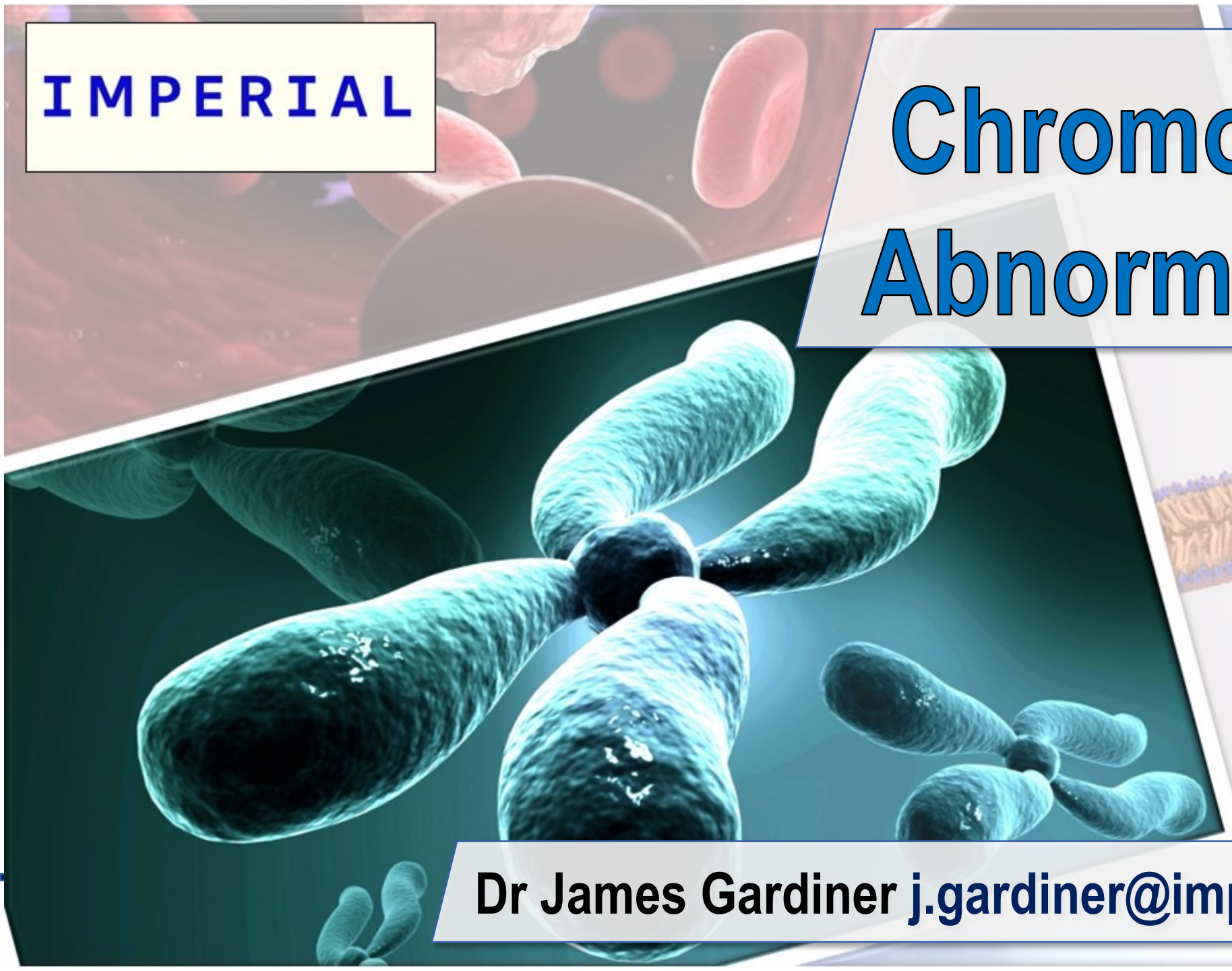
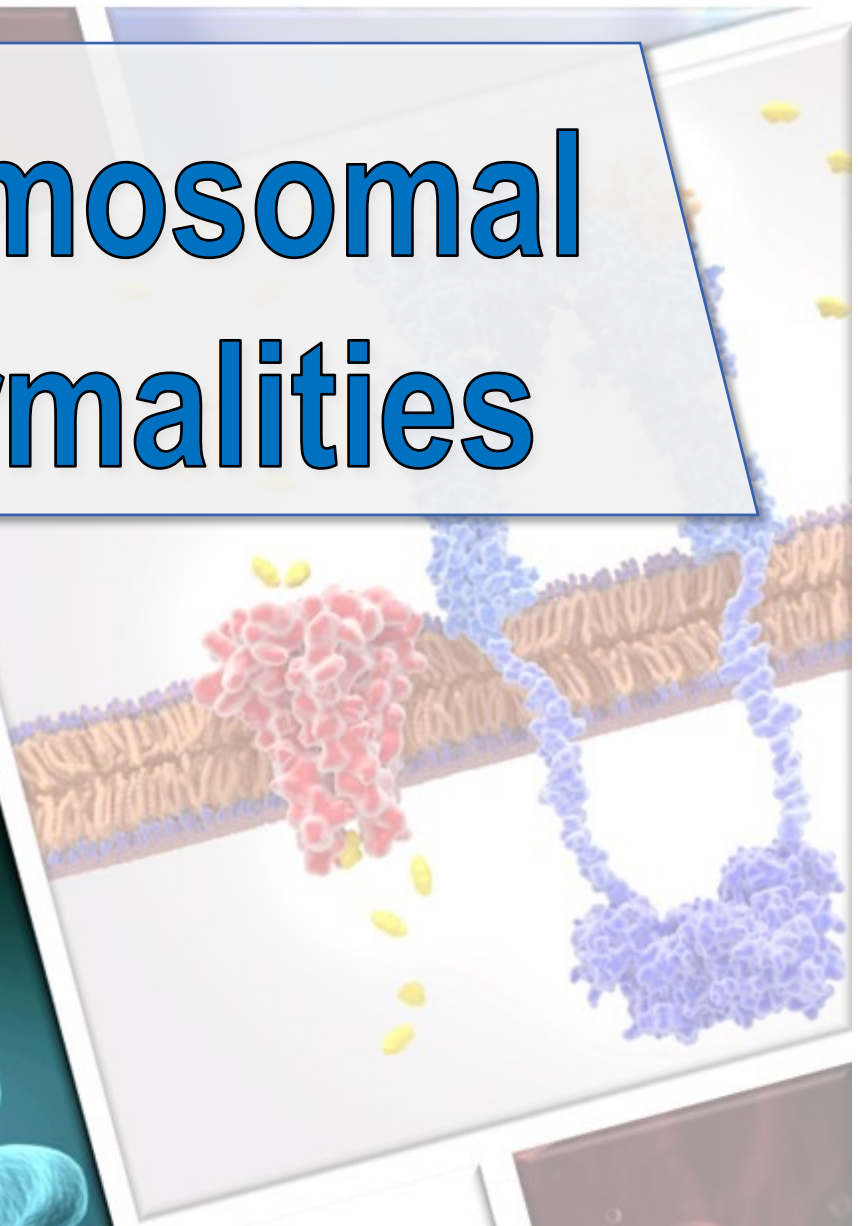
IMPERIAL

Chromosomal Abnormalities





Dr James Gardiner j.gardiner@imperial.ac.uk

Session Plan

Part 1

- Understand human karyotype
- Understand chromosome banding
- Understand chromosomal nomenclature

Part 2

- Aneuploidy
- Explain the increased genetic risks associated with advanced maternal and paternal age
- Explain the basis of chromosomal non-disjunction
- Understand the relevance to clinical disease

Part 3

- Overview of other types of chromosomal abnormalities
- Understand the basis of other chromosomal changes
- Understand their relevance to clinical disease

1-POM-4-3: Chromosomal abnormalities: Recall the normal human karyotype, chromosome banding and nomenclature, explain the basis of nondisjunction and other types of chromosomal abnormalities, and their relevance to disease.

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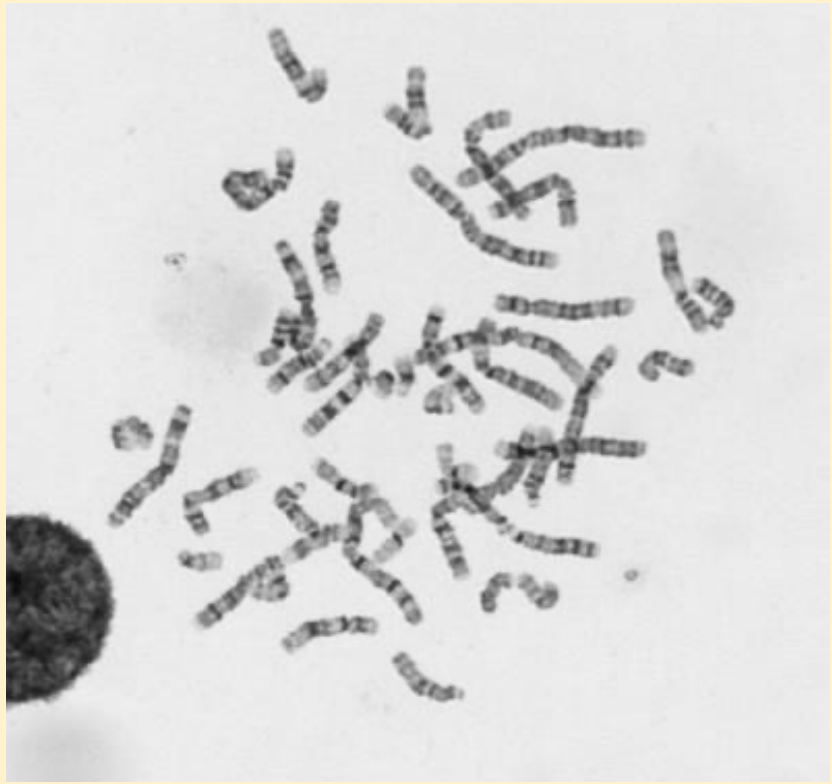
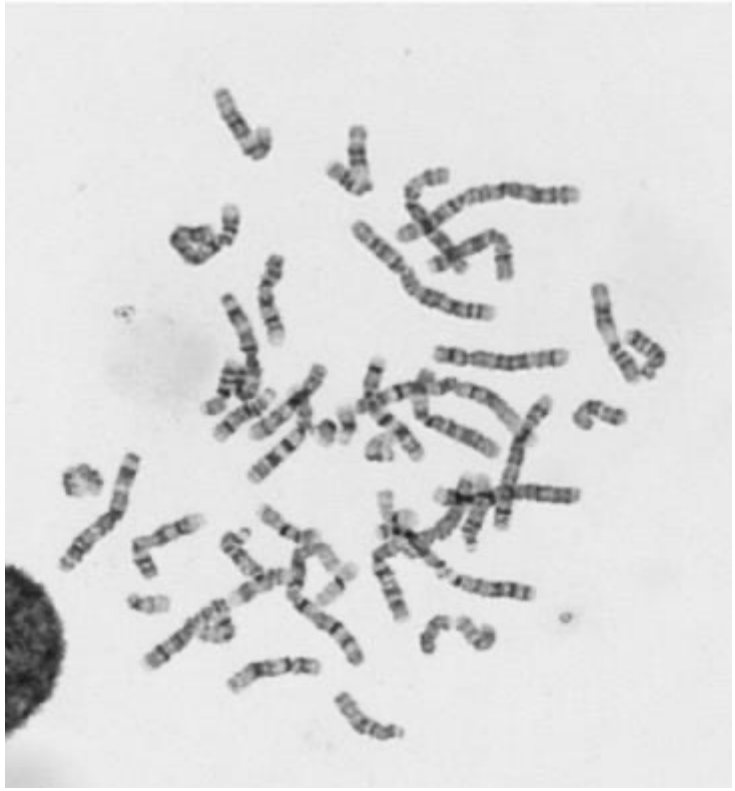
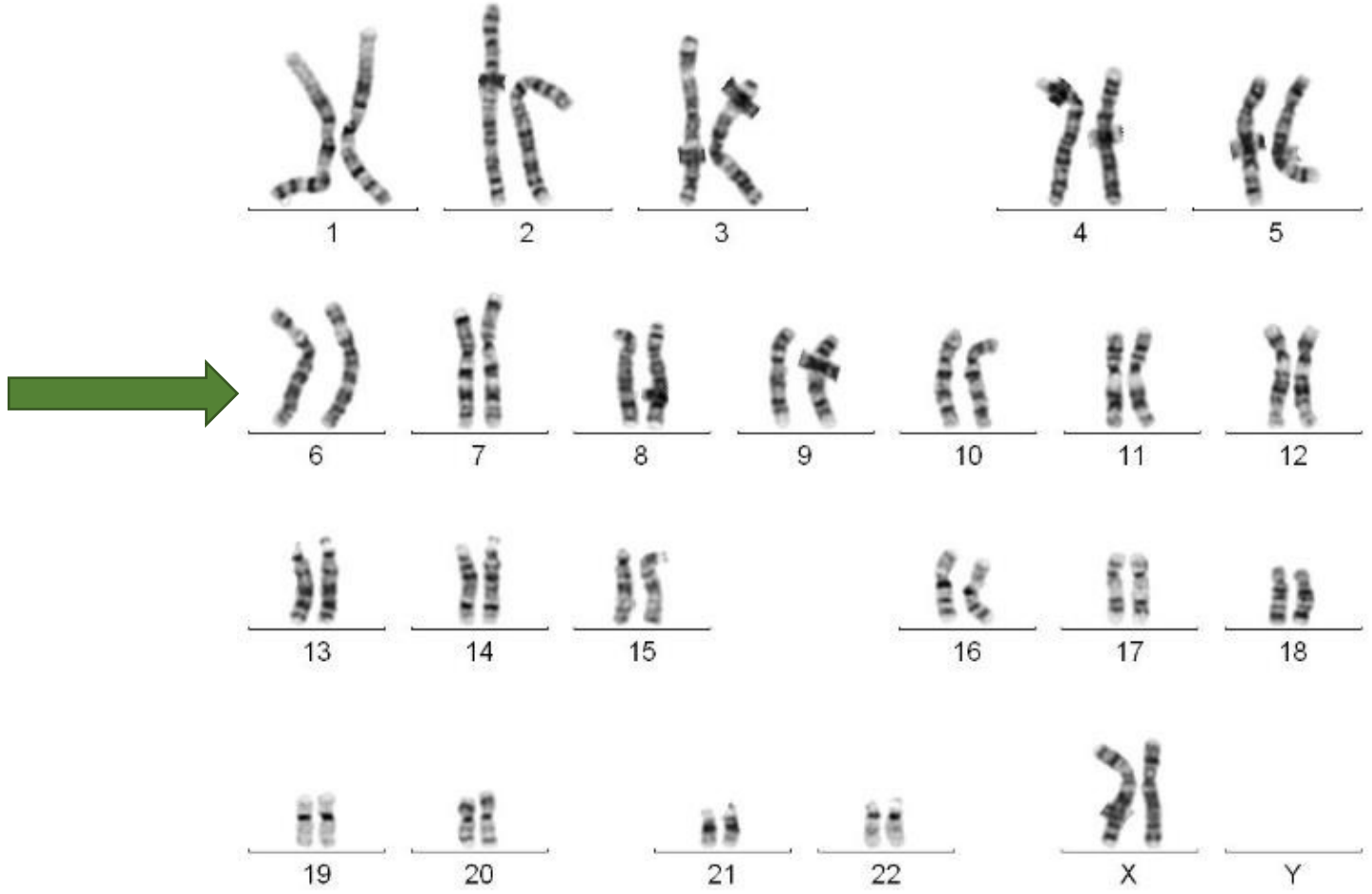
Prepare karyotype

