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Degree programme: M.sc (PBG)

Year: 1<sup>st</sup> year

Semester: 2<sup>nd</sup> semester

Course name: Principles of Cytogenetics

Course code: GP.502 (2+1)

Topic name:

1. Chromosome classification
2. Chromosome banding

Date: 29/03/2020

Course instructor name: Dr. Nilanjaya

## Chromosome classification

A functional chromosome has three essential elements: a centromere, a pair of telomeres, and origins of replication.

The **centromere** appears as a constricted region on the chromosome. It serves as the attachment point for spindle microtubules—the filaments responsible for moving chromosomes in cell division. Before cell division, a multiprotein complex called the kinetochore assembles on the centromere; later, spindle microtubules attach to the kinetochore.

Centromeres could be classified as follows

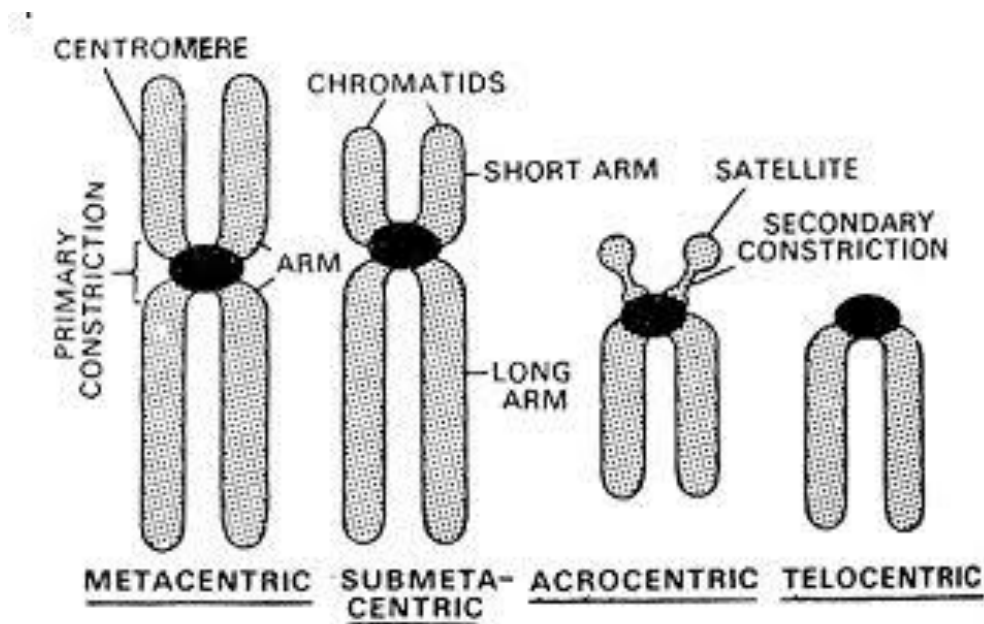
1. Localized centromeres: constitutes the normal condition in which a chromosome possesses permanently localized region to which spindle fiber attaches during chromosome movement
2. Neocentromeres : this are formed under certain conditions in which the centromere region is replaced by a secondary center of movement
3. Non localized centromeres : are those in which the spindle attachment is not confined to a strictly localized chromosome area
  - a. Polycentromeres: each chromosome is attached by many spindle fibers. ex:- Ascarid nematodes
  - b. Holocentromeres: this centromeres are diffuse in nature where every point along the chromosome shows the centromeric activity ex:-Hemiptera, Homoptera.

**Telomeres** are the specific DNA sequences and associated proteins located at the tips of whole linear chromosomes. Just as plastic tips protect the ends of a shoelace, telomeres protect and stabilize the chromosome ends. If a chromosome breaks, producing new ends, the chromosome is degraded at the newly broken ends. Telomeres provide chromosome stability. Research shows that telomeres also participate in limiting cell division and may play important roles in aging and cancer.

Origins of replication are the sites where DNA synthesis begins; unlike centromeres and telomeres, they are not easily observed by microscopy.

**On the basis of the location of the centromere, chromosomes are classified into four types:**

1. **Metacentric:** chromosome with a centromere located in middle of chromosome; two arm are almost equal
2. **Submetacentric :** chromosome with a centromere located slightly away from the mid-point
3. **Acrocentric:** centromere occupying sub terminal position
4. **Telocentric:** centromere at its terminal position



**On the basis of function chromosome are classified into**

1. **Autosomes :** this are responsible for determination of body parts and function
2. **Sex chromosomes :** usually there will be one pair of sex chromosomes in every organism which determine sex of the individual

**On basis of number of centromere chromosomes are classified into**

1. **Monocentric** : having one centromere only
2. **Dicentric** : having two centromere , which is found in some species of wheat
3. **Polycentric** : having more than two centromere, found in some round worms
4. **Acentric**: chromosome without centromere.

