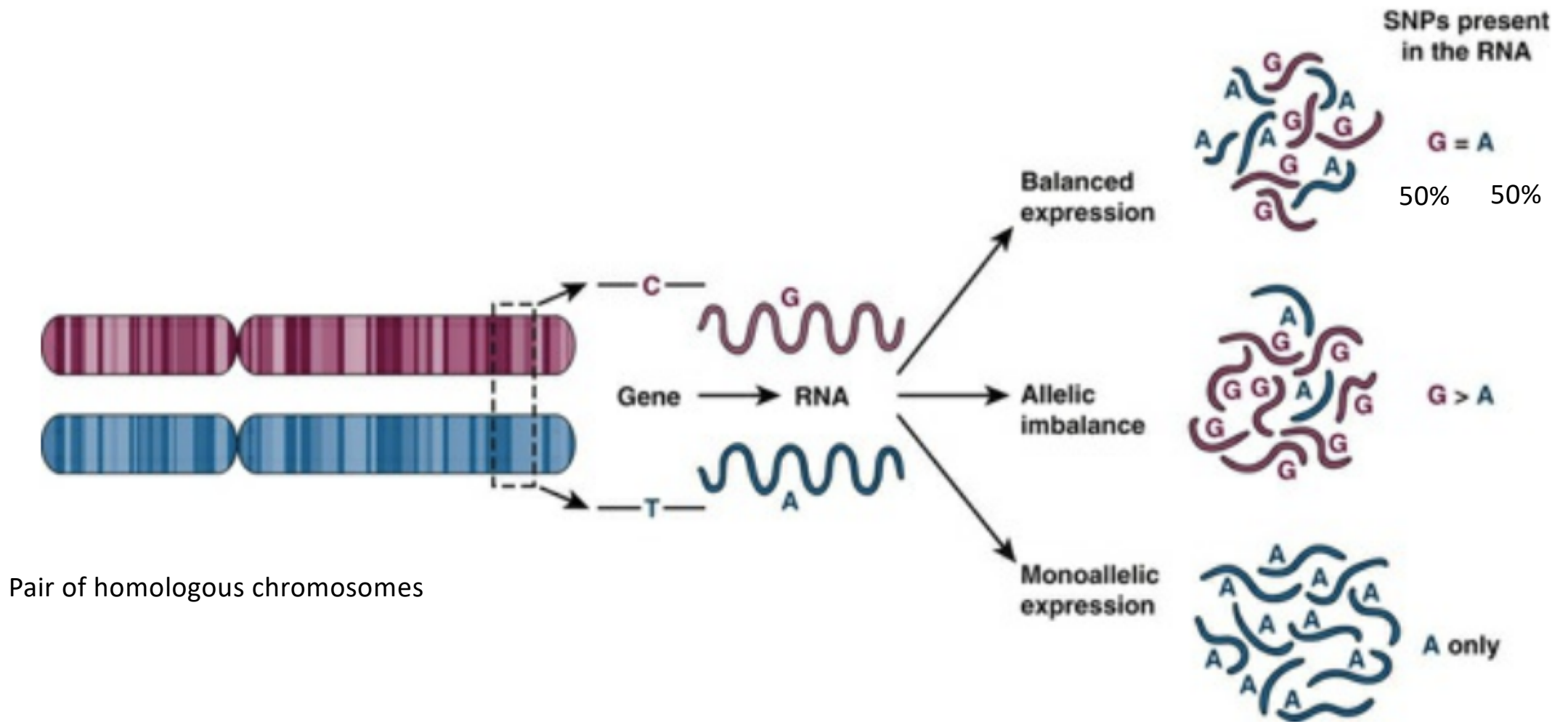
