Botany

M.Sc. (Semester IV)

M. BOTEC-1

(Unit 1)

Topic - Chromosome banding
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Chromosome banding

It refers to alternating light and dark regions along the length of a chromosome, produced after staining with a dye. A band is defined as the part of a chromosome that is clearly distinguishable from its adjacent segments by appearing darker or lighter with the use of one or more banding techniques.

In the late 1960s Caspersson postulated that differences in DNA base composition might produce differential intensity patterns along the length of chromosomes when fluorescent DNA binding dyes were applied to chromosome spreads and thus the concept of chromosome banding was born. Buskholder and Weaver (1977) reported that differences in the binding of non-histones to DNA in different segments of the metaphase chromosome may be involved in mechanism of G-and C banding. They have authored an excellent paper on DNA Protein interactions and chromosome banding in Experimental Cell Research.

Why to study banding pattern?

This allows us to see smaller pieces of the chromosome so that we could identify smaller structural chromosome abnormalities not visible on routine analysis.

Groups of banding techniques

Bands distributed along the length of whole chromosome e.g (i) G-band (ii) Q-band (iii) R-bands	stain a restricted number of specific bands or structures. They reveal Centromeric bands, C-bands and nucleolus organizing regions.
G-and R- bands can be Bright field or Fluorescent	C- banding methods donot permit identification of every chromosome in the somatic cell complements but can be used to identify specific chromosomes.

Bright field G-bands-

They take their name from the Giemsa dye, but can be produced with other dyes. In G- bands, the dark regions tend to be heterochromatic, late replicating and AT rich. The bright regions tend to be euchromatic, early replicating and GC rich.

Bright field R- bands-

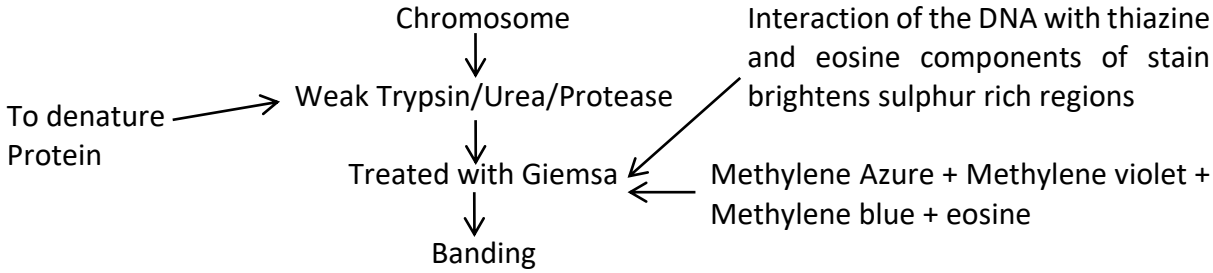
These R-bands are approximately the reverse of G-bands (R-reverse). The dark regions are euchromatic and the bright regions are heterochromatic.

Flouresent G-and R bands-

These bands are the photographic negative of bright field versions I.e. the reverse of the bright field G-bands and R-bands.

Q- bands are like fluorescent G-bands but certain heterochromatic regions are more brightly stained with Q-banding. Q banding uses a stain called quinacrine.

G-banding techniques



Advantages-

1. Used in identification of bands rich in sulphur content.
2. Used in the identification of chromosomal abnormalities.
3. Gene mapping
4. G- bands may reflect stronger chromatin condensation.

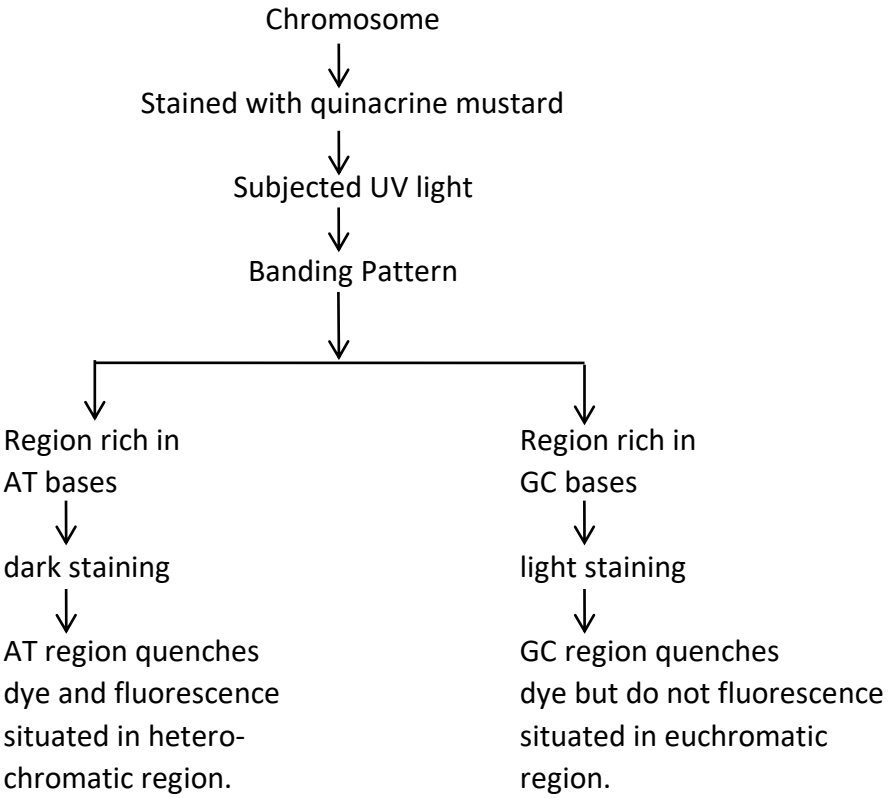
Disadvantages-

it is not suitable for plant chromosome. It requires pretreatment by salt or proteolytic enzyme.

