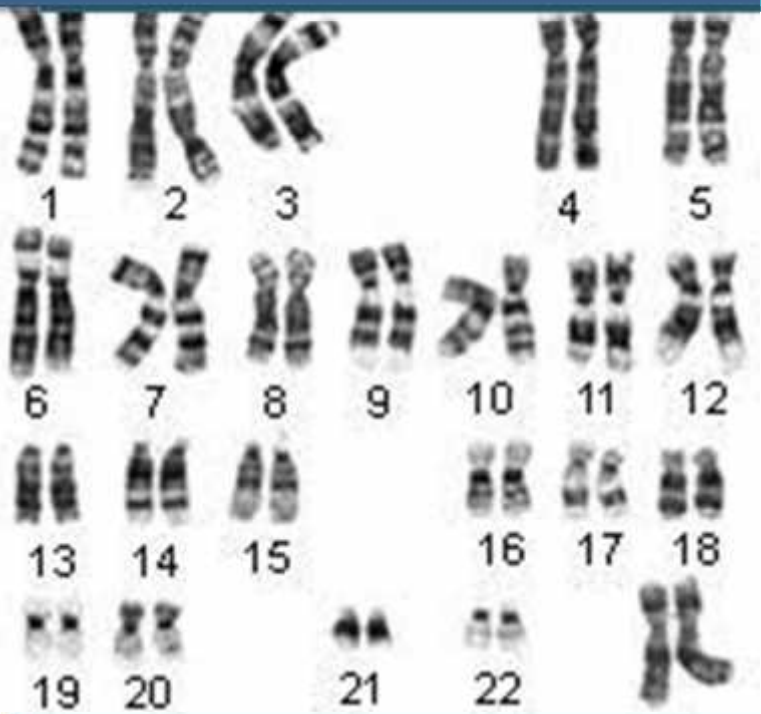
MUCOM
2017 - 2018

Human Karyotyping Lab



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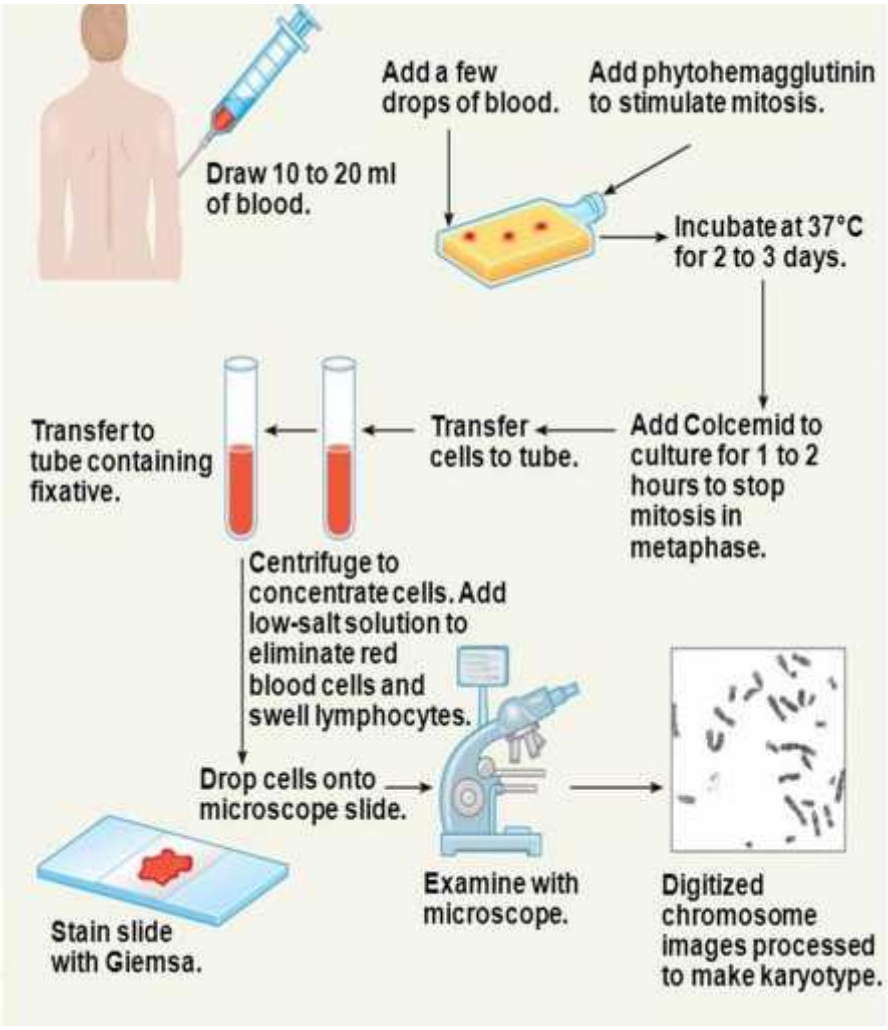
HUMAN KARYOTYPING