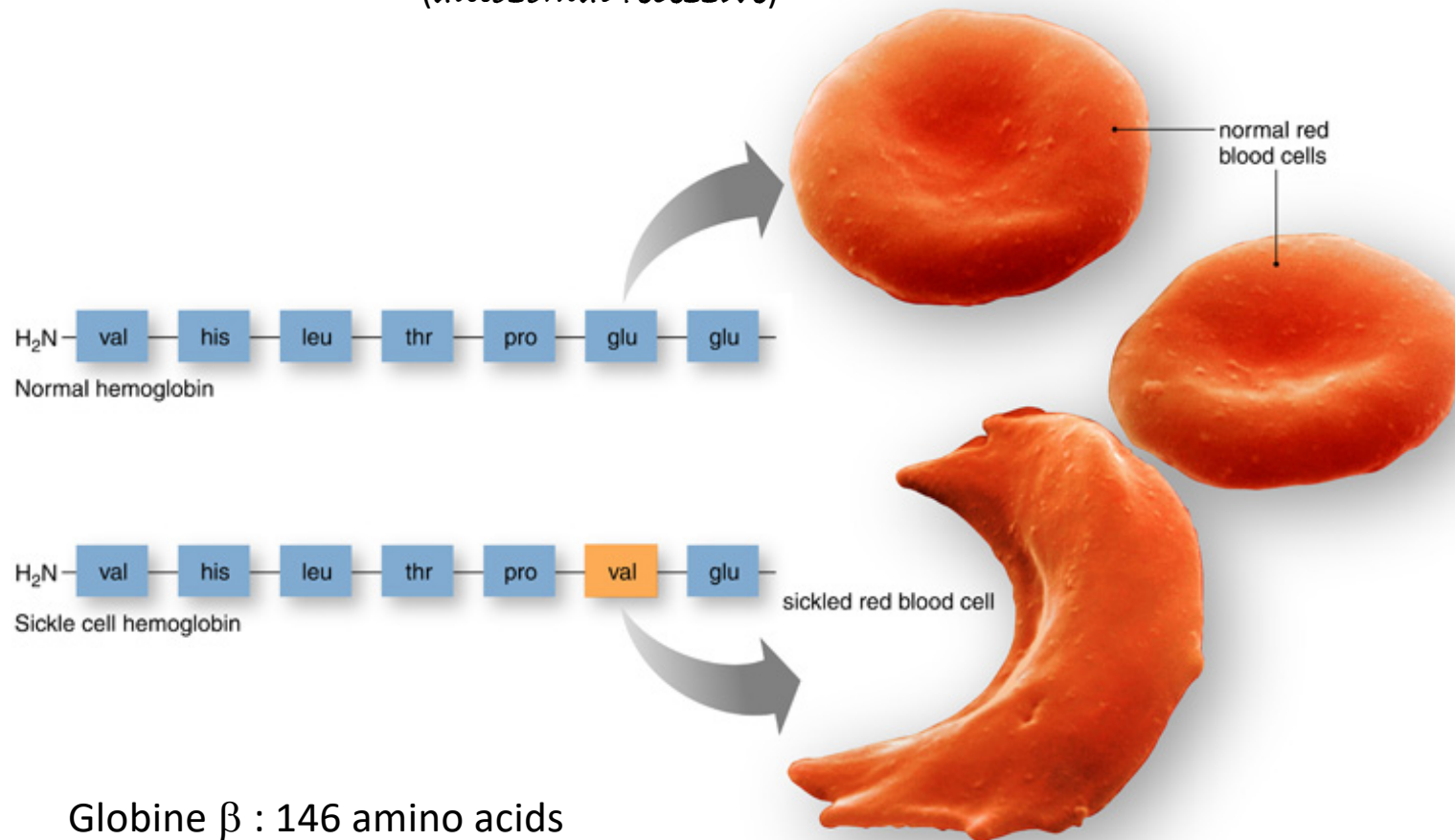


