MEDICAL ACADEMY NAMED AFTER S.I GEORGIEVSKY VERNADSKY CFU

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TOPIC: CYTOGENETIC METHOD

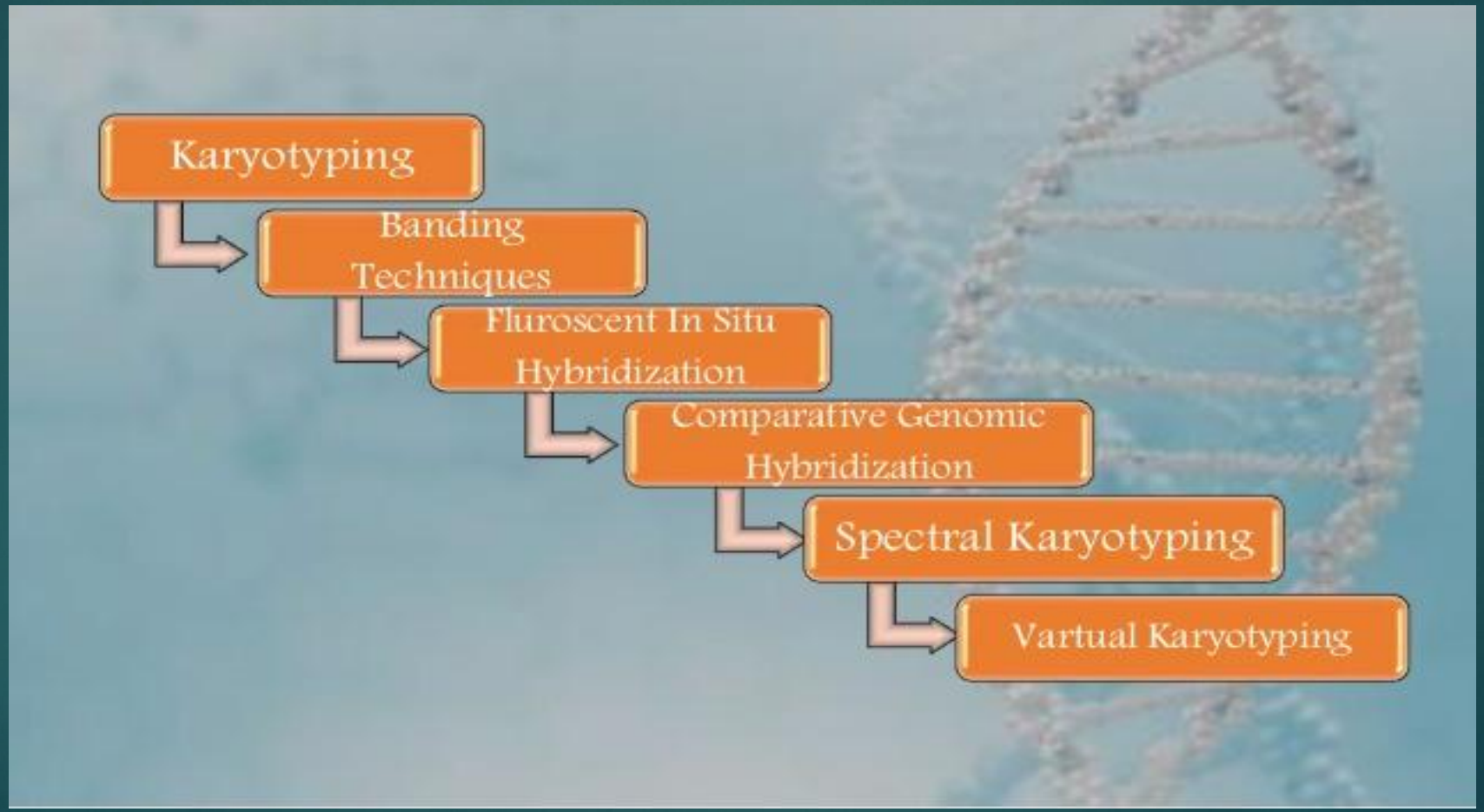
INTRODUCTION

- Cytogenetics involves the examination of chromosomes to identify structural abnormalities present in the chromosome.

Why cytogenetic methods?

- Molecular cytogenetic methods are use to detect chromosomal aberrations.
- There are many chromosomal aberration are observe in the population.