- **Karyotype:** an ordered display of the 23 pairs of human chromosomes in a typical somatic cell.
- **Karyotyping:** is a test to examine chromosomes in a sample of cells. This test can help identify genetic problems as the cause of a disorder or disease.

In order to analyze chromosomes, the sample must contain cells that are actively dividing (in metaphase in mitosis)

Procedure and principle of G-band karyotype

1. **Collect** the sample (blood, bone marrow, amniotic fluid or tissue from the placenta).
2. **Incubate** drops of blood sample with special media contain a **mitogene** that stimulate cell division such as PHA or granulocyte colony stimulating factor (G-CSF).
3. **Stopping the cell division** at metaphase (when they are maximally condensed) by add **Colcemid** that attach to mitotic spindle and prevent the cell enter to anaphase.
4. **Hypotonic treatment** of the cells by low concentrated **KCl**, the KCl swells the cell but ovoid excess exposure as it may cause rupture of the cells.
5. **Fixing** the cells by fixative solution.
6. **Making the chromosome slides** by dropping a solution from a height of about half meter (18 inches), drop two or three drops of fluid onto each side.(in this step the cell will burst).
7. Allow the slides to **dry** then add **trypsin**
8. **Stain** the slide by immersion in fresh **Giemsa** stain.
9. Finally **exam** the slide to microphotograph good spreads
10. Construction of G-banded karyotype.



❖ How read the chromosomes slide

- Chromosomes are digitally arranged so that they are matched with their **homologue** or "partner" chromosome.
- They are numbered according to size, location of centromere and arrangement of bands.

Common Symbols Used in Karyotype Nomenclature

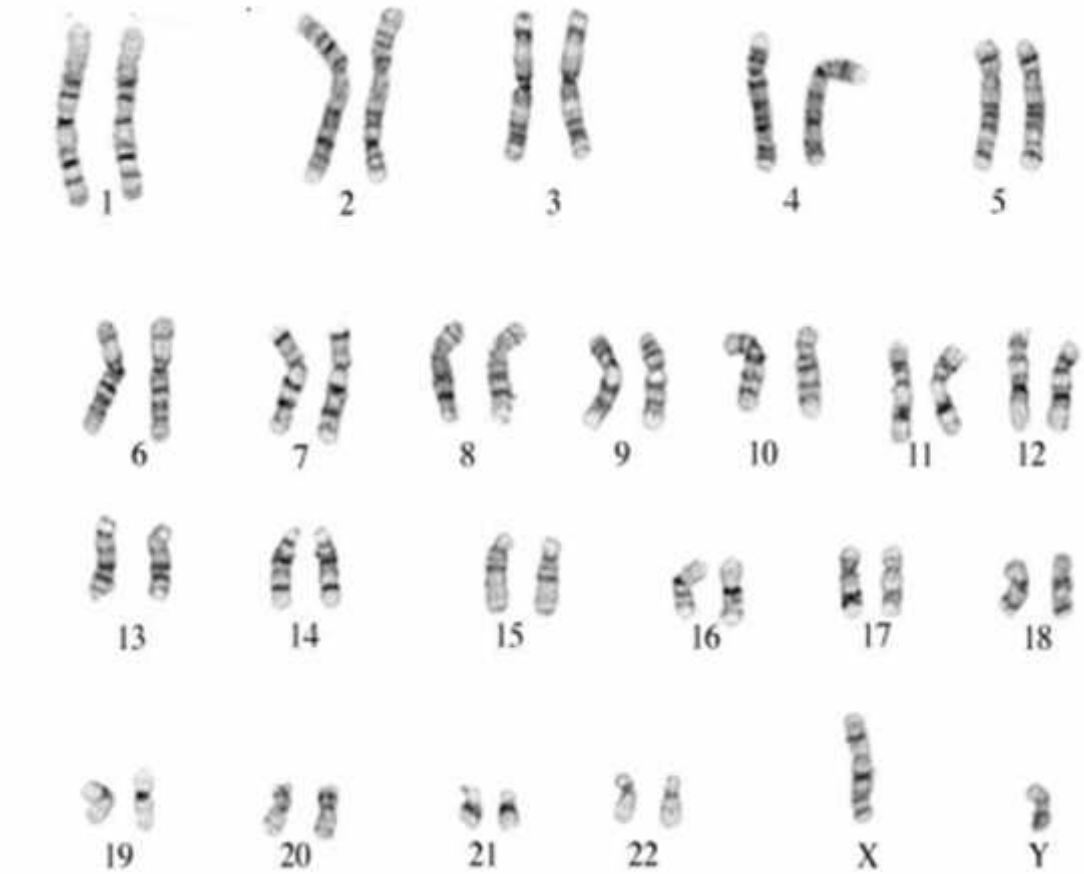
1-22	Autosome number
X, Y	Sex chromosomes
(+) or (–)	When placed before an autosomal number, indicates that chromosome is extra or missing
p	Short arm of the chromosome
q	Long arm of the chromosome
t	Translocation
del	Deletion
inv	Inversion
i	Isochromosome
r	Ring chromosome