1. Collect ~5ml heparinised venous blood
   - Can use amniotic cells, CVS
2. Isolate white cells
3. Culture in presence of phytohaemagglutinin
   - Stimulates T-lymphocyte growth/differentiation
4. After 48 hours add colchicine
   - Causes mitotic arrest – metaphase
5. Place in hypotonic saline
6. Place on slide
7. Fix and stain



Karyotype Gisema stain

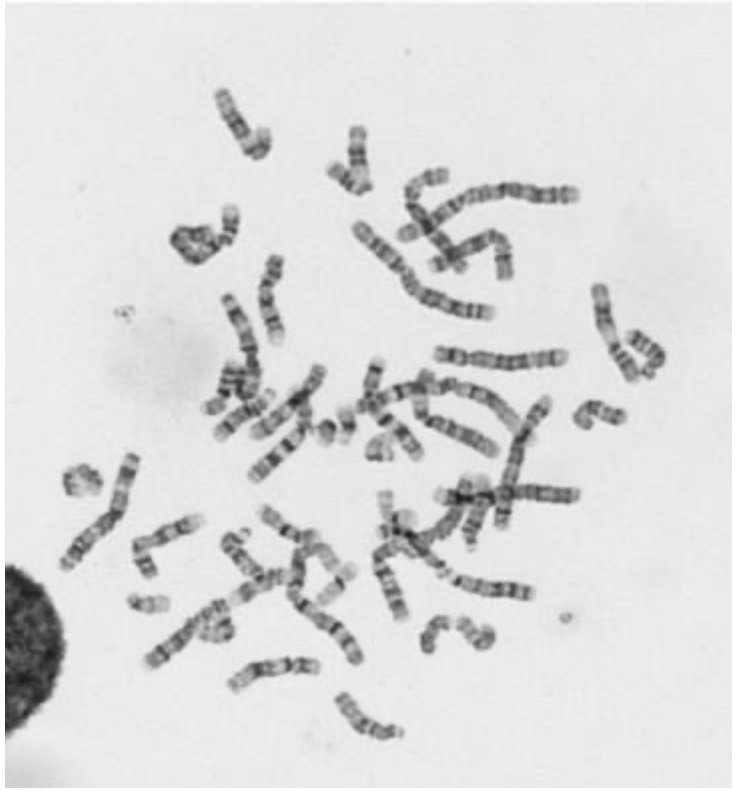


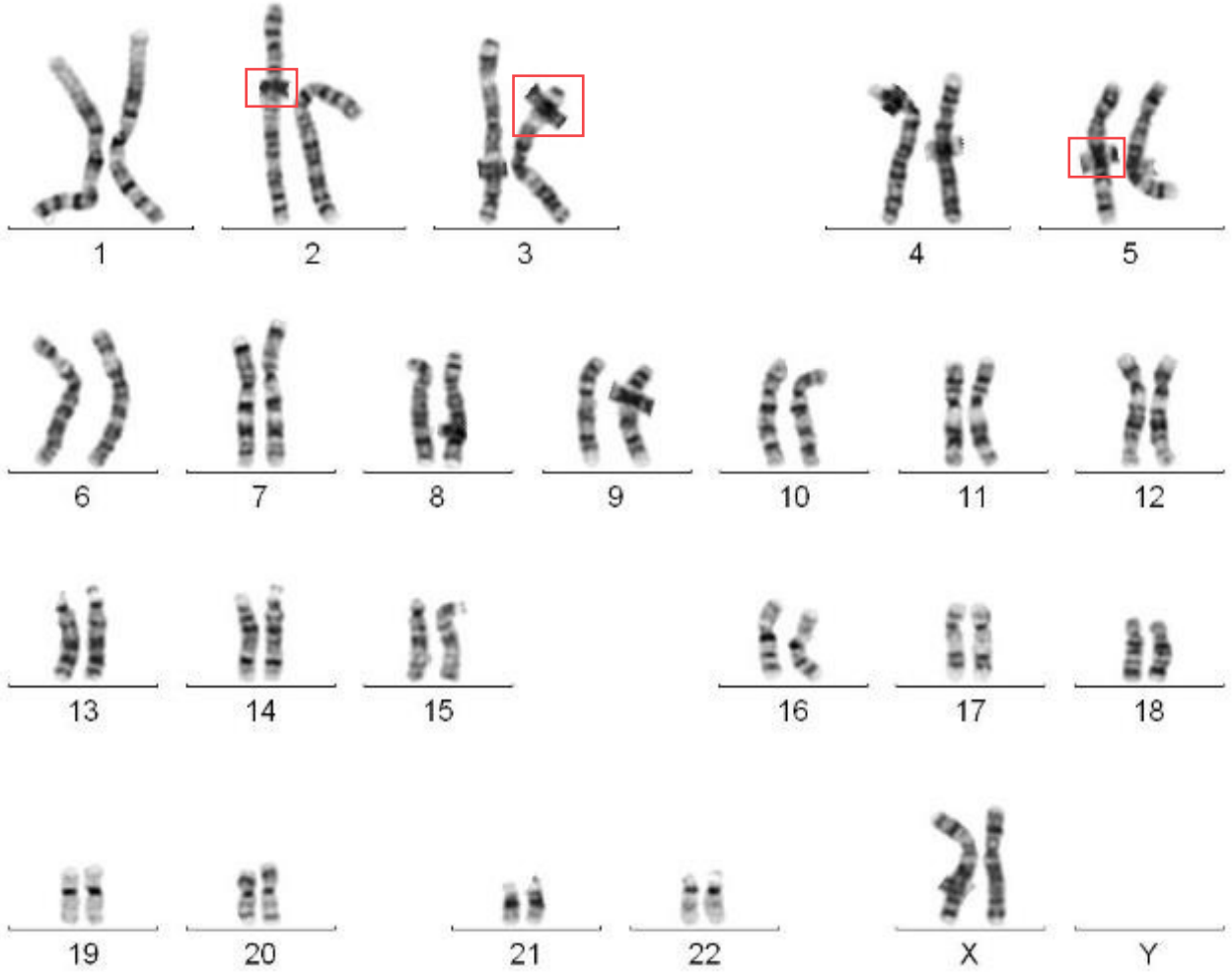


46, XX

1-POM-4.3 Recall the normal human karyotype

Karyotype gisema stain

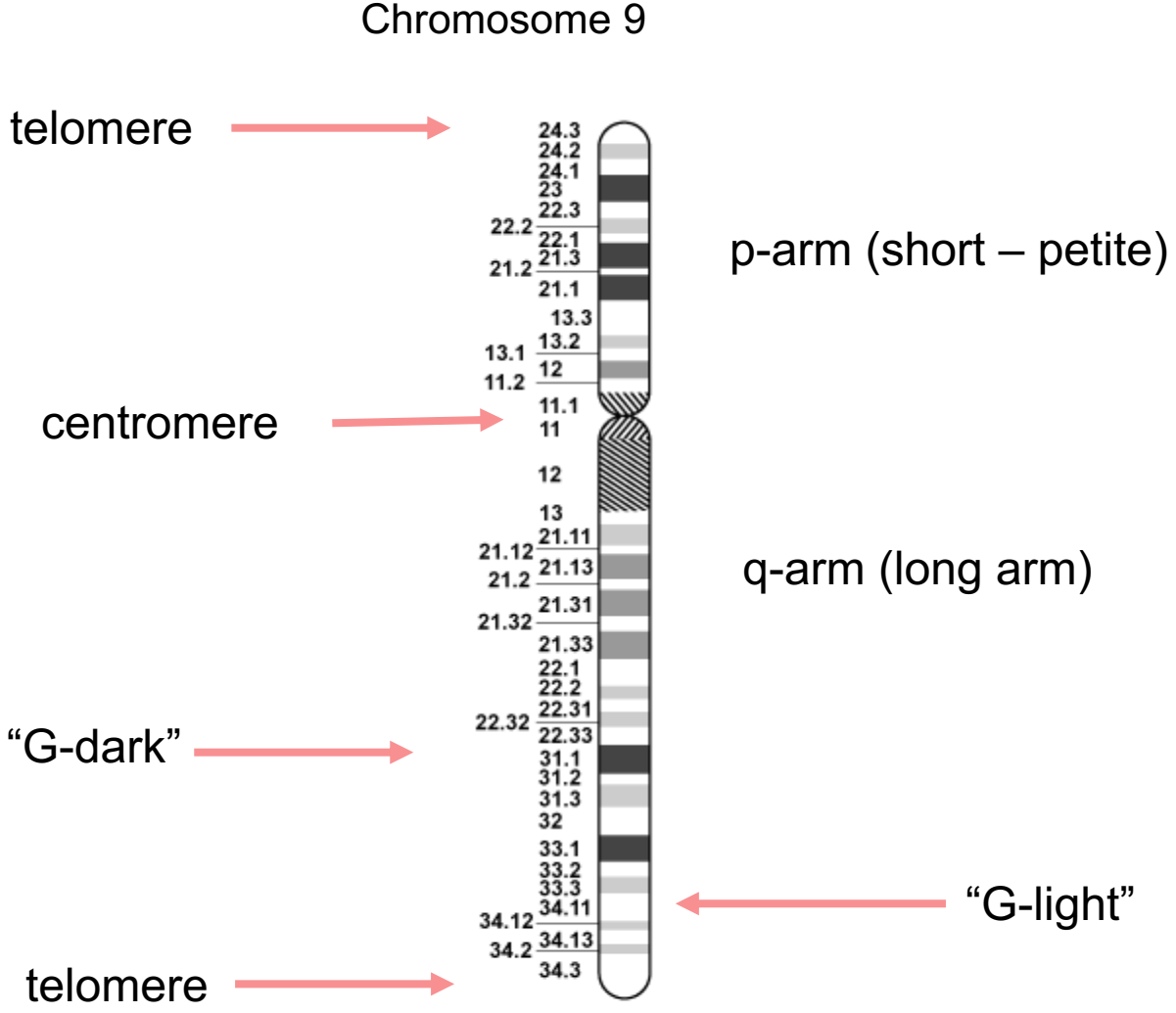




46, XX

1-POM-4.3 Recall the normal human karyotype

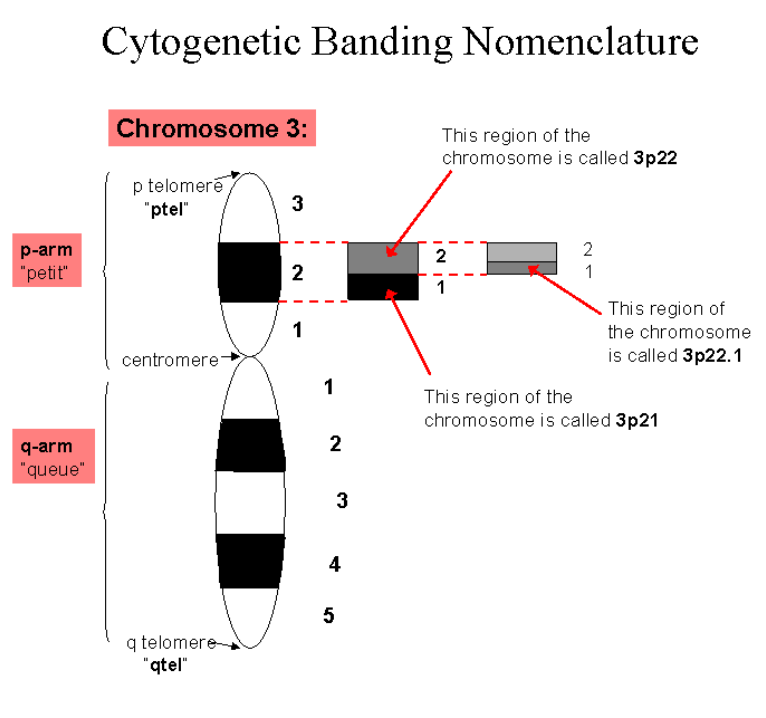
G-banded architecture



Ideogram

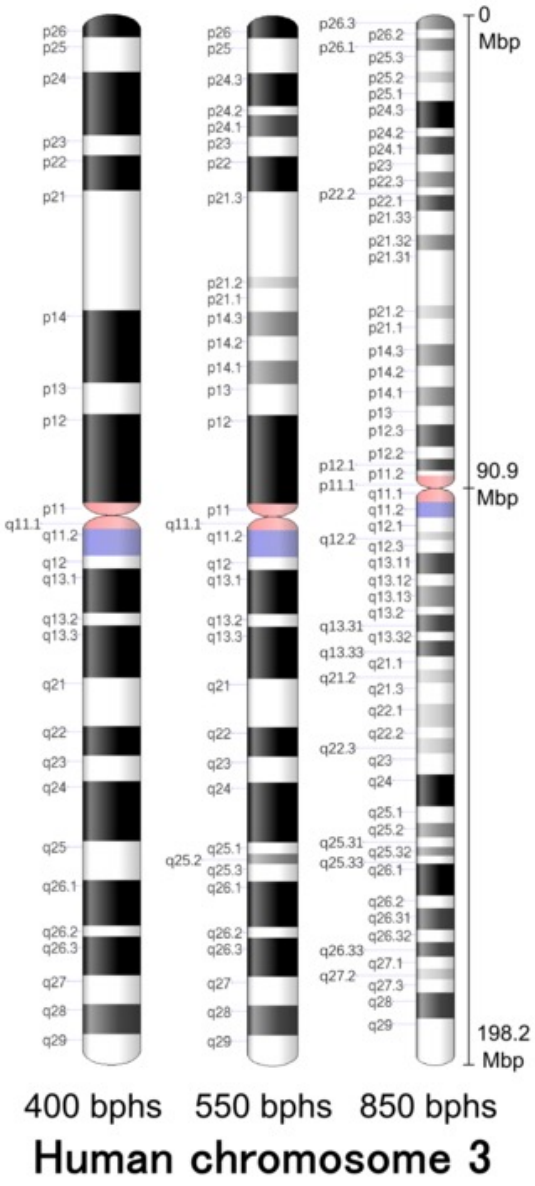
- Chromosomes have some common structural features
- **Giemsa staining** leaves a recognizable pattern of bands