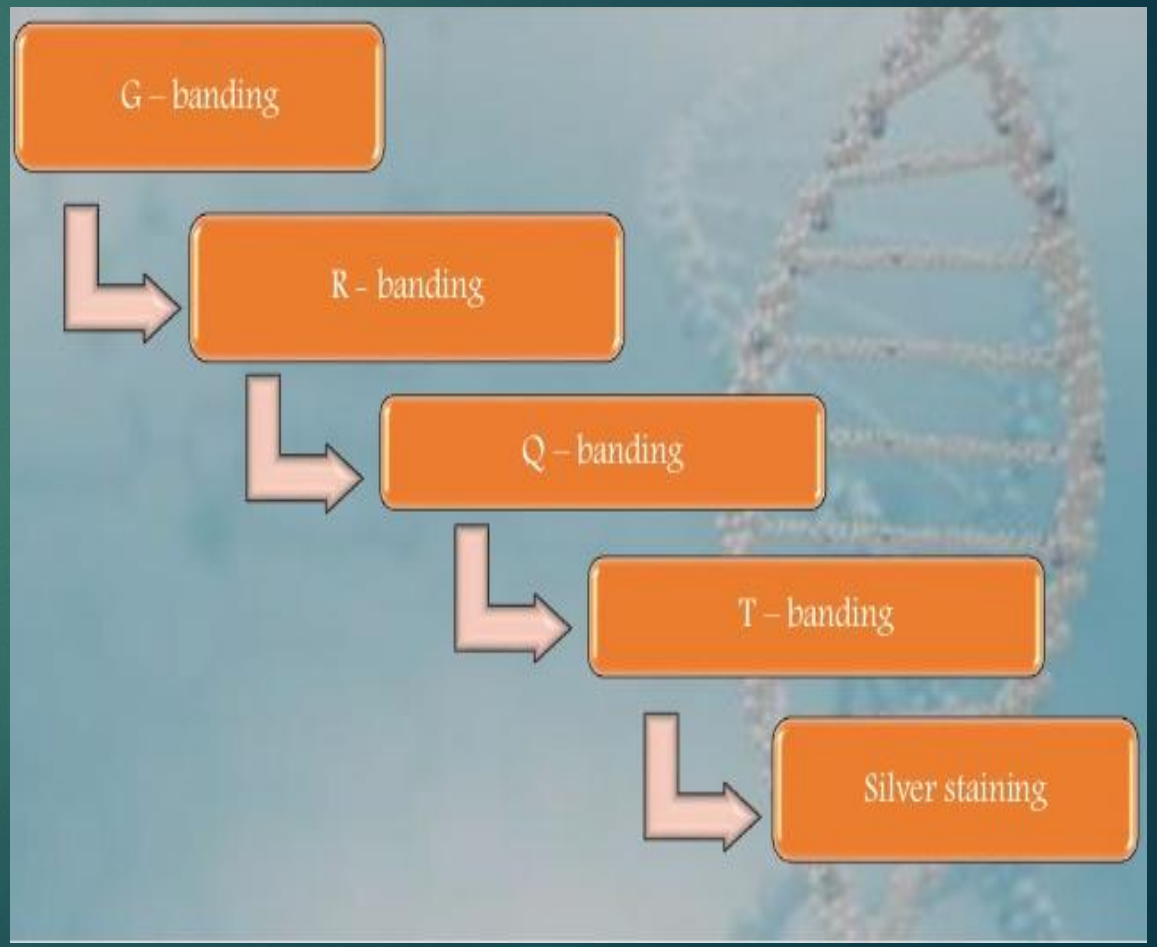
THE MOLECULAR CYTOGENETIC METHODS ARE:-



KARYOTYPING

- Karyotyping is the test to exam chromosome in a sample of cell. This test can identify genetic problems as the cause of a disorder or disease.
- This method produce a characteristic pattern of contrasting dark and light transverse bands on the chromosomes.
- Banding makes it possible to identify homologous chromosome by visualization of chromosomes.
- Banding of homologous chromosome allows chromosome segments and arrangements to be identified.
- It helps to detect the chromosomal abnormalities and chromosomal alterations.
- The most widely used banding methods are G-banding(Giemsa-banding) and R-banding(reverse-banding).


Cell fixed in metaphase
Chromosomes stained
Picture taken
Enlarged
Cut out
Matched by banding patterns
Arrange them in order.


G – banding
R - banding
Q – banding
T – banding
Silver staining

FLUROSCENT IN SITU HYBRIDIZATION (FISH)

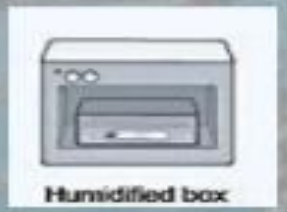
It is a cytogenetic technique that uses fluorescent probes that bind to only those parts of the chromosome with high degree of sequence complementarity.