❖ Test Applications

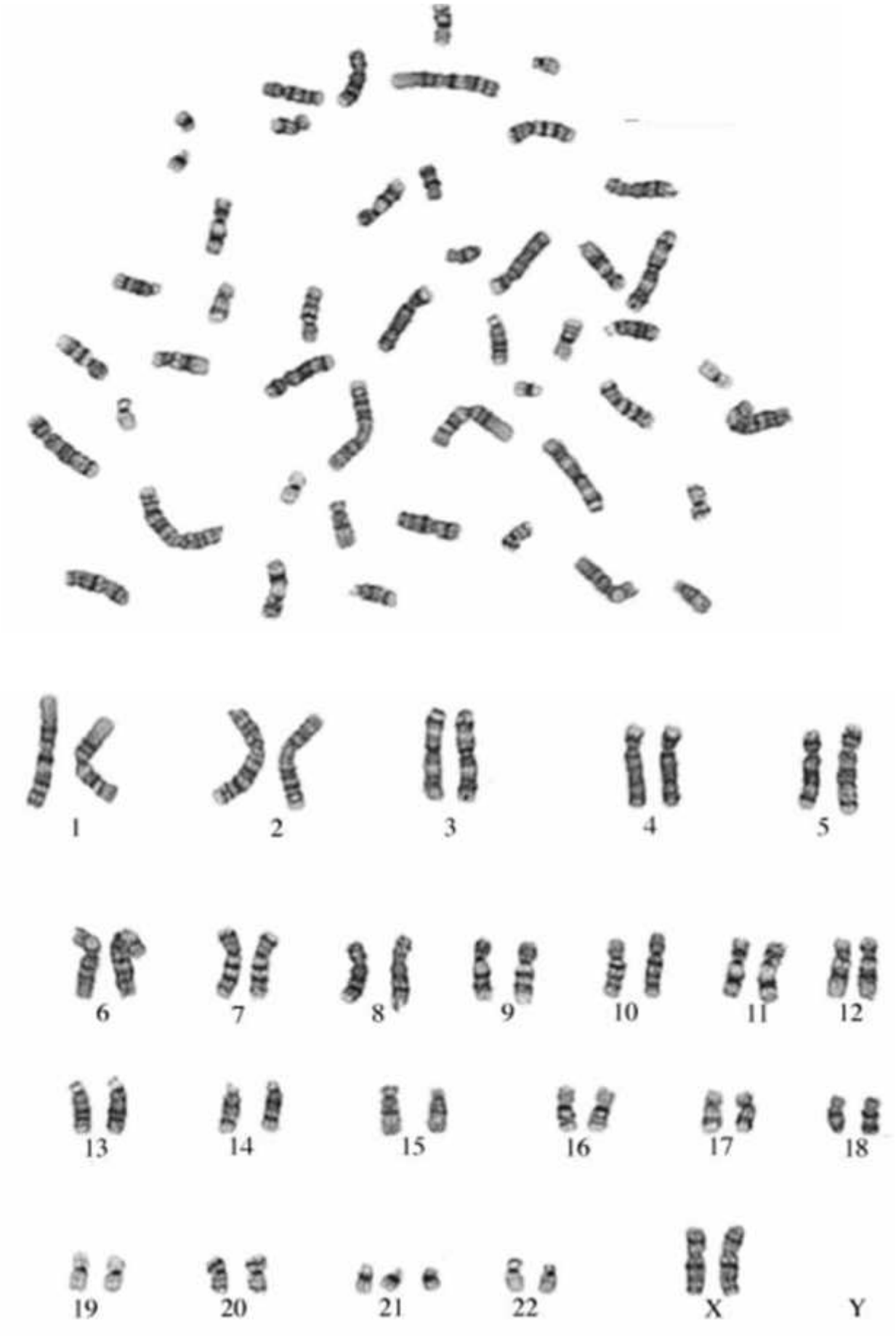
Karyotyping is widely used to detect chromosomal abnormalities; wither numerical or structural abnormalities.