The **usual case** : balanced, biallelic expression



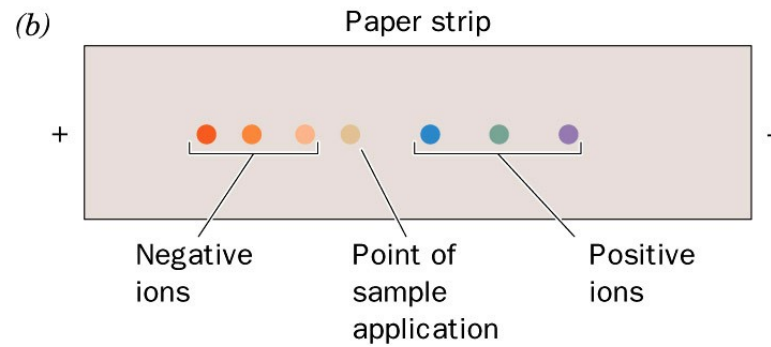
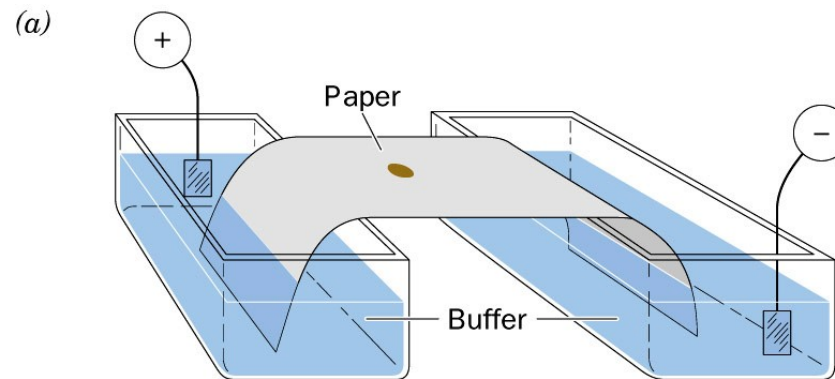
Sickled cells disease

(autosomal recessive)



Globine β : 146 amino acids
Only 1 difference, at position 6

Electrophoresis :



Paper electrophoresis

Linus Pauling, 1949: HbS has different charge.

Electrophoresis .