What do the band numbers mean



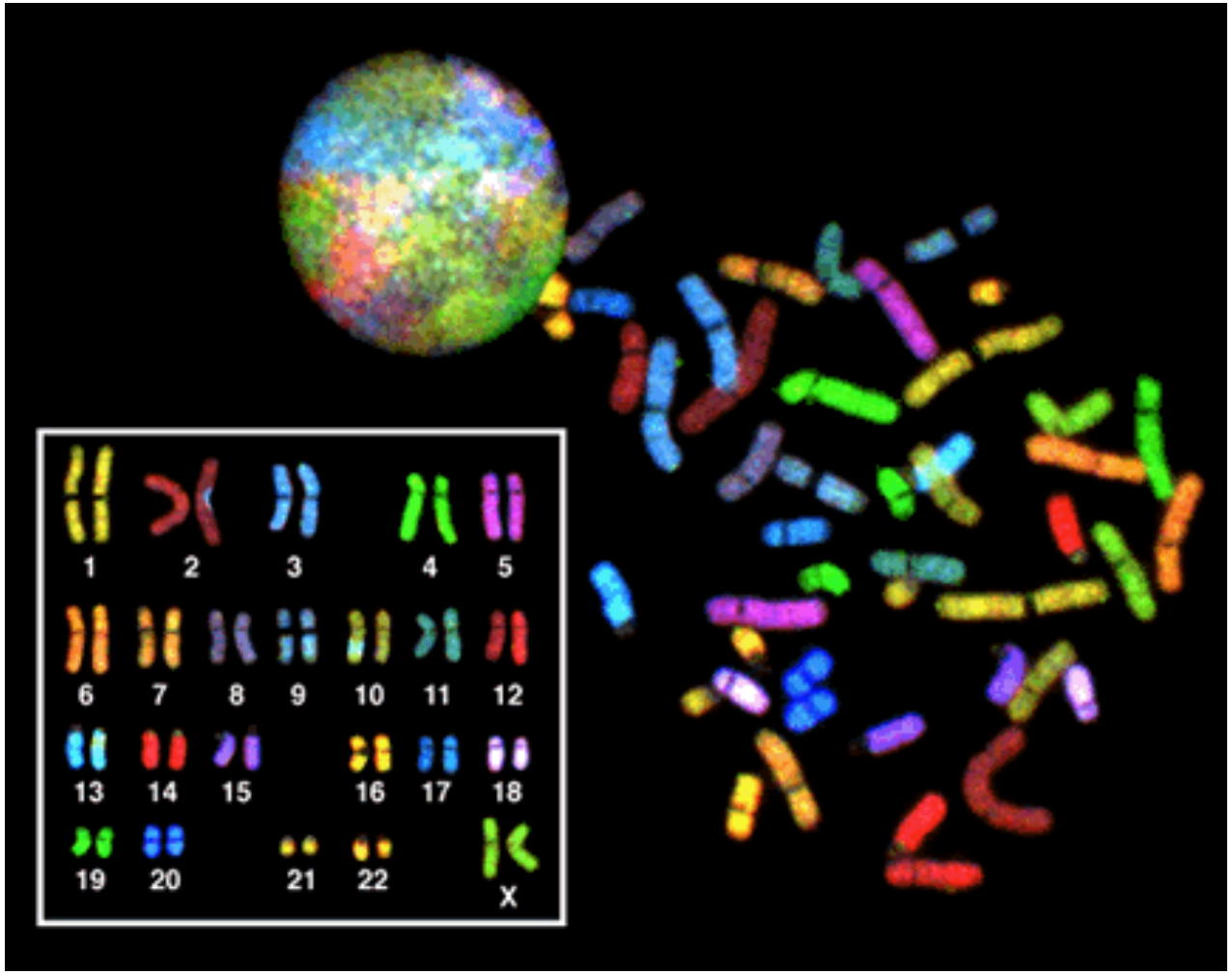
- Bands caused by differently staining
- Bands originally identified with low level of resolution
  - Only a few band visible per chromosome eg 1,2,3
- Improved technology more bands visible
  - Named as sub-bands eg 11,12,21,22,23 etc
  - Further improvements sub-sub-bands 22.1,22.2 etc

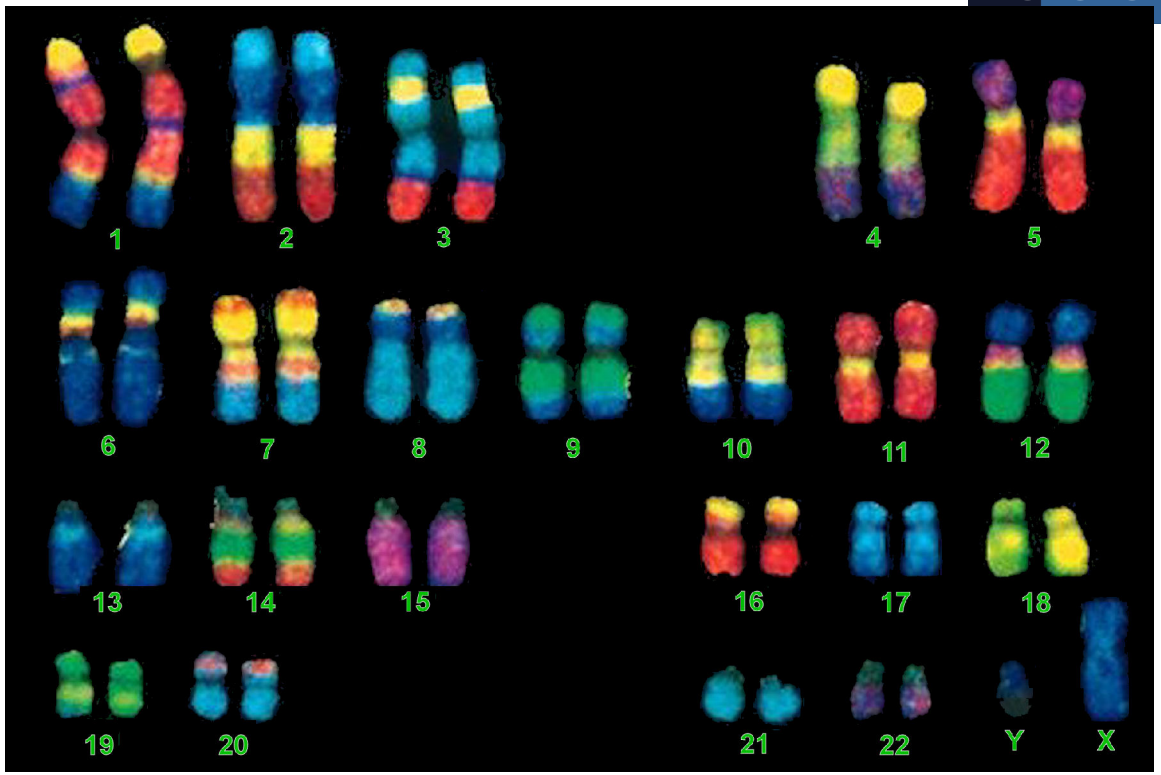
- Improved resolution helps identify smaller aberrations



- Current level of detail chromosome 3
  - bphs – bands per haploid set
- Bands do not represent genes or families of genes
- Regions of different compaction
  - Dark (heterochromatin) more compact fewer genes
  - Light (euchromatin) more open more genes
  - Now often done prophase rather than metaphase

Karyotype fluorescent stain





1-POM-4.3 Recall the normal human karyotype

Whole genome sequencing

We are beginning to see genomic medicine

Human genome project

£5 billion, 11 years

Whole genome sequencing

~£600 1 day (analysis takes longer- AI)

NHS 100000 genomes – 2013-2018

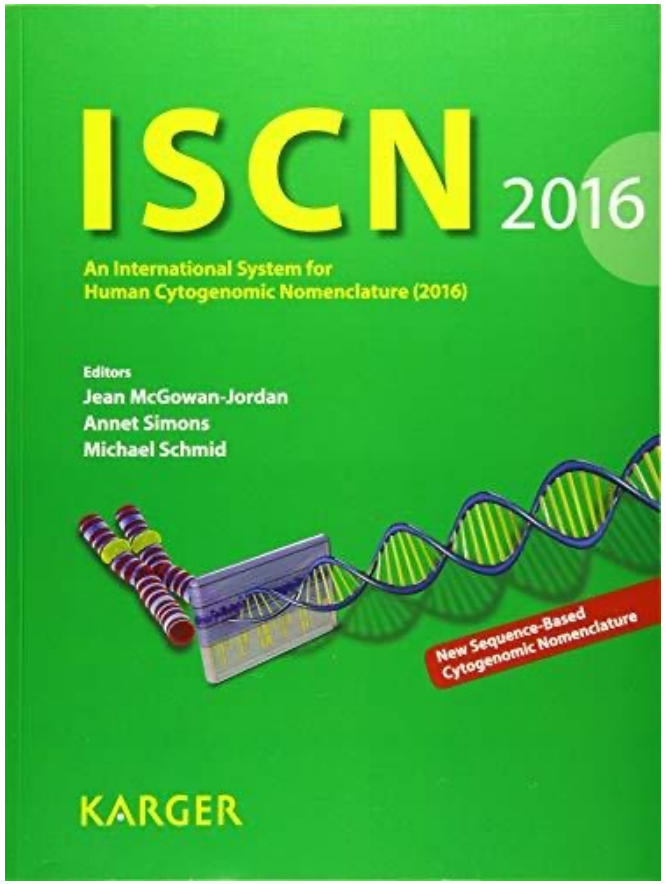
NHS 500000 genomes 2019-23/24 (5mil 2026)

WGS for patients some types of cancer (all patients)

WGS all children with suspected abnormality – September 22

Study to investigate feasibility of WGS all newborns

Standard Nomenclature



abbreviation	Meaning
p	Short arm
q	Long arm
pter	Tip of short arm
qter	Tip of long arm
cen	centromer
del	deletion
der	Derivative chromosome (contains extra material)
dup	duplication
ins	insertion
inv	inversion
t	translocation
+/− Before chromosome number	Gain/loss whole chromosome
+/− After chromosome number	Gain/loss part of chromosome

Progress check

Part 1

- Karyotype – chromosome count of individual
- Performed on metaphase chromosomes
- Various staining methods classically Gisema
- Gisema stain basis of nomenclature
- Can detect major chromosome abnormalities
- Whole genome sequencing now being used

Part 2

Part 3

1-POM-4-3: Chromosomal abnormalities: Recall the normal human karyotype, chromosome banding and nomenclature, explain the basis of nondisjunction and other types of chromosomal abnormalities, and their relevance to disease.

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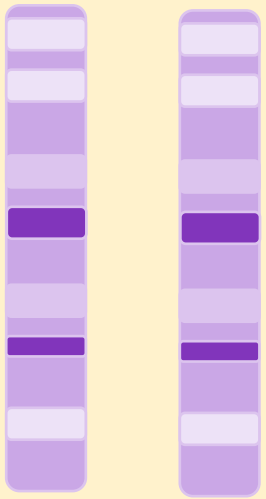
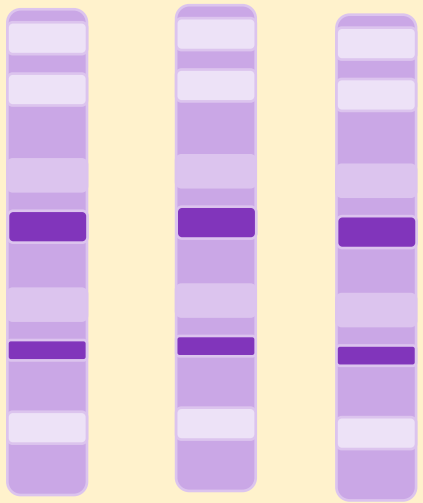
Aneuploidy – abnormal number of chromosomes