- **Numerical chromosomal abnormalities.**
  A. **Triploidy** (three copies of each chromosome) or **Tetraploidy** (four copies)
  B. **Aneuploidy**, a deviation from the euploid number, represents the gain (+) or loss (–) of a specific chromosome. Two major forms of aneuploidy are observed:
     1. **Monosomy** (loss of a chromosome), the most common example is
        - Turner syndrome (monosomy X). 45 X or 45,XO
     2. **Trisomy** (gain of a chromosome), the most common examples are
        - Down syndrome (trisomy 21), 47,XY,+21 or 47,XX,+21
        - Edward syndrome (trisomy 18), 47,XY,+18 or 47,XX,+18
        - Patau syndrome (trisomy 13), 47,XY,+13 or 47,XX,+13
        - Klinefelter syndrome, 47,XXY
        - Trisomy X, 47 XXX
- **Structural chromosomal abnormalities.**
  1. Translocations, example
     - 46,XX,–14,+t(14;21), or 46,XY,–14,+t(14;21) (Down syndrome)
     - 46,XX, t(9;22) or 46,XX, t(9;22) Philadelphia chromosome, in Chronic myelogenous leukemia
  2. Deletion, example
     - 46,XX, del(5p) or 46,XY, del(5p), Cri-du-chat syndrome.
  3. Inversion
  4. Isochromosome
  5. Ring chromosome