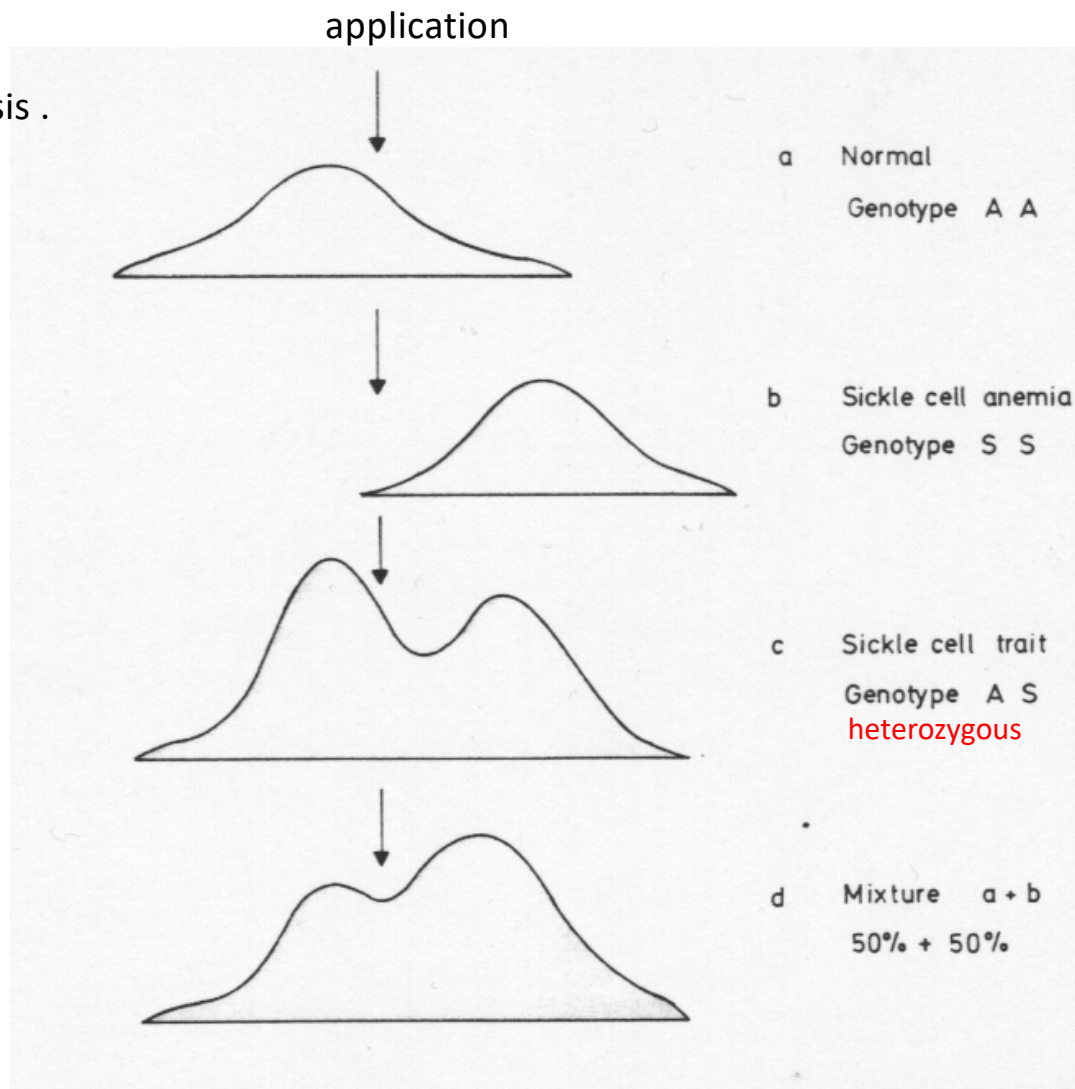
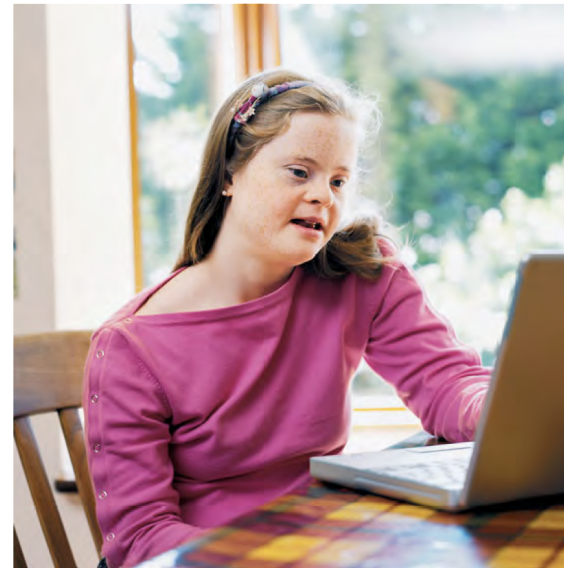
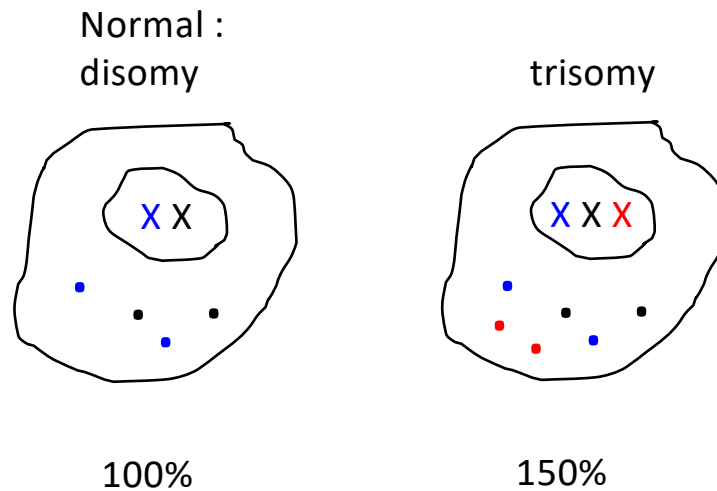


Fig. 7.27 a-d. Zone electrophoresis diagram of hemoglobins at pH = 6.9. a Normal homozygote (AA); b patient with sickle cell anemia (SS); c sickle cell trait (AS); d mixture of equal parts of HbA and HbS. Arrow, starting point of electrophoresis. (From Pauling et al. 1949 [195])