It is used to detect and localize the presence or absence of specific DNA sequence on chromosome

FISH is often used for finding specific features in DNA for use in genetics counselling, medicine, and species identification.

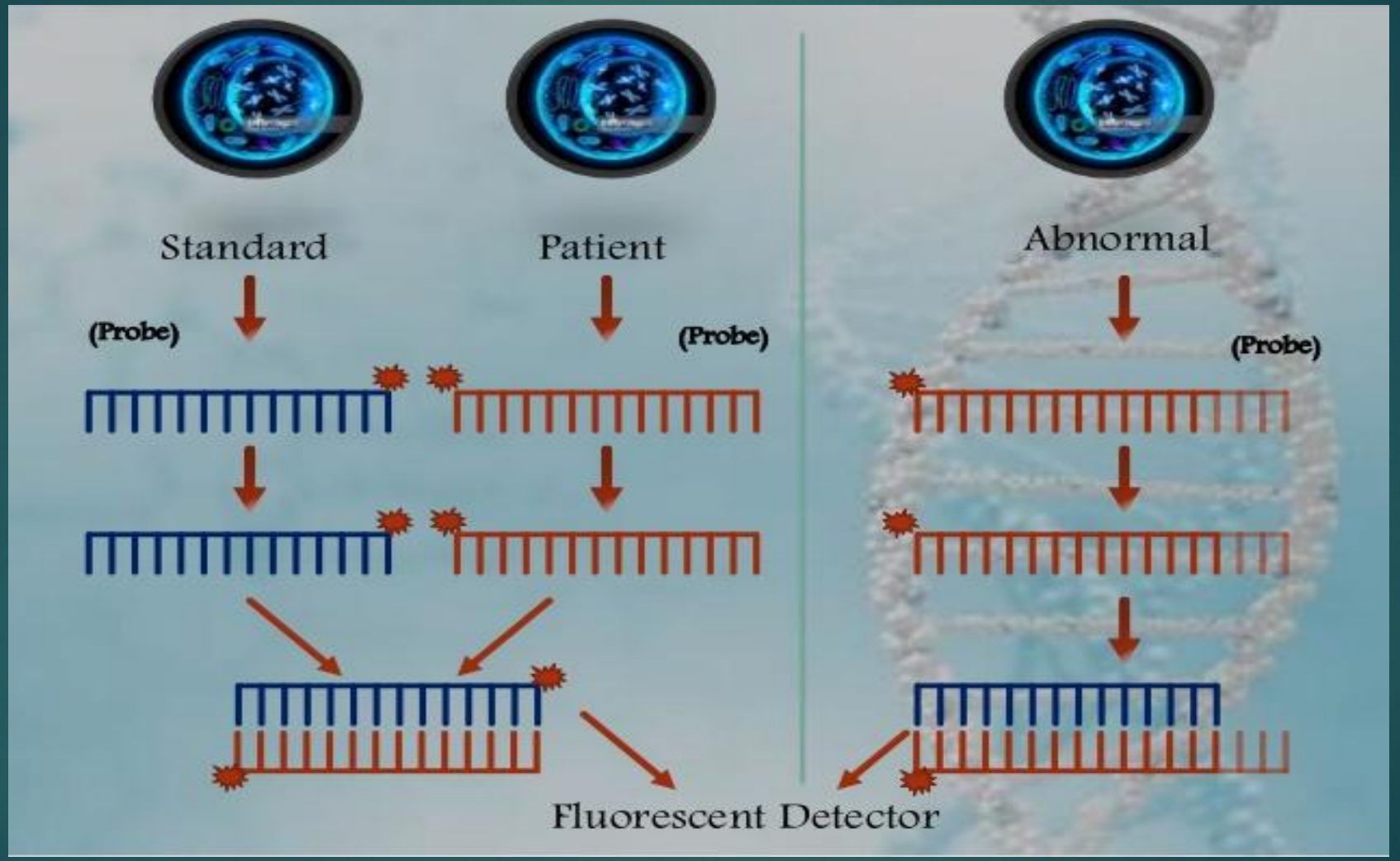
Fluroscent In Situ Hybridization

- **Step I – Denaturation**
  - ➢ Conversion of double stranded dna in to single stranded dna
- **Step II – Hybridization**
  - ➢ Application of probe DNA to slide & overnight incubation at 37°C
  - ➢ Binding of probe DNA to trarget DNA.
- **Step III – Post hybridisation washing & detection**
  - ➢ Washing of unbound probe DNA.
  - ➢ Application of counter stain &
- **Step IV – counter stain**
  - ➢ Application of counter stain.
- **Step V – Visualization**
  - ➢ visualization using fluorescence microscopy.