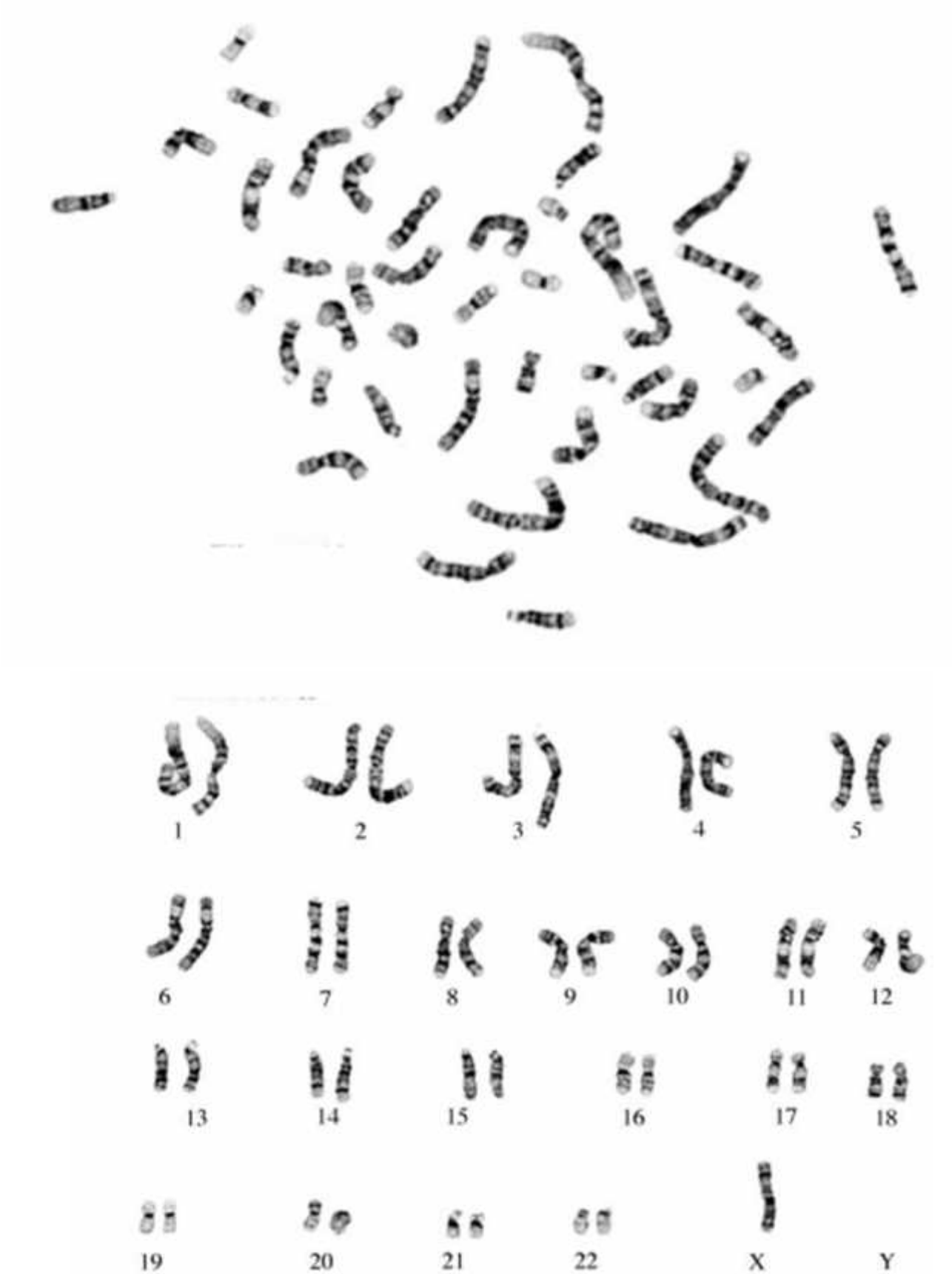
46,XY Normal male



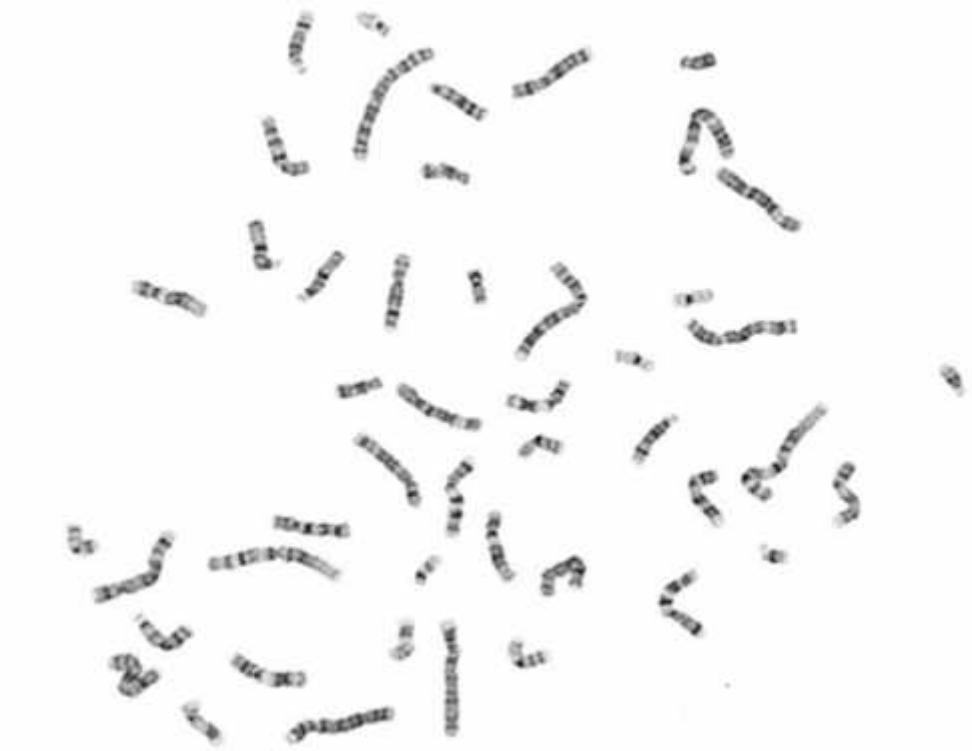
47, XX, +21 (Trisomy21), a female with Down syndrome