## **Chromosome banding**

Chromosome band is defined as a part of a chromosome which is clearly distinguishable from its adjacent segments by appearing darker or lighter with various banding methods.

### **Why we need to study banding pattern?**

Chromosome banding allow us to see and analyze smaller pieces of the chromosome, which in turn help us to identify smaller structural chromosome abnormalities not visible on a routine analysis

### **Classification of banding techniques**

Based on

- GC and AT rich regions.
- Constitutive heterochromatin region

Note: Always chromosomes harvested at metaphase stage which are highly condensed and whose diameter is long enough are used for chromosome banding studies which is achieved by treating cells with tubulin inhibitor, such as colchicine or demecolchicine ,that depolymerize the mitotic spindle and so arrest the cell at this stage

### **Different banding techniques**

- Q(Quinarcine) banding-Casperson et.al(1958)
- C(Centromic/Constitutive heterochromatin)banding-Linde and Laursen(1971)
- G(Giemsa) banding-Summer et.al(1973)
- R(Reverse) banding

#### **1. Q(Quinarcine) banding**

A fluorochrome, quinacrine mustard, was the first compound to reveal a reproducible banding structure, or Q-banding, within metaphase chromosomes.Q-banding results in bright-fluorescent bands against a dull-fluorescent background.

## 2. C(Centromeric/Constitutive heterochromatin)banding

C-banding procedure clearly differentiate between euchromatin and heterochromatin in metaphase chromosomes.

## 3. G(Giemsa) banding

G banding reveals Chromomere-like differentiation along the length of the chromosomes.

## 4. R(Reverse) banding

Preferentially stains euchromatin in a way that was the reverse of G-banding.

### Chromosome-Banding Techniques

Name of Technique	Basis of Technique	Organism Studied
Q-banding	Binding of AT-specific fluorochromes such as quinacrine and 4',6-diamidino-2-phenylindole (OAPI), to DNA	Reptiles, birds, mammals
G-banding	Giemsa staining after incubation in warm Sodium chloride and sodium citrate or trypsin	Fish, amphibia, reptiles, birds, mammals
R-banding	Giemsa staining after incubation in hot buffer	Mammals
C-banding	Giemsa staining after alkali treatment	Most plants and animals

G- and Q-banding procedures for mitotic-metaphase chromosomes appear to reveal the chromomere structure that was seen previously only in prophase chromosomes. The G bands of chromosomes are most commonly observed by treating chromosome preparations in either saline sodium citrate solution or the enzyme trypsin, followed by staining with Giemsa dye. G-Banding generally provides a better resolution of chromosome substructure than other procedures and, in prophase chromosomes, may reveal as many as a thousand bands per genome. The locations of G bands in prophase chromosomes coincide with the locations of chromomeres in conventional preparations.

Mechanism of G-banding is unclear, but the G bands appear to be the sites of AT-rich DNA and DNA that tends to replicate later in the cell cycle. G bands are thought to be relatively scarce in DNA sequences coding for genes, and they may contain proteins, specific for AT-rich DNA, that may be responsible for the differential staining reaction. The regions between G bands are representative of a short, interspersed type of repetitive DNA sequence.

Chromosomes treated at high temperatures (such as 86°C) in a salt solution, followed by staining with Giemsa dye, develop R bands, which seem to correspond to regions between chromomeres and thus are the reverse of G-banding.

The technique of C-banding is applied universally to reveal the location of heterochromatin in mitotic-metaphase chromosomes. In a typical procedure, chromosomes that have been spread on glass slides are sequentially treated with an acid or alkali, followed by incubation in a salt buffer and staining with Giemsa. The C bands correspond to the locations of constitutive heterochromatin in pachytene chromosomes.

### **Uses of Chromosome Banding**

G-and R-banding are the most commonly used techniques for chromosome identification (karyotyping) and for identifying abnormalities of chromosome number, translocations of material from

one chromosome to another, and deletions, inversions or amplifications of chromosome segments. Comparisons of chromosome banding patterns can confirm evolutionary relationships between species and also reveal changes in karyotype that may have been important in speciation.