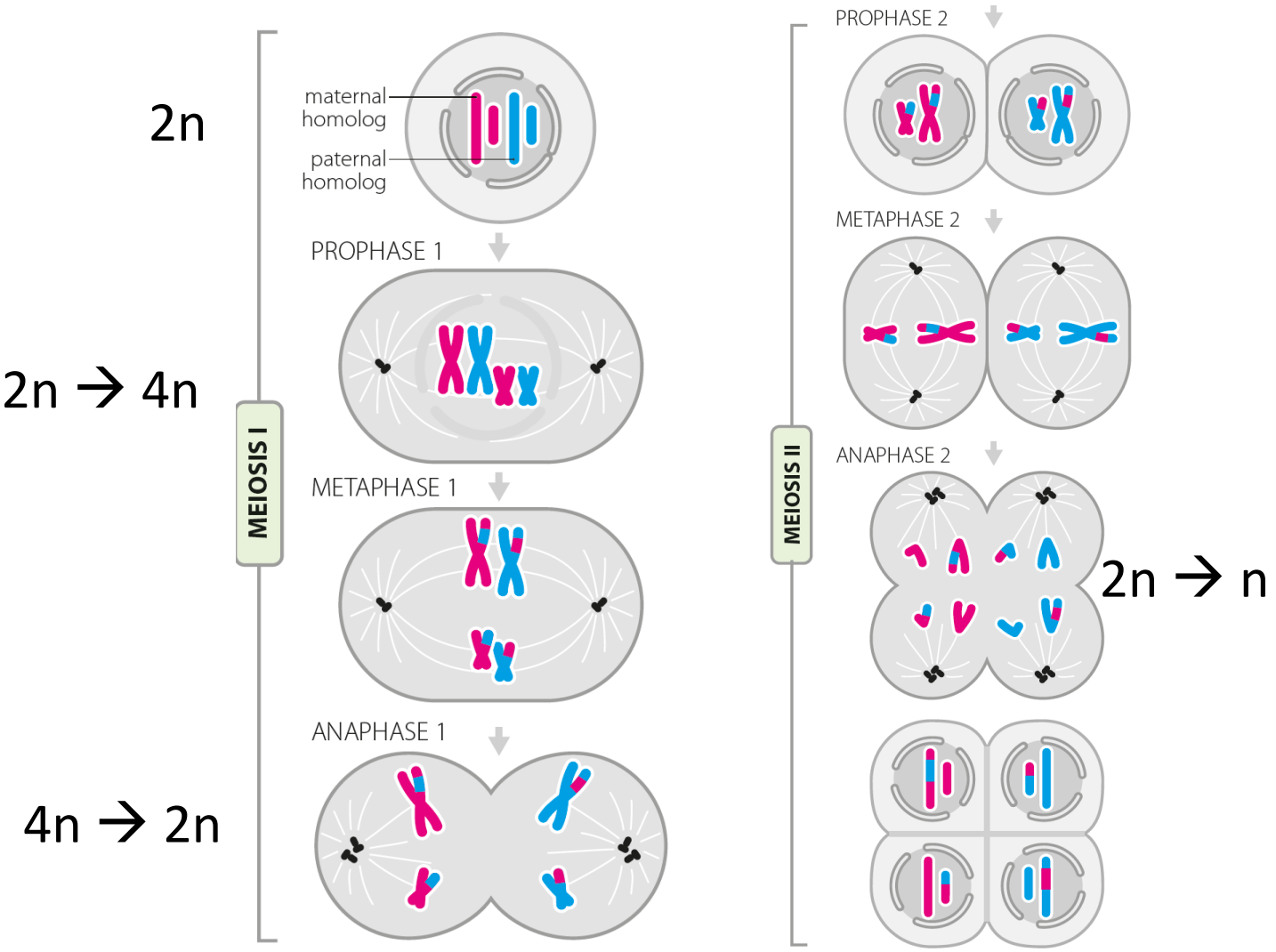
sufficient



Too much
Trisomy

Insufficient
Monosomy

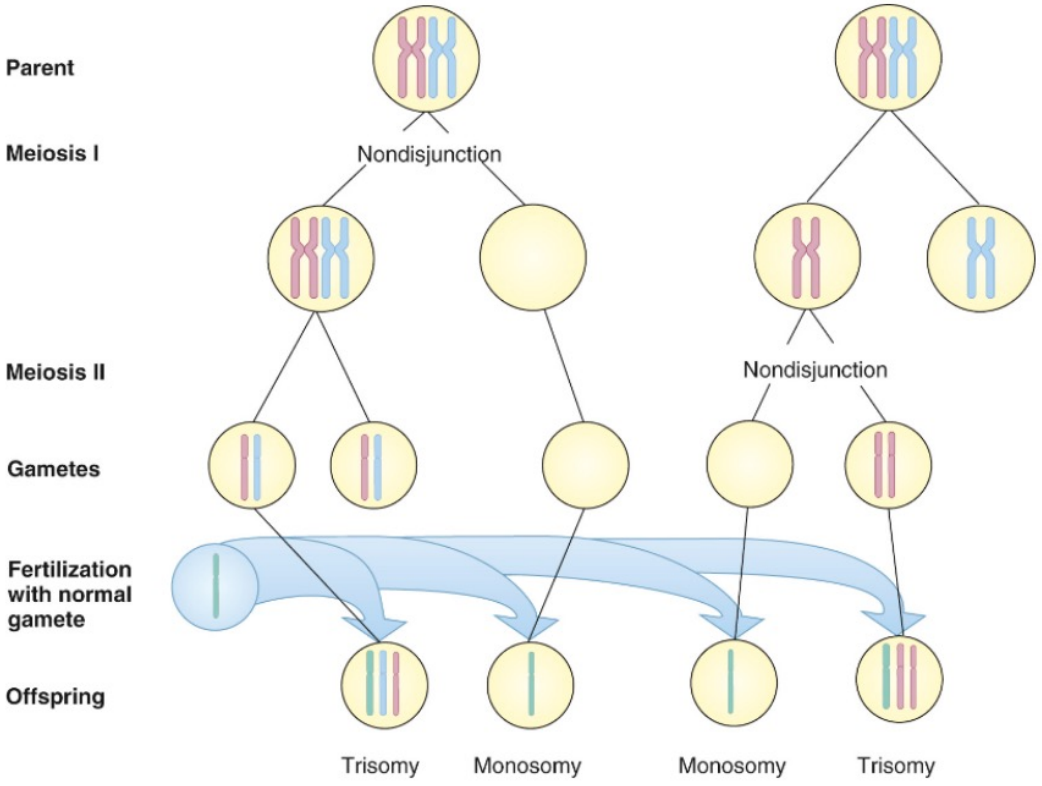
Normal meiosis



Purpose of meiosis

- To achieve **reduction** from **diploid** (2n=46) to **haploid** (n=23)
- To ensure **genetic variation** in the gametes
- Enables **random assortment** of homologues and **recombination**

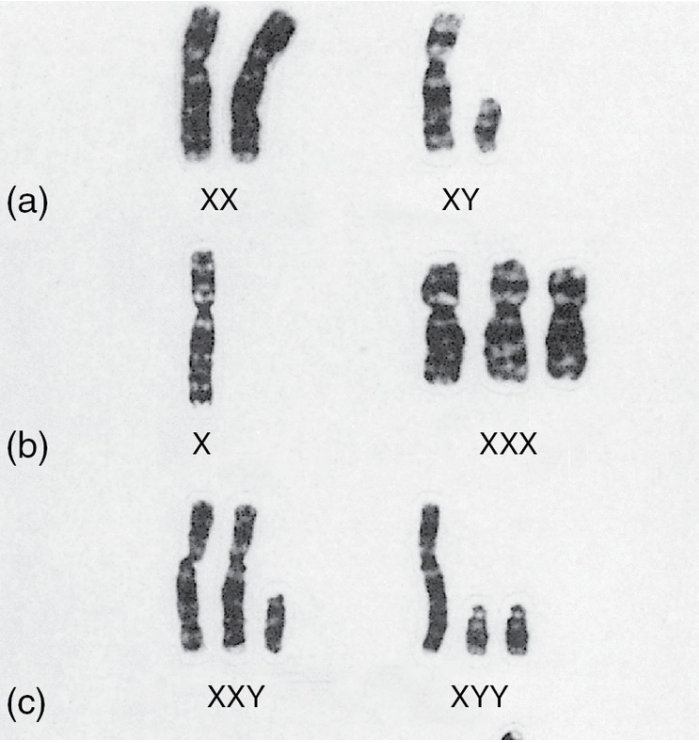
Non-disjunction



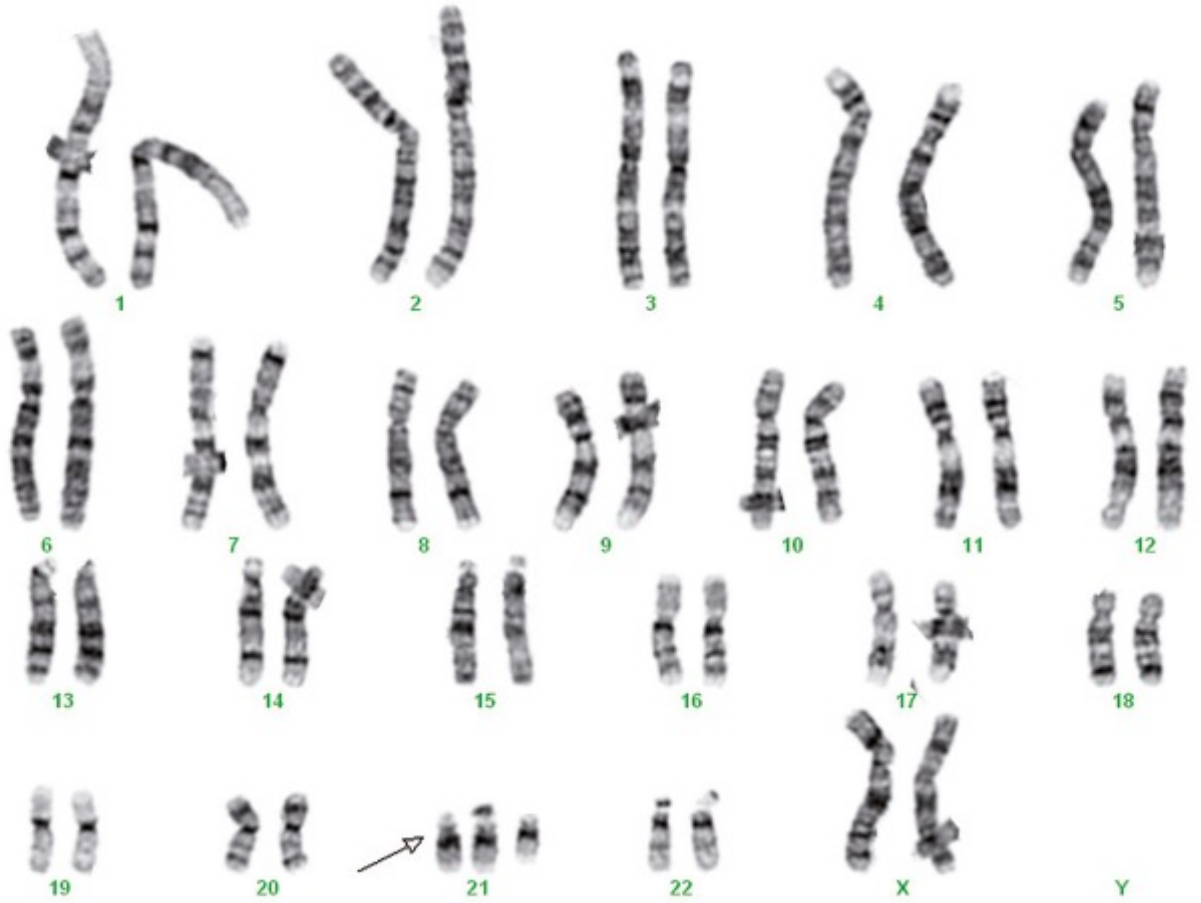
- Non-disjuncture results in uneven number of chromosomes in daughter cells.
- Can occur in either meiosis I or meiosis II
  - Meiosis I all daughter cells affected
  - Meiosis II half affected
- Always results in either +1 or -1 chromosome
  - When fertilized either trisomy or monosomy

Sex chromosome aneuploidy

- Most common form of aneuploidy
- Affect 1 in 400 males and 1 in 650 females
- Why is sex chromosome imbalance tolerated?
- **X-inactivation** of excess X chromosomes
  - Only one X-chromosome active
- **Low gene content** of Y chromosome
- Why if inactivated does abnormal number X have effect
  - Both X and Y chromosome have PAR
  - PAR- pseudo-autosomal region