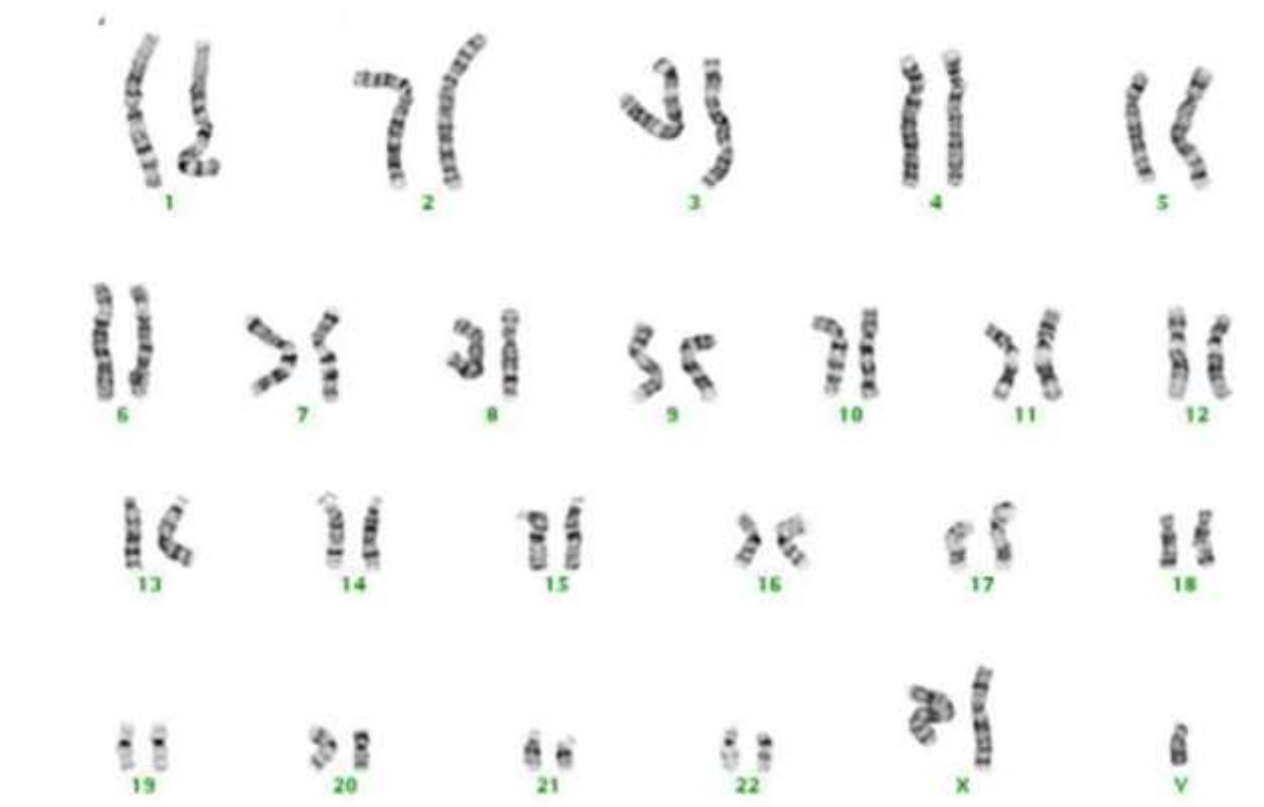


45, XO (Monosomy X) a female with Turner's syndrome



47, XXY a male with Klinefelter syndrome