G- Banding not used in Plants.

1. Plant mitotic metaphase chromosome is 10 times more shorter than human chromosome. Hence difficult to demonstrate the arrangement of bands at this level of saturation with G banding technique.

Q-banding techniques



Advantages

a) Simple and versatile
b) Used where G- bands ares not accepted
c) Used in study of chromosome heteromorphism
d) This is used to identify human and mice chromosome

Disadvantages

- Tendency to fade during examination Photodegrdation
- Uv light breaks the chemical bond

C-banding techniques

C-banding represents the constitutive heterochromatin and the banding is caused by differential staining reactions of the DNA of heterochromatin and euchromatin.

Constitutive heterochromatin is located near the centromere, at telomeres and in the nucleolar organizer regions. It is composed of highly repetitive DNA.

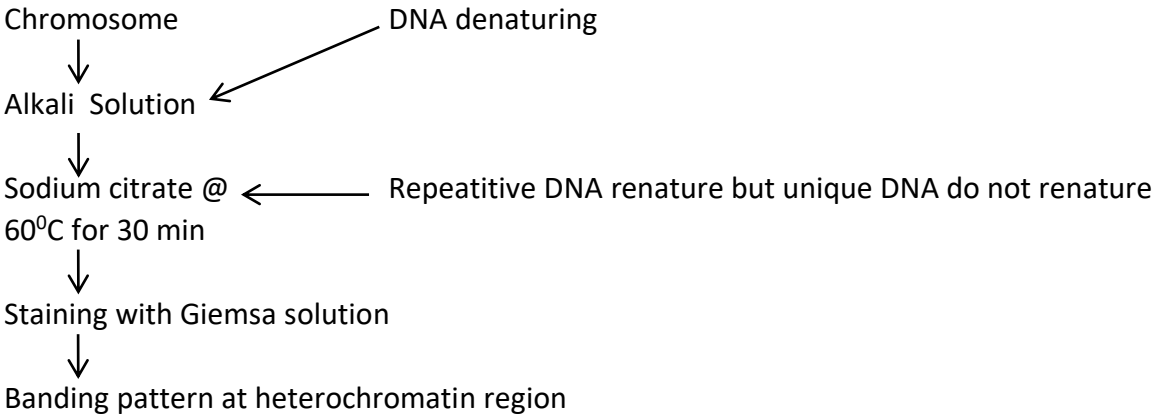
The banding method is a complex technique of denaturation and renaturation of DNA.

DNA denaturation occurs by treatment with acid, alkali or increased temperature. Sub-sequently renaturation of DNA occurs in treatments with sodium citrate at 60^0C.

By these treatment, the repetitive DNA (heterochromatin) denatures but low repetitive and unique DNAs do not re-nature. This results in differential staining of the specific chromosome regions.

Classification of C-bands by Linde-Laursen (1978) in barley chromosome-

1. Centromeric bands
2. Intercalary bands
3. Telomeric bands
4. Bands beside the sec. constriction in the short arm of satellited chromosomes.



Advantages :

1) Identification of chromosomes particulary in insects and plants
2) Identification of centromere position
3) Gene mapping

Disadvantages:

1) C-banding methods do not permit identification of every chromosome in the somatic cell complement

T- banding (Terminal banding)

- It uses high temp, pH 6.7 and giemsa staining
- Most efficient banding pattern for chromosome terminal banding
- Identification of translocation

N- banding techniques

Chromosome
↓
Air dried
↓
Treated with 5% trichloroacetic
acid at @ 95^0c for 30 min
↓
Treated with 0.1 N Hcl at @ 60^0C
for 30 min
↓
Banding pattern in structural non-histone proteins linked to NoR region.

Remember-

1. Q (Quinacrine) was worked out by Caspersson *et al.* in 1958
2. G (Giemsa) was worked out by Summer *et al.* in 1971
3. N (NoR) was worked out by Matsui and Sasaki in 1973
4. C (Centromeric)was worked out by Linde and Laursen in 1978.

N- banding

	Advantages	**Disadvantages**
1.	Used in identification of Ncleoloar organizer region	Time consuming both in technique and reagent preparation
2.	Superior banding pattern	

Applications of chromosome banding:

1. Chromosome identification
2. Chromosome abnormalities
3. Chromosome of cultured cells
4. Chromosome banding and cancer.
5. C-bands used for paternity testing and gene mapping

Disadvantages-

1. The ineffectiveness of determining small translocations
2. Detecting microdeletions
3. Characterizing the chromosome of cell lines which are complex.