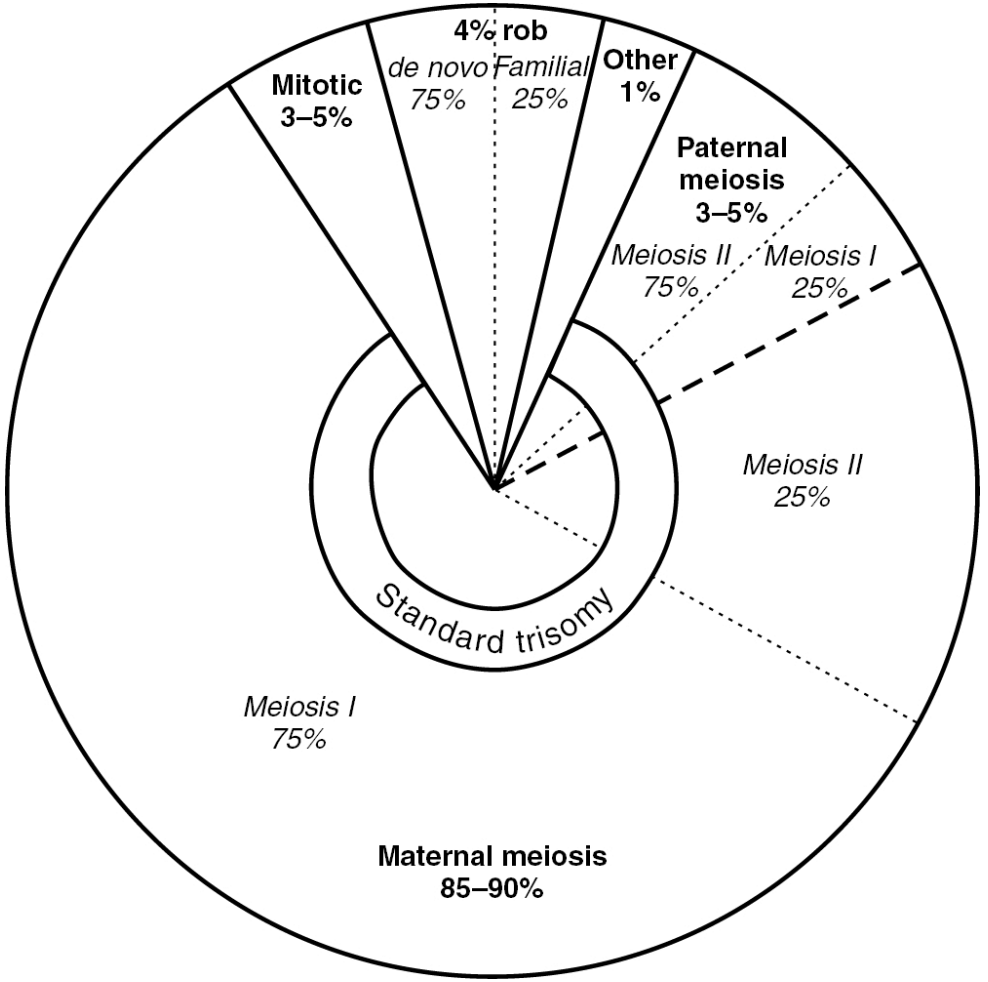


Trisomy 21



Karyotype:
47 +21 or 47, XX +21

Most trisomy 21 arises in maternal non-disjunction



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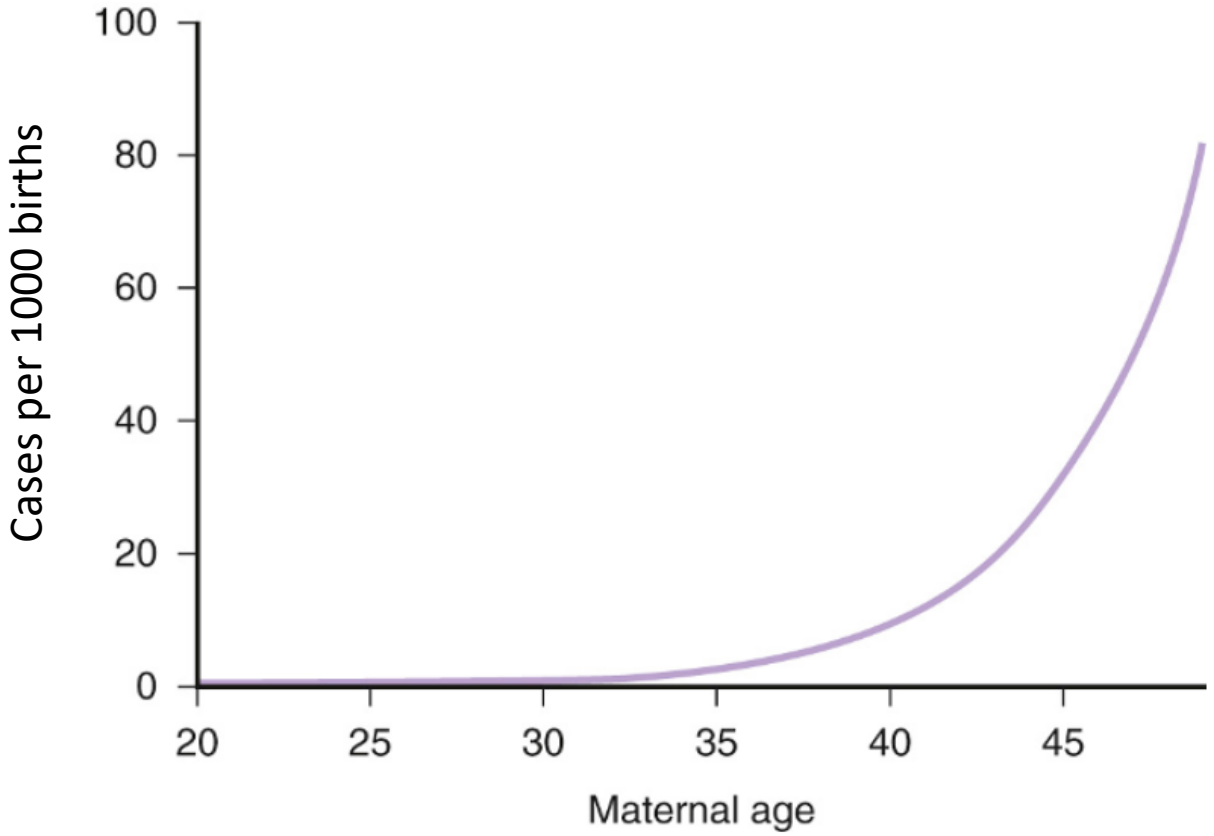
Risk of maternal non-disjunction increases with age



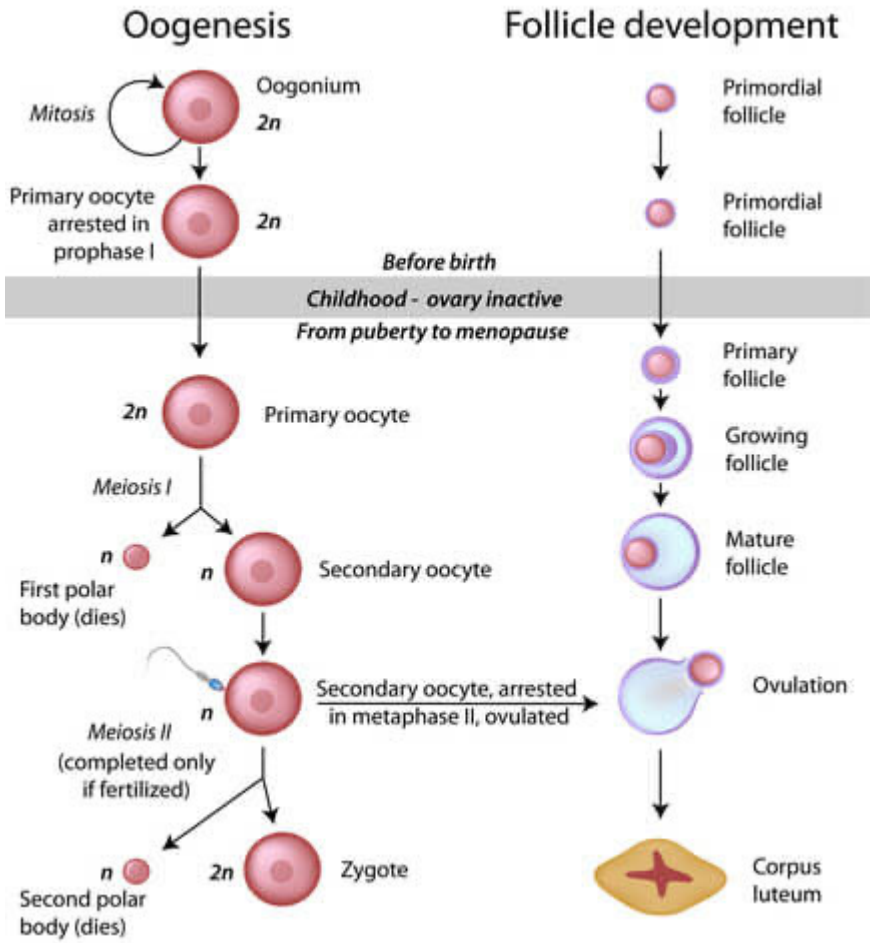
Risk of down syndrome vs maternal age

Maternal Age	Risk
Less than 30	1 in 1000
35	1 in 400
40	1 in 40
45	1 in 25

However 75% of down syndrome babies born to mothers under 35.
90% of children born to mothers of this age

1-POM-4-7: Age-associated risk: Explain the increased genetic risks associated with advanced maternal and paternal age. (Tag DA)

Why is there a maternal age effect



Vulnerability of oogenesis

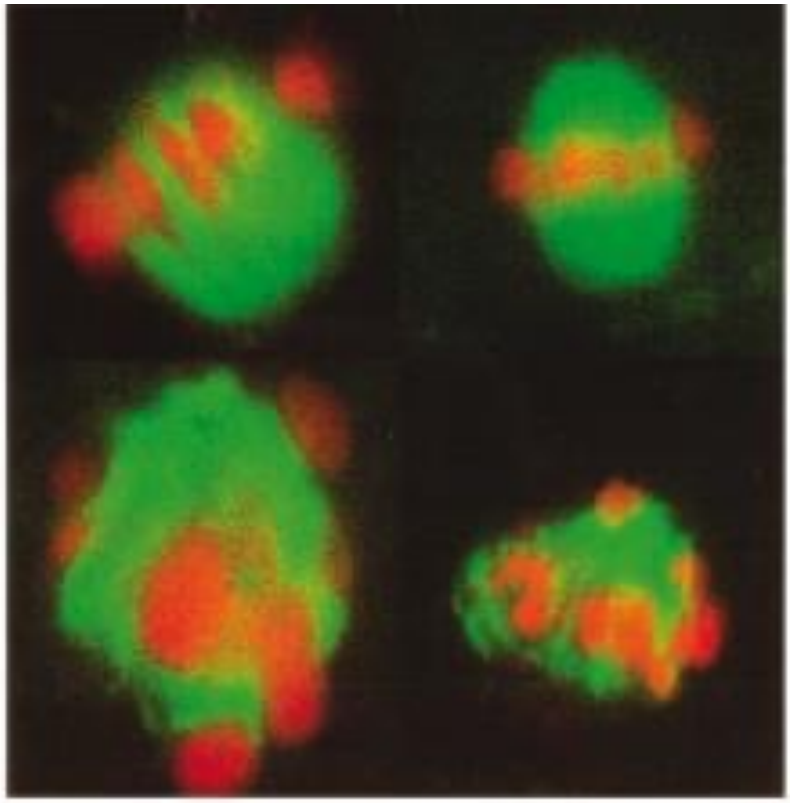
- Paused in utero in prophase I until puberty
- Secondary oocyte arrests in metaphase II
- Only competes if fertilized
- One primary oocyte yields only one ovum
- Finite number of primary oocytes

1-POM-4-7: Age-associated risk: Explain the increased genetic risks associated with advanced maternal and paternal age. (Tag DA)

oogenesis

Female non-disjunction

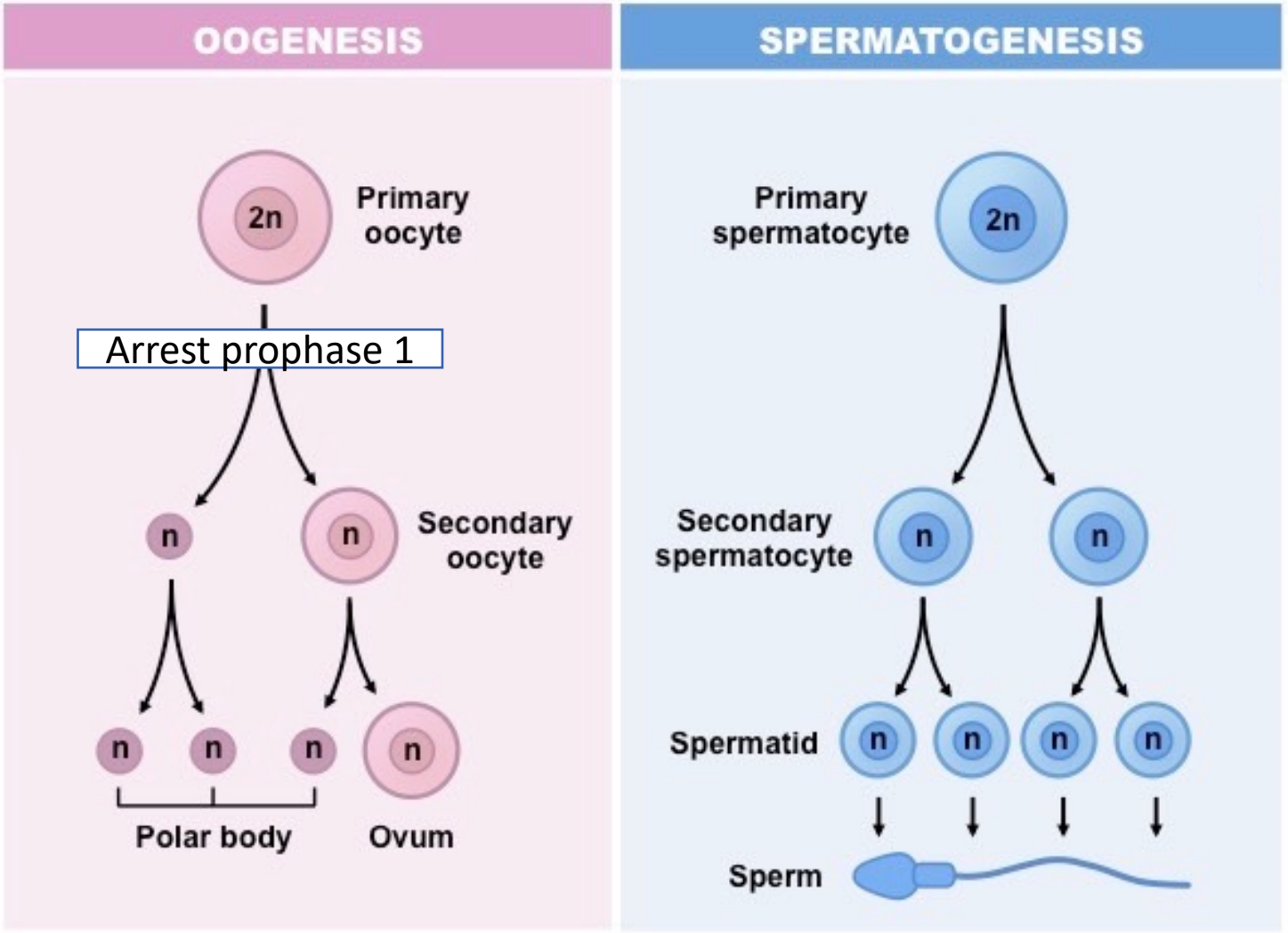
- Most **aneuploidy** caused by non-disjunction arises in oogenesis
- Likely due to **degradation** of factors which hold homologous chromatids together



22 year old ♀

40 year old ♀

Paternal age effect



Vulnerability of male meiosis

- **No equivalent to oocyte mitotic arrest**
- Primary spermatocytes undergo **~23 mitotic divisions per year** and potentially accumulate defects

1-POM-4-7: Age-associated risk: Explain the increased genetic risks associated with advanced maternal and paternal age. (Tag DA)

Paternal Age Effect

Paternal Age not risk factor increased aneuploidy

Does affect a subset of single gene disorders

caused by point mutations in *FGFR2, FGFR3* and *RET* including:

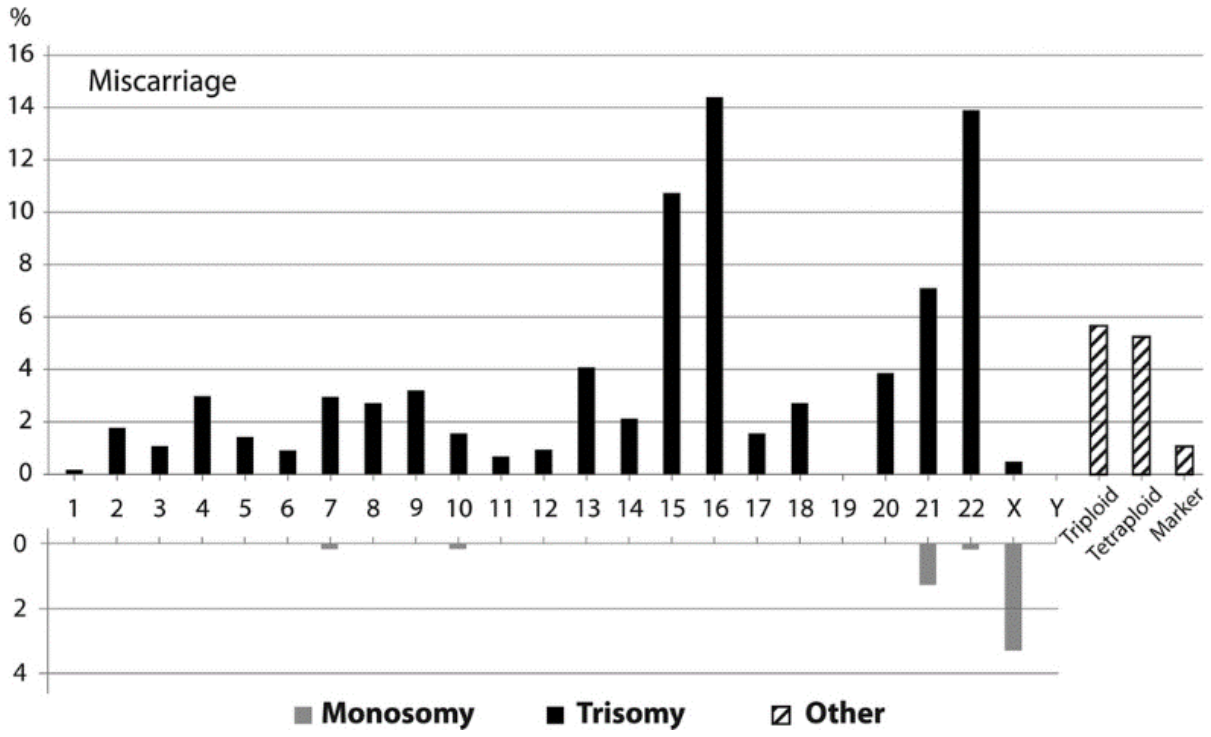
- Apert syndrome
- Crouzon syndrome
- Pfeiffer syndrome