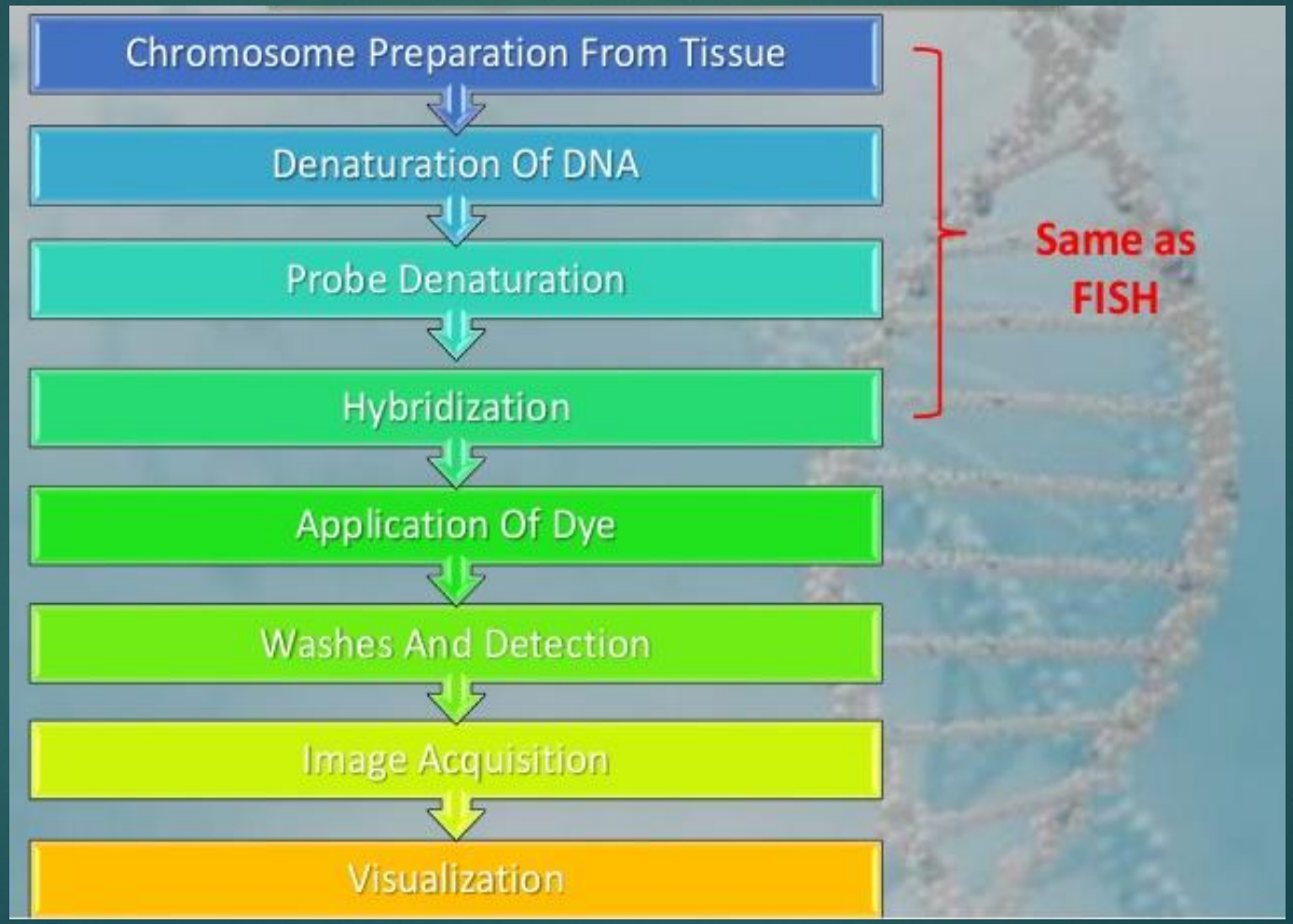
COMPARATIVE GENOMIC HYBRIDIZTION (CGH)

- Comparative genomic hybridization is a molecular cytogenetic method for analysing copy number variations with the help of hybridization technique.
- It helps in detection of balanced rearrangements of chromosomes and for the comparison of normal and suspected DNA samples.