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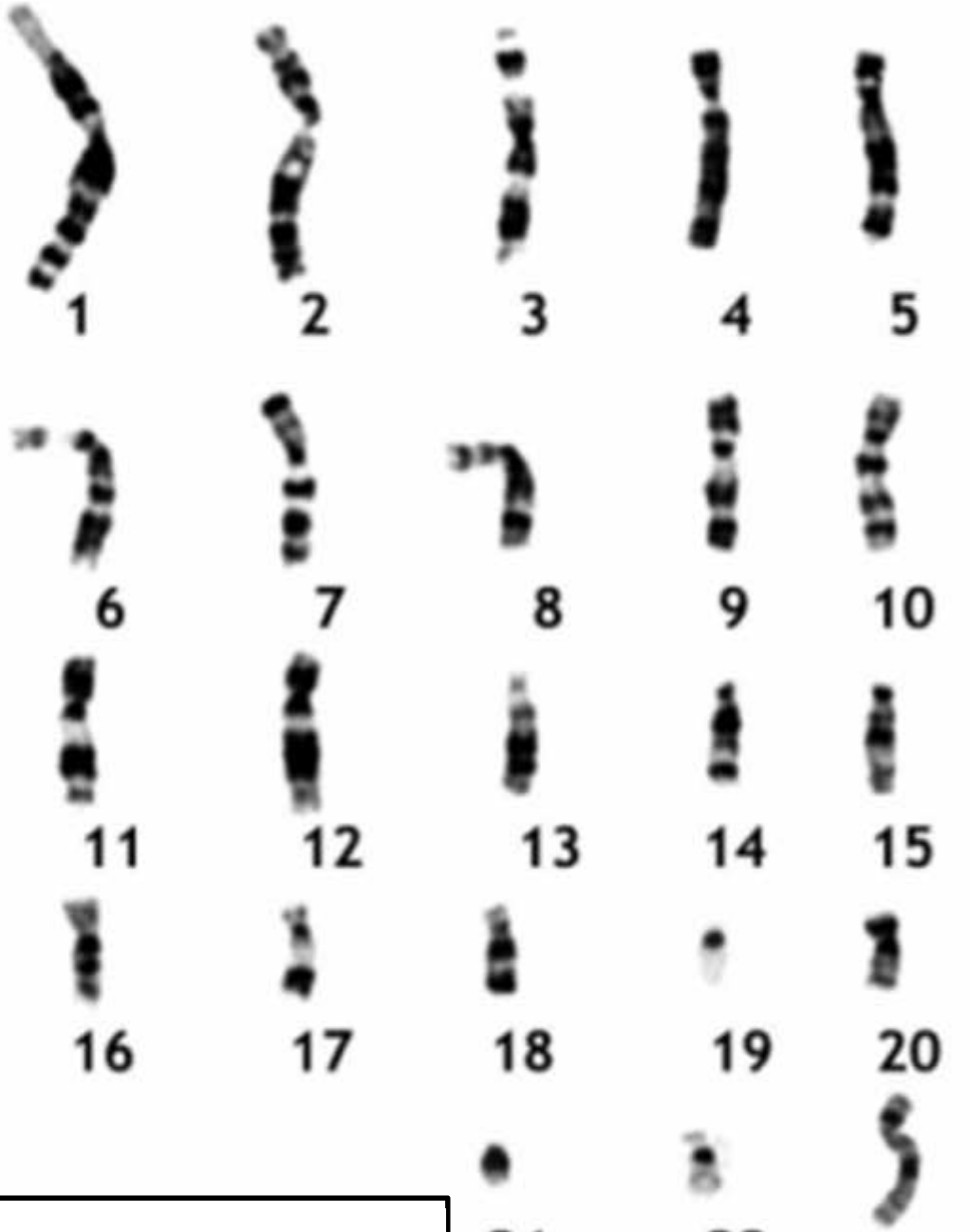

Cut & paste karyotyping activity

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