Thought to be enhanced by ‘selfish spermatogonial selection’ resulting from a selective advantage over neighbouring wild type cells

Paternal role in aneuplody

- Although no age effect some aneuploidy paternal
  - smoking a risk factor (not maternal)
- 80% 45 X
- 46% 47 XXY
- 100% 47 XYY
- 8% 47 +21

Pregnancy risk and aneuploidy



- Aneuploidy – 5% still births 50% spontaneous abortions
- 5% all clinically recognized pregnancies
- Trisomy of all chromosomes has been detected prenatally
- Most trisomies not compatible with life
  - 21, 18, 13
- Monosomy is very poorly tolerated
- Estimated 50% preimplantation embryos

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Mosaicism

- Presence of two or more populations of cells with different genotypes
  - X-inactivation results in mosaic expression
- Mosaicism can arise two mechanisms
  - Non disjuncture during early development
  - Loss of extra chromosome in early development
- Results in generally milder phenotype
  - Some lethal aneuploidy survivable if mosaic (trisomy 9 or 8)
  - Most common mosaic 46,XX/45,X mosaic 46,XY/45,X
- Everyone thought to be mosaic

Progress check

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- Karyotype – chromosome count of individual
- Performed on metaphase chromosomes
- Various staining methods classically Gisema
- Gisema stain basis of nomenclature
- Can detect major chromosome abnormalities

Part 3

Part 2

- Aneuploidy - chromosomes not 46 or multiple of 23
- Common during pregnancy
- Most not compatible with life
- Sex chromosomes exception
- Can be maternal or paternal
- Maternal age risk factor

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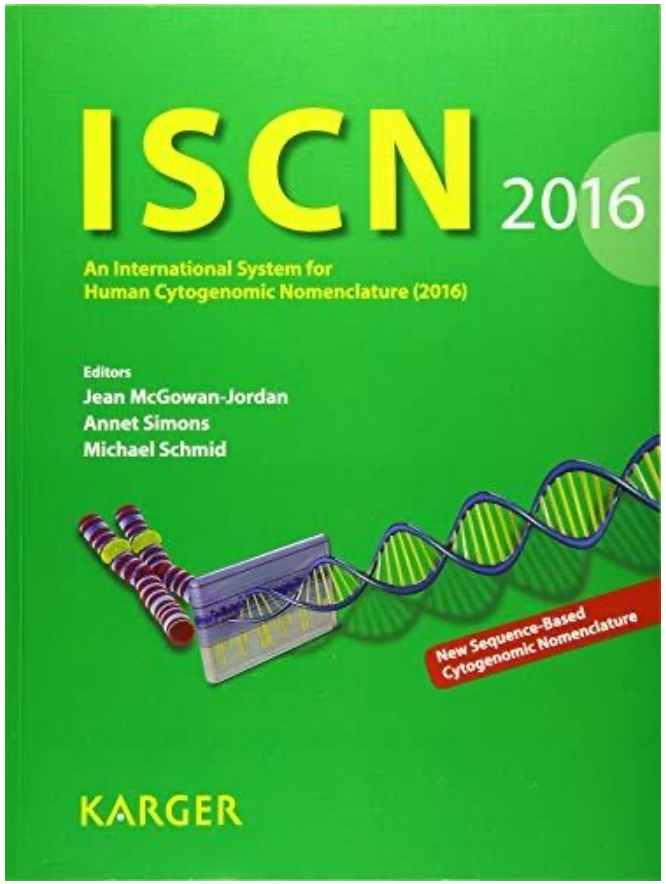

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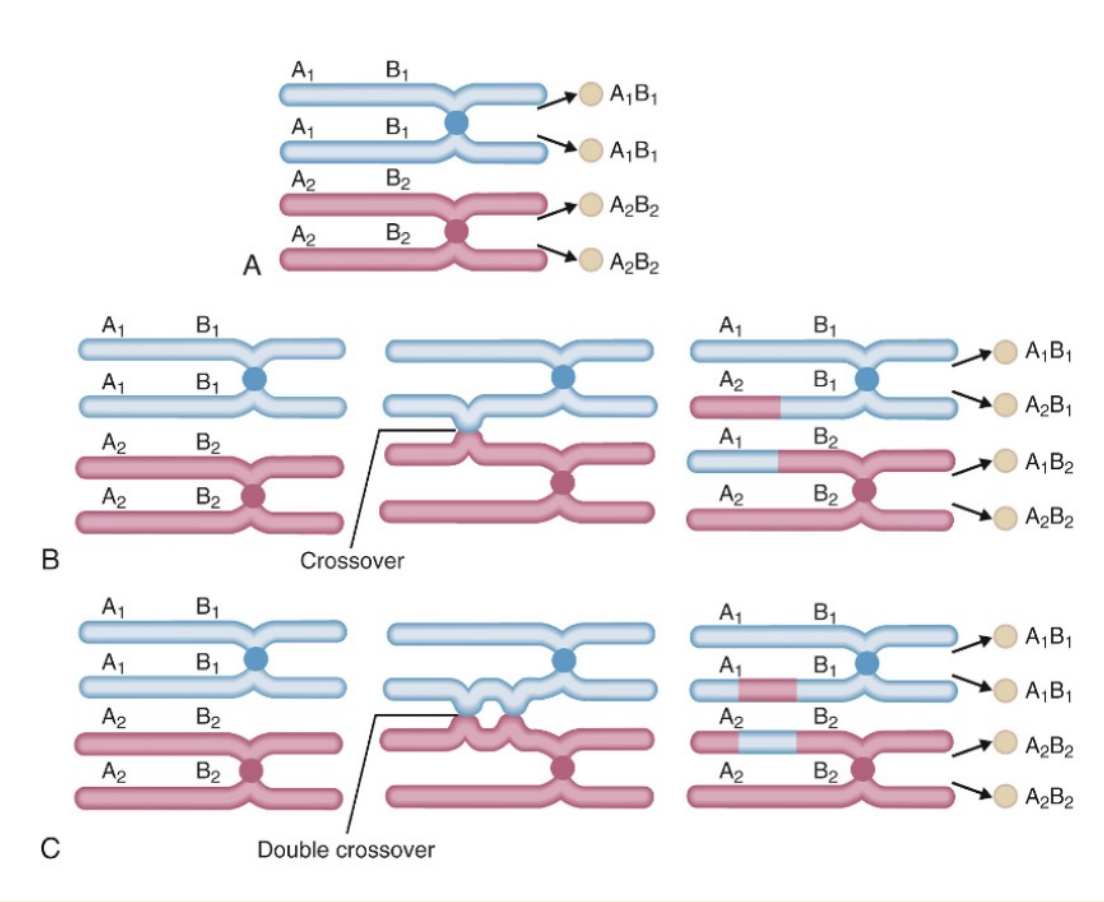
Abnormalities of chromosome structure

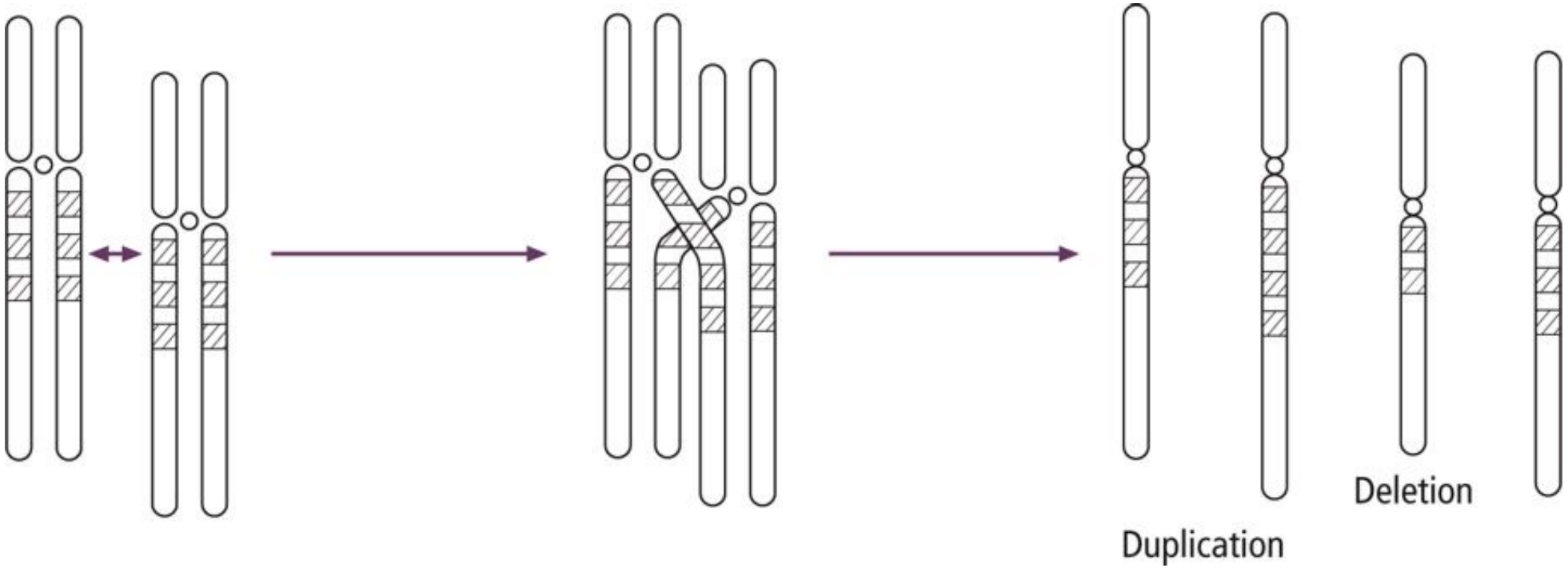
1-POM-4-3 Explain the basis of other types of chromosomal abnormalities, and their relevance to clinical disease

Cross over chromosomes

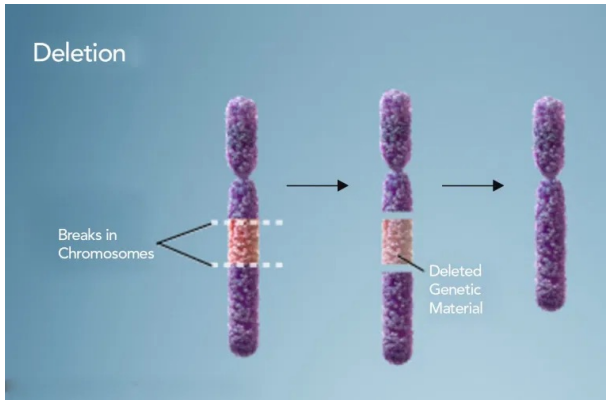


- Occurs in prophase I
- increases genetic diversity
- Pairs of chromosomes align
- Chiasma form and crossover occurs
- 1-3 times per chromosome per meiosis
- However sometime goes wrong