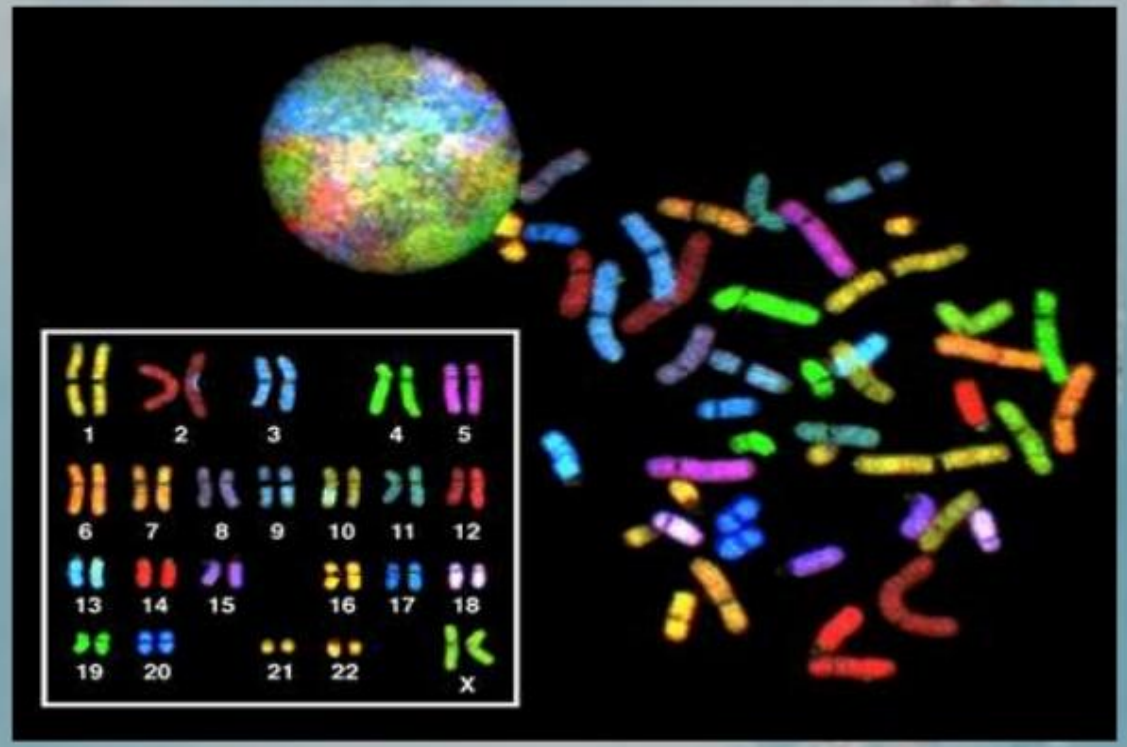

Standard
Patient
Abnormal
(Probe)
(Probe)
(Probe)
Fluorescent Detector

SPECTRAL KARYOTYPING(SKY)

- Spectral karyotyping is cytogenetical techniques used to simultaneously visualize all the pairs of chromosome in an organism in different colours.
- SKY allow determination of the precise molecular address of chromosome breakpoints associated with the deletion translocation and insertion.
- SKY can discern the aberrations that cannot be detected very well by convectional banding techniques and fluorescent in situ hybridization(FISH).

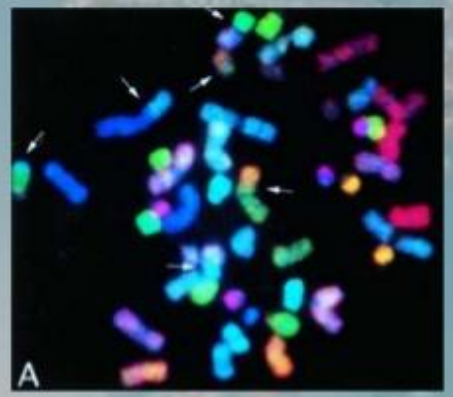

Chromosome Preparation From Tissue
Denaturation Of DNA
Probe Denaturation
Hybridization
Application Of Dye
Washes And Detection
Image Acquisition
Visualization
Same as FISH

Spectral Karyotype Observation



Some Abnormalities Seen In SKY

Translocation



CONCLUSION

- Cytogenetic techniques like FISH,CGH and SKY are the available advanced diagnostic tools to detect such chromosomal abnormalities and to prevent the spread in the population.


THANK
YOU