Genetic dosage is important

Trisomy 21
47, XX +21



Each chromosome is used equally :

2 chromosomes : protein concentration = 100%

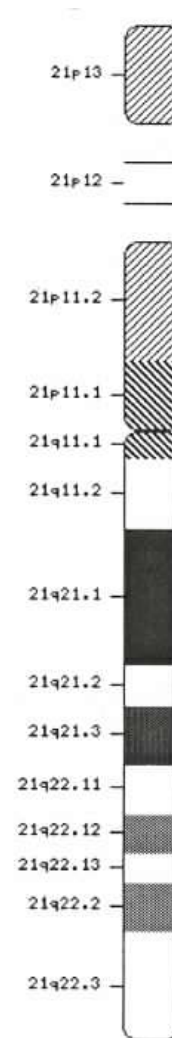
3 chromosomes : protein concentration = 150 %

Chromosome 21

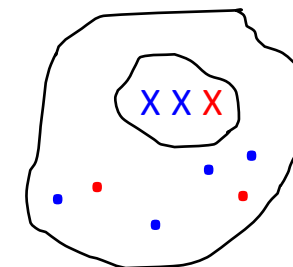
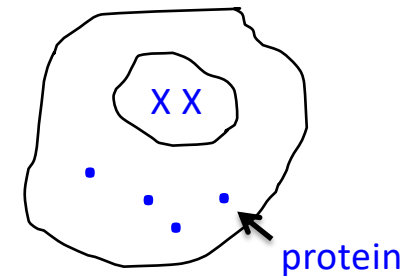
47 millions base pairs

352 protein coding genes

In trisomy 21,
352 proteins are
expressed about 150% .

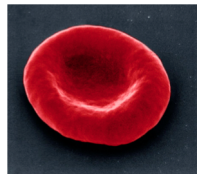


Normal (disomic) cell :

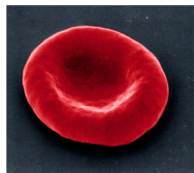
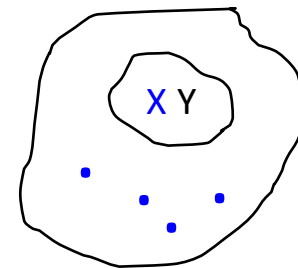
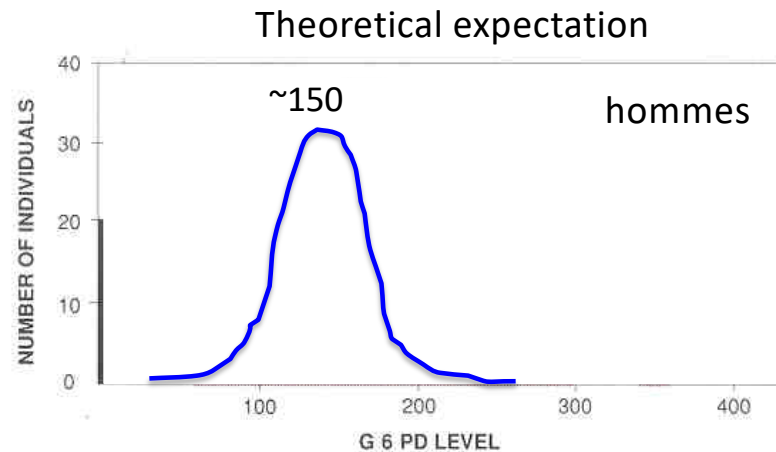


Trisomic cell.