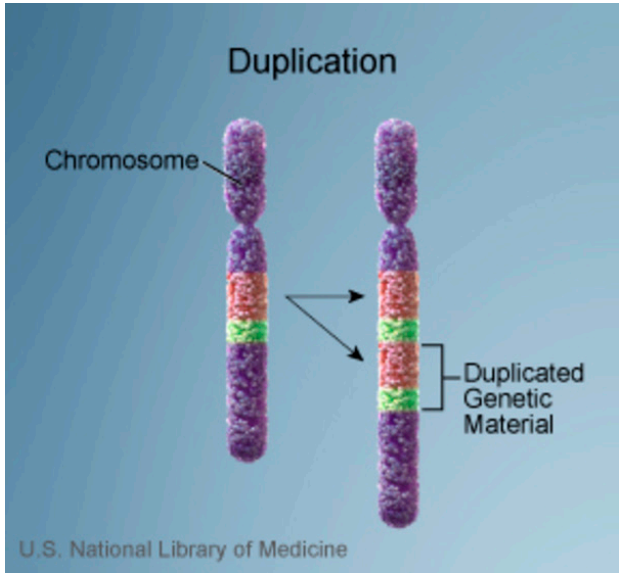
Unequal crossover



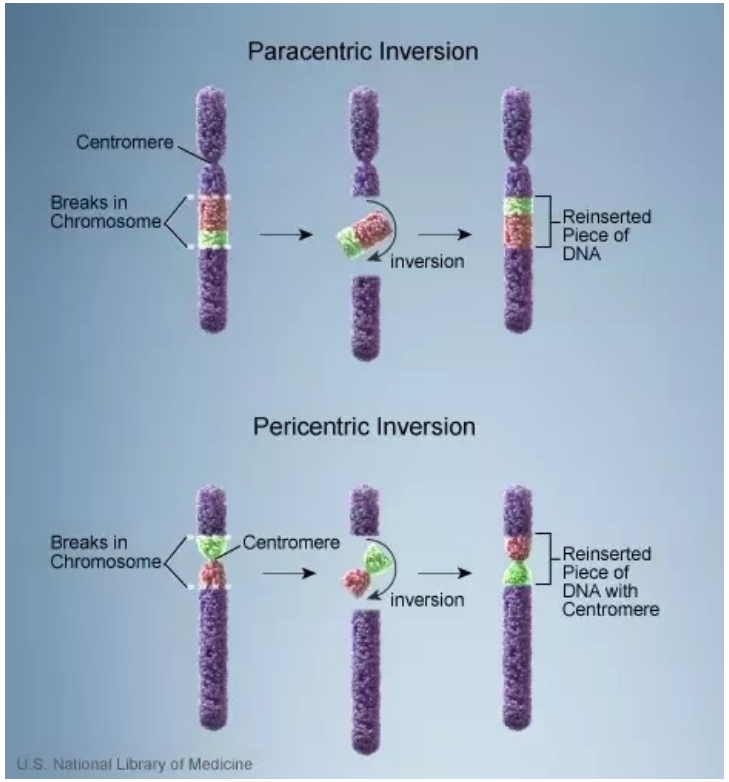
Single chromosome abnormalities



Can be the result of unequal cross over
Breaks in chromosome
Can occur at ends



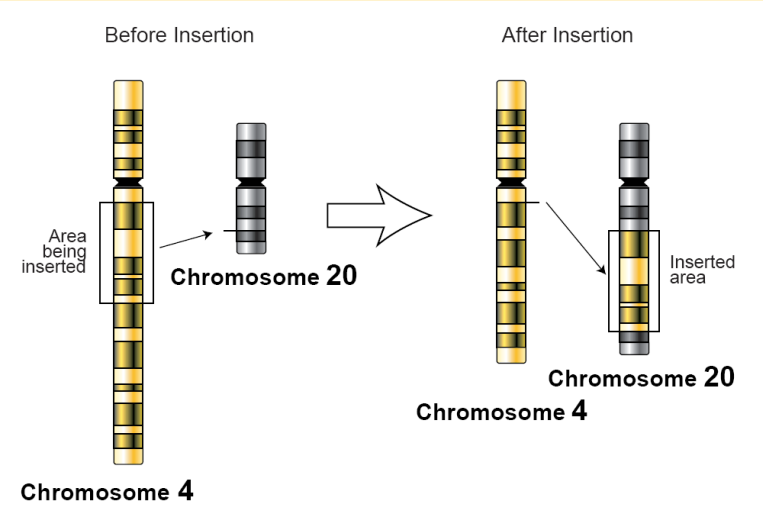
Most often caused by unequal cross over

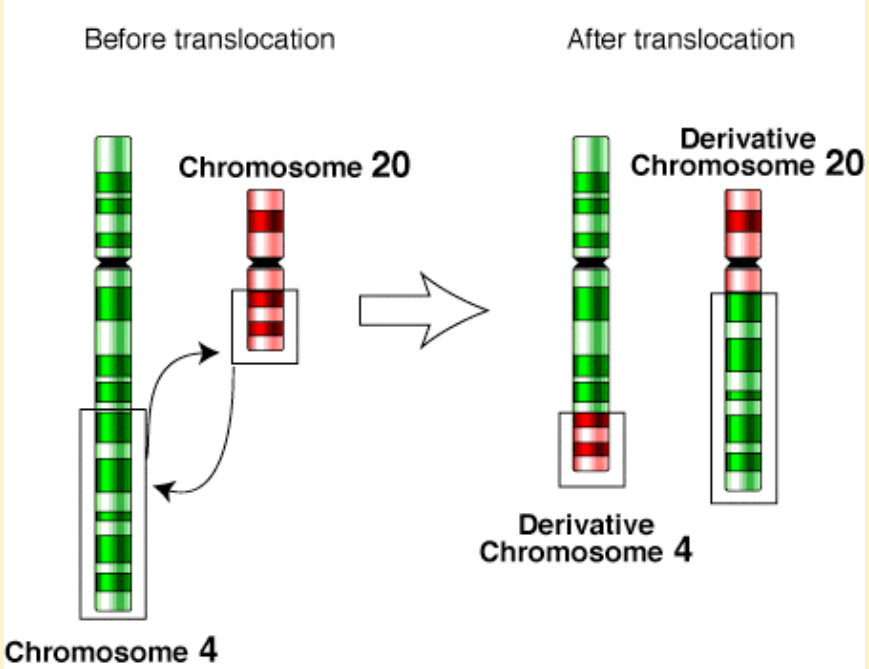


Carriers often unaffected
Estimated to occur in 1 in 1000 people
Can cause reproductive problems
Children with deletions/insertions

1-POM-4-3 Explain the basis of other types of chromosomal abnormalities, and their relevance to clinical disease

Two chromosome abnormalities



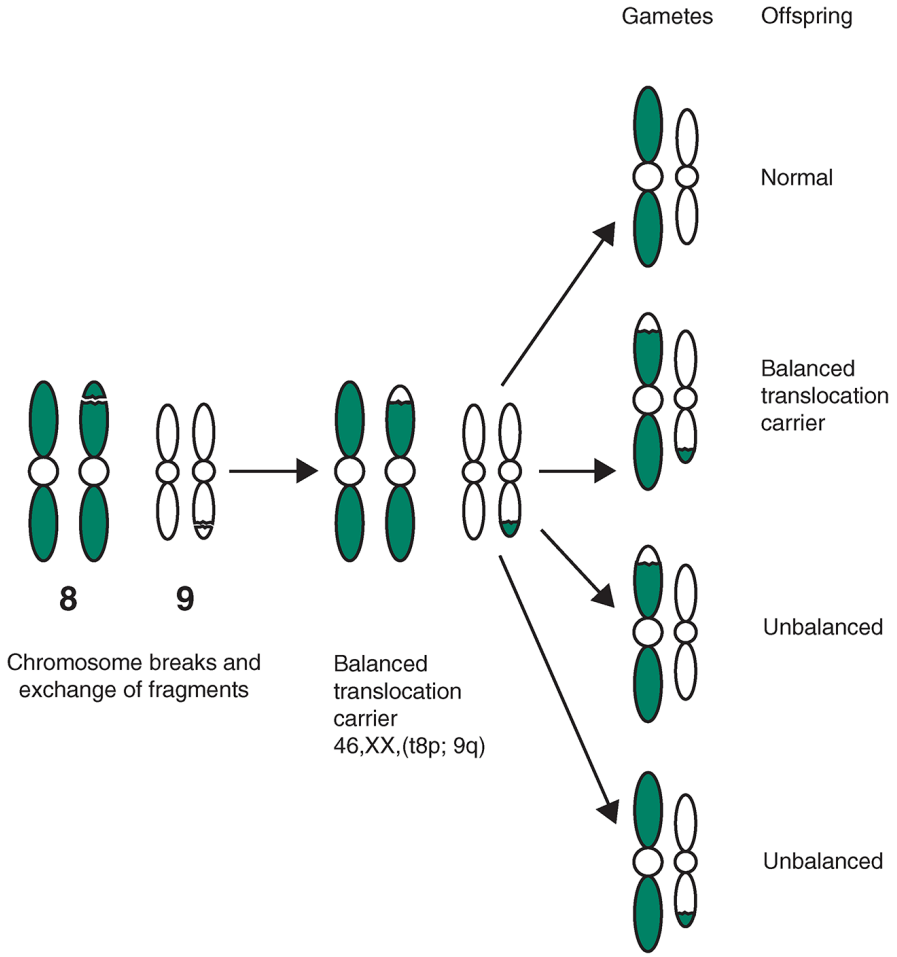


If balanced does not affect carrier may cause problems in off spring

- can cause partial trisomy or monosomy (eg Cri-du-chat syndrome, 5p monosomy)

Can occur in Somatic cells cf Philadelphia chromosome t(9;22)(q34;q11) CML (Genetics of cancer)

Inheritance of chromosome abnormalities



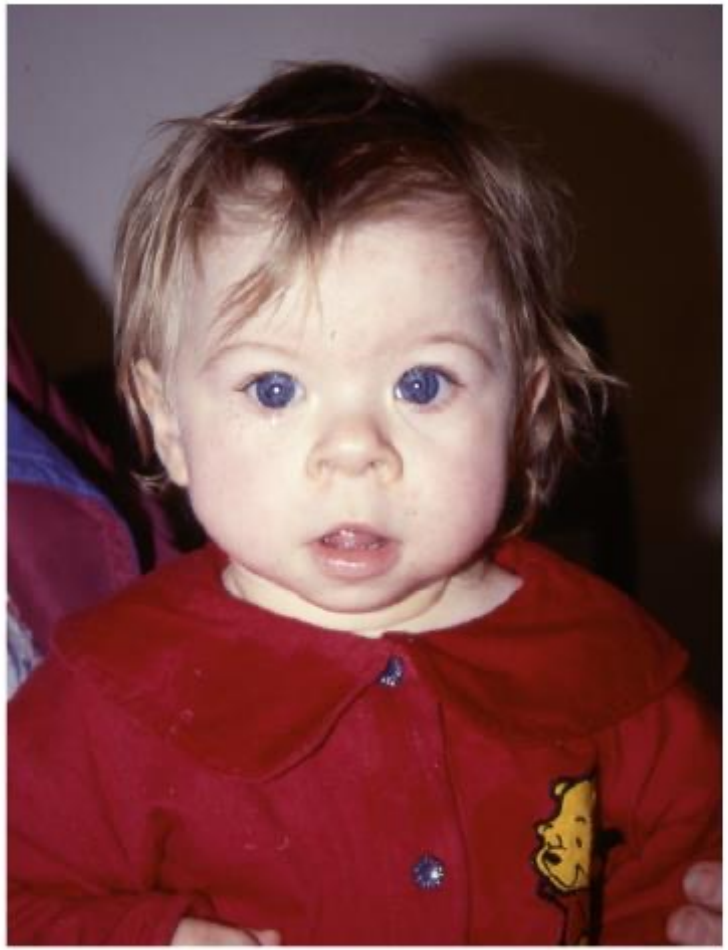
Many chromosomal abnormalities are de-novo
Some people are unidentified carriers
Can have offspring which are affected.

1-POM-4-3 Explain the basis of other types of chromosomal abnormalities, and their relevance to clinical disease

Chromosomal deletions

- Microscopic – be detected easily in microscope
  - Cri-du-chat syndrome 46,XY,del(5p)
- Microdeletion- seen in v high resolution banding; molecular genetics
  - Despite name still 20+ genes deleted
  - Velocardiofacial/DiGeorge syndrome 22q11.2 del

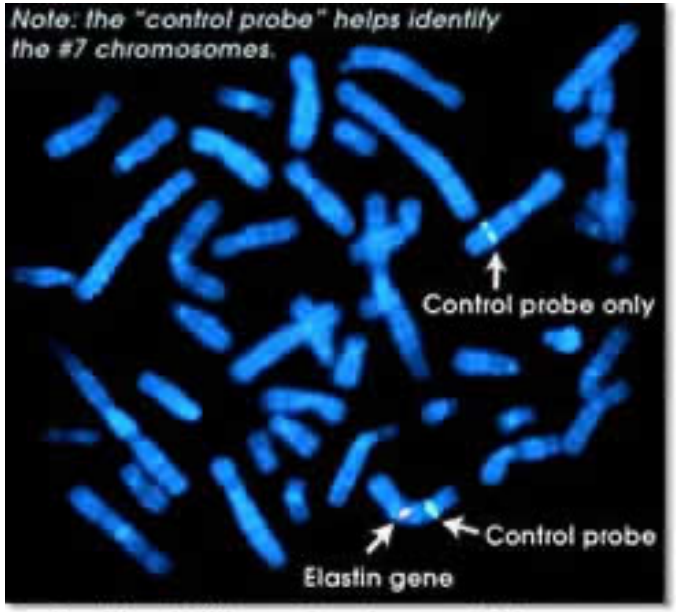
Williams syndrome (7q11.23 deletion)



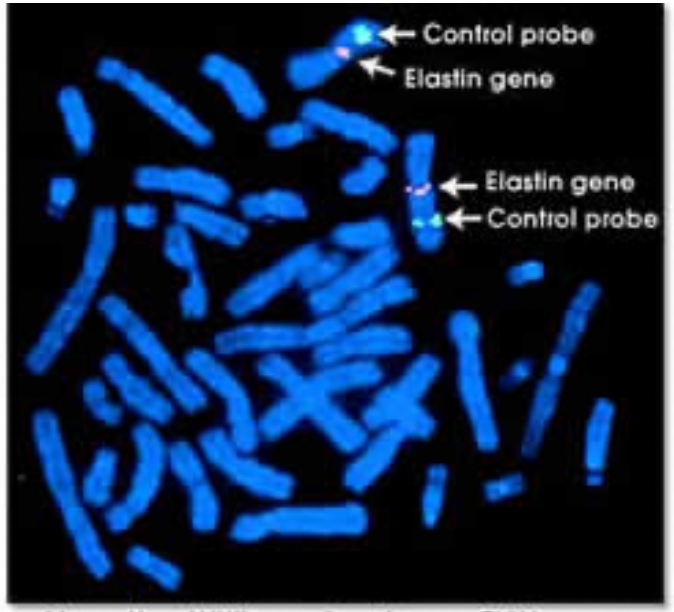
- Long philtrum
- Short, upturned nose
- Arched eyebrows
- Supravalvular aortic stenosis
- Friendly, social ‘cocktail party’ personality – an absence of social anxiety

- Phenotypes caused by imbalance of genes
  - Deletion from one chromosome
- which are unrelated apart from their location

Williams syndrome (7q11.23 deletion)



Positive Williams Syndrome FISH assay
(*Chromosome 7*)
The elastin gene is found on only one chromosome. The other copy carries an elastin gene deletion.



Negative Williams Syndrome FISH assay
(*Chromosome 7*)
The elastin gene is found on both chromosomes. This individual does not have Williams Syndrome.

