form large supercoiled loops or domains. The diameter of these looped domains is 80-100nm. These loops are anchored to non-histone protein scaffolding also called nuclear matrix. This protein scaffolding may be nuclear matrix, or may be an enzyme **topoisomerase II**. Topoisomerase II might be helping in untangling of DNA if the DNA gets intertwined. Attachment occurs at

DNA sequences called as MARs or SARs – for matrix or scaffold attachment regions. These regions contain a predominance of A-T rich sequences. The metaphase chromosome appears to be composed of these numerous loops giving a fuzzy appearance. The presence of topoisomerase II at the bottom of each loop would ensure that the loops are topologically isolated from one another. The **SMC (Structural Maintenance of Chromosomes)** proteins are key components of the machinery that condenses and holds sister chromatids together after chromosome duplication. **Cohesin** is an SMC-protein-containing ring-shaped complex that is required to link the two daughter DNA duplexes (sister chromatids) together after DNA replication. The chromosome condensation that accompanies chromosome segregation also requires a related SMC-containing complex called **condensin**. Condensin is also a ring-shaped complex. It may use its ring-like nature to induce chromosome condensation. The associations of these proteins with the nuclear scaffold may serve to enhance their functions by providing an underlying foundation for their interactions with chromosomal DNA. The fiber is organized in loops, scaffolds and domains that give a final packing ratio of about 1000 in interphase chromosomes and about 10,000 in mitotic chromosomes. In interphase cells, chromosomes are localized to largely non overlapping **"territories"** in the nucleus.

Reference

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