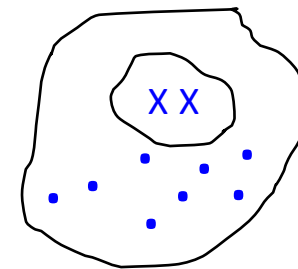
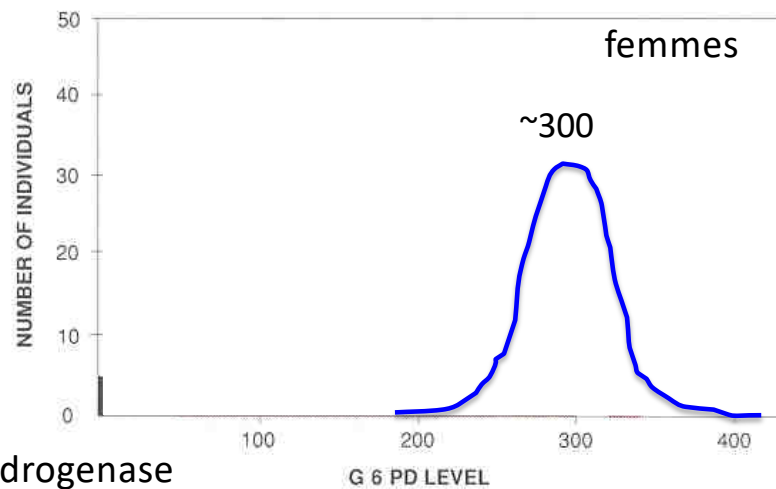
For genes located on the X chromosome
women (X X) should produce twice as much of a protein compare to men (only 1 X)



R. B. C.
from a man



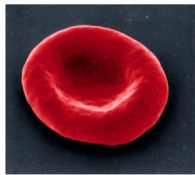
R. B. C. from
a woman



Glucose-6-Phosphate Dehydrogenase

Observations do not confirm the expectation.

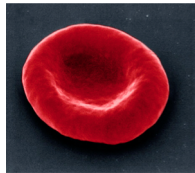
G6PD gene is located on X chromosome.



R. B. C. from a man



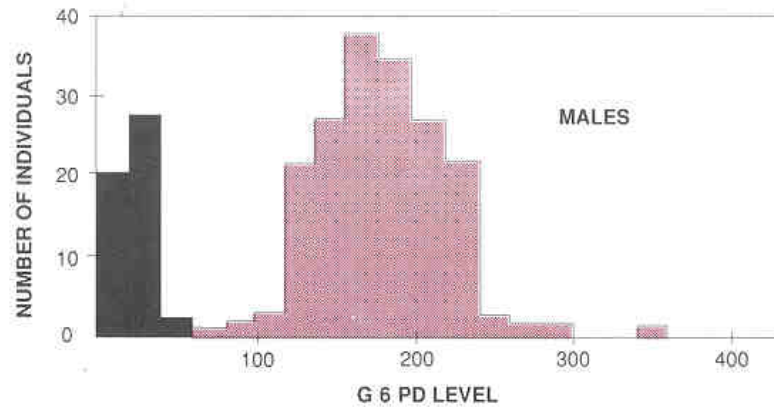
Identical amount of G6PD



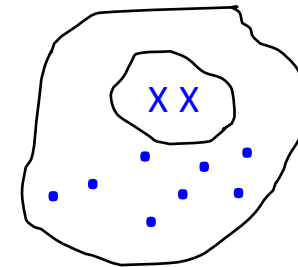
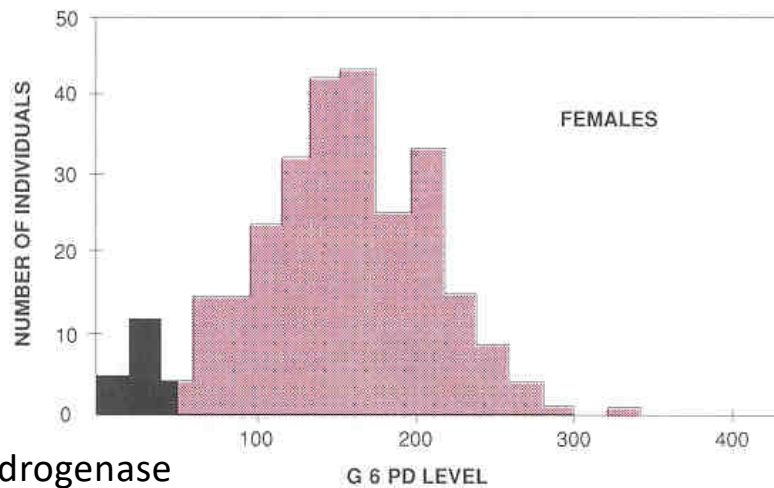
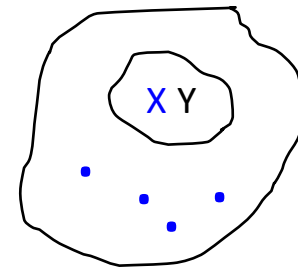
R. B. C. from a woman