Deletion too small to detect using standard karyotyping

Can be detected using targeted FISH

- Fluorescent in situ hybridisation

Lack of elastin on affected chromosome

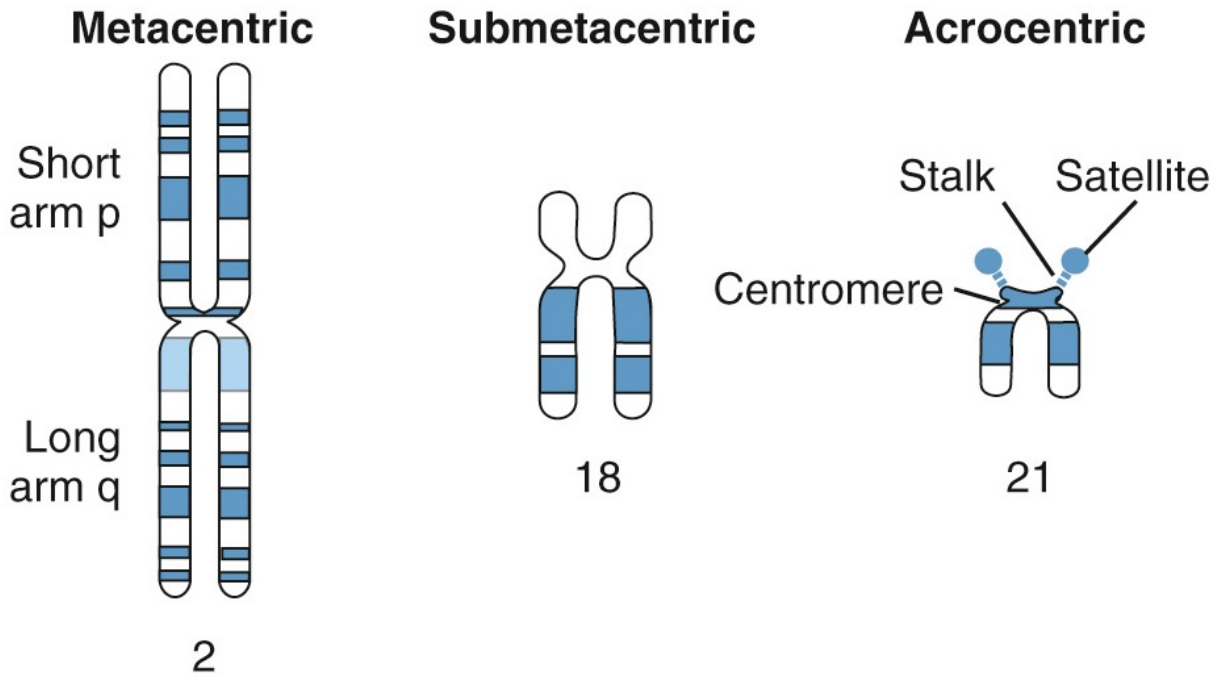
7q11.23 duplication syndrome

- Delayed speech development
- Autistic behaviours that affect social interaction and communication
- Dilatation of the aorta
- Flat eyebrows
- Broad nose and short philtrum

Duplications usually have a milder phenotype than the corresponding deletion

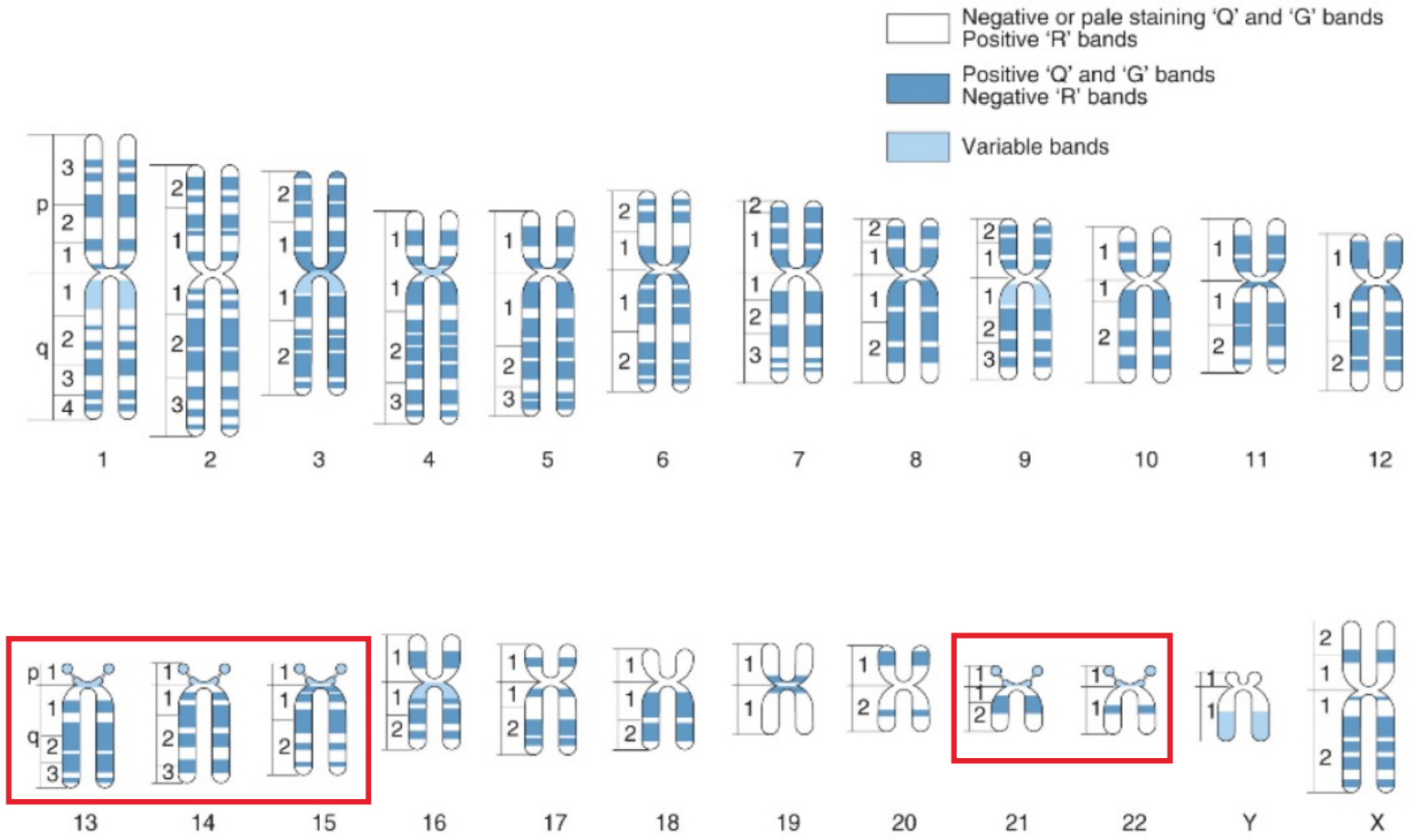
Robertsonian translocation

Classes of chromosomes



1-POM-4-3 Explain the basis of other types of chromosomal abnormalities, and their relevance to clinical disease

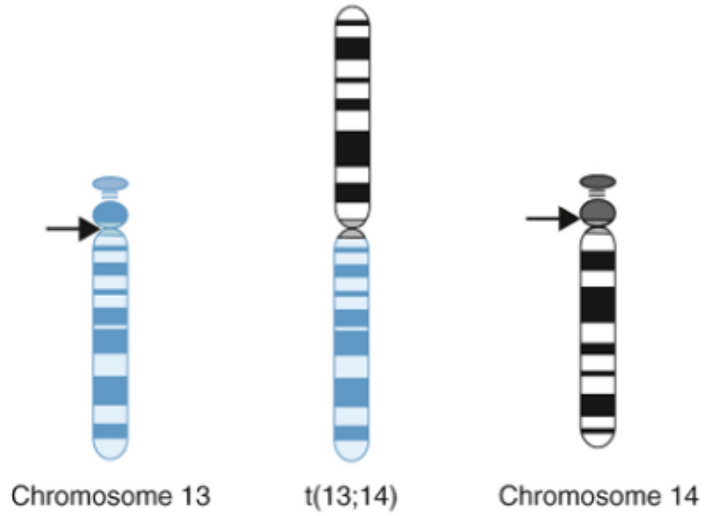
Different classes of chromosomes



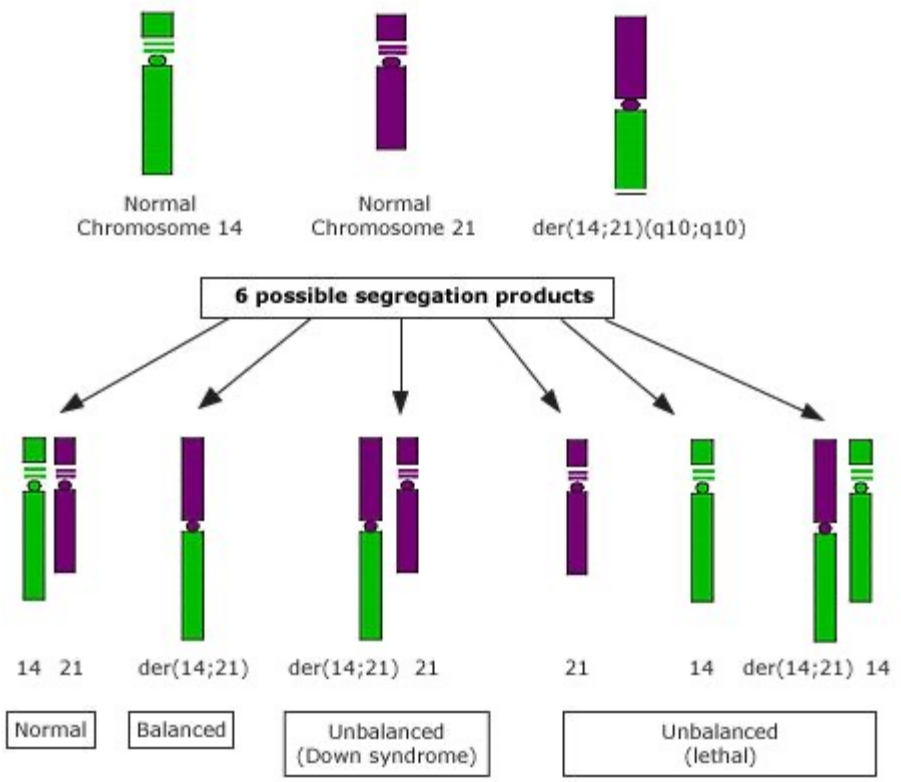
1-POM-4-3 Explain the basis of other types of chromosomal abnormalities, and their relevance to clinical disease

Robertsonian translocation

- Affects 1 in 1000 people
- Occurs between acrocentric chromosomes
- Can be homologous or non homologous
- Most common 13 and 14; 14 and 15; 14 and 21
- Most people show no affects
- Can cause problems in offspring

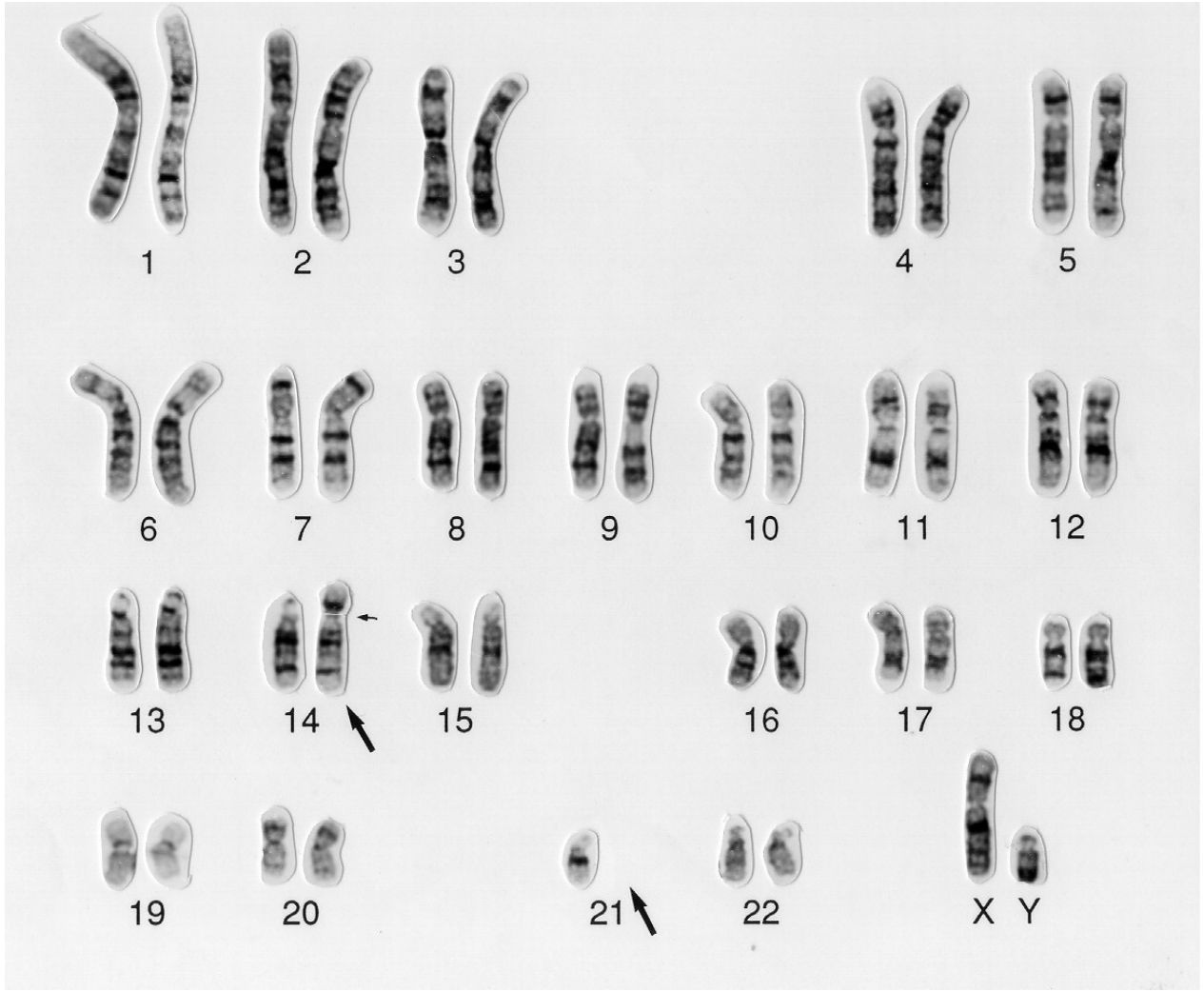


Robertsonian translocation-in reproduction



- Whilst silent in carriers can produce affected offspring
- Same true for other chromosomal abnormalities

Robertsonian translocation - karyotype



45, XY, rob (14;21) (q10;q10)
45, XY, t(14;21) (q10;q10)
45, XY, der(14;21) (q10;q10)

Progress check

Part 1

- Karyotype – chromosome count of individual
- Performed on metaphase chromosomes
- Various staining methods classically Gisema
- Gisema stain basis of nomenclature
- Can detect major chromosome abnormalities

Part 2

- Aneuploidy - chromosomes not 46 or multiple
- Common during pregnancy
- Most not compatible with life
- Sex chromosomes exception
- Can be maternal or paternal
- Maternal age risk factor

Part 3

- Variety of chromosomal abnormalities
- Deletions, insertions, translocations
- May be obvious in karyotype or not visible
- Many silent in carriers but apparent in offspring

1-POM-4-3: Chromosomal abnormalities: Recall the normal human karyotype, chromosome banding and nomenclature, explain the basis of nondisjunction and other types of chromosomal abnormalities, and their relevance to disease.

Progress check

Part 1

- Karyotype – chromosome count of individual
- Performed on metaphase chromosomes
- Various staining methods classically Gisema
- Gisema stain basis of nomenclature
- Can detect major chromosome abnormalities
- Whole genome sequencing now being used

Part 2

- Aneuploidy - chromosomes not 46 or multiple
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