Glucose-6-Phosphate Dehydrogenase

Observation :



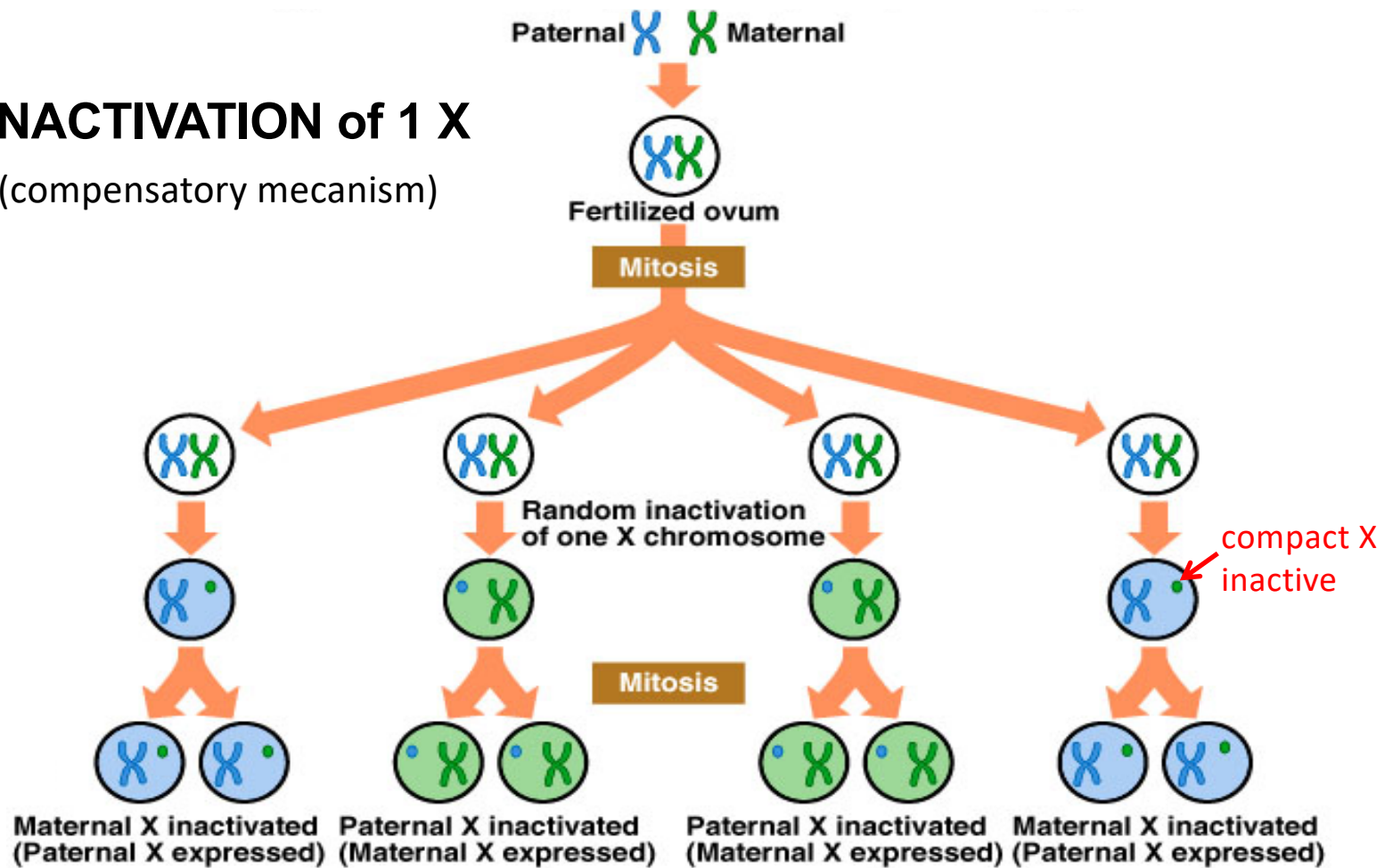
Theoretical expectation:



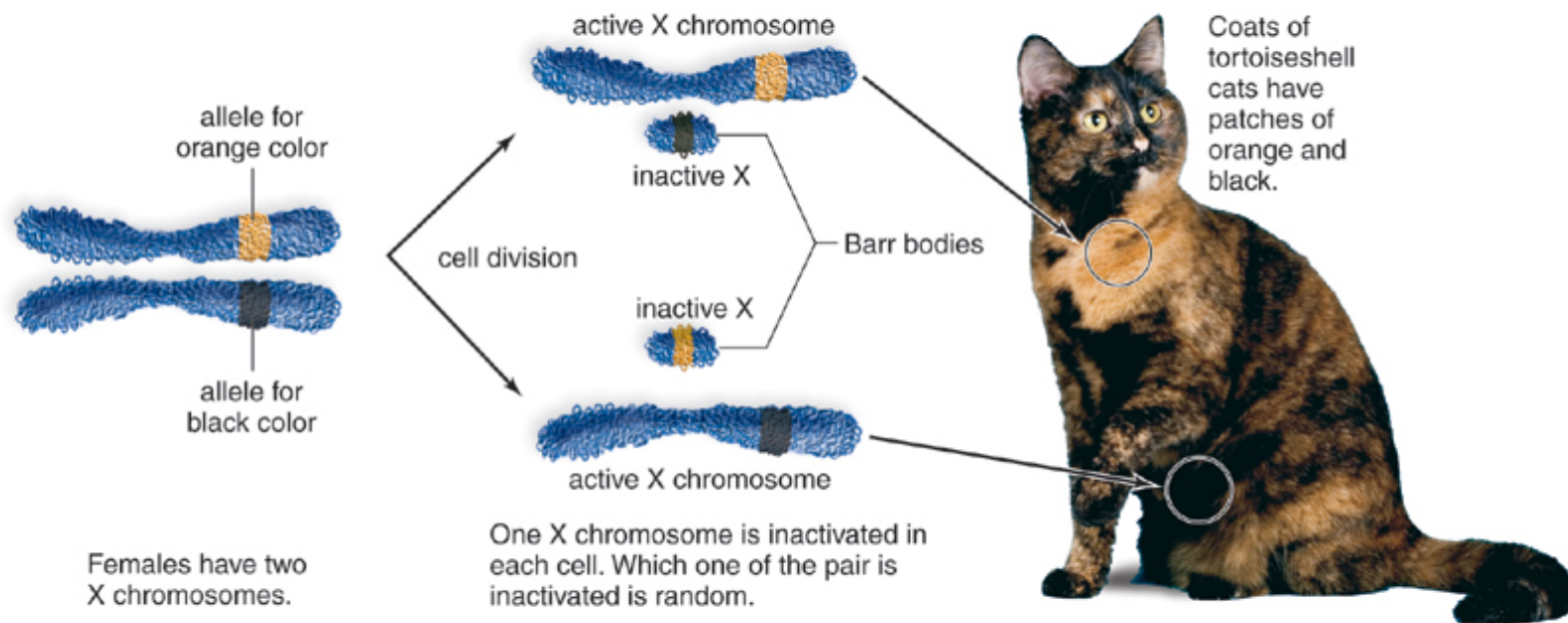
Women and men have the same amount of G6PD
because women and men have only 1 **active** X chromosome.

INACTIVATION of 1 X

(compensatory mechanism)



X-inactivation



© Chanan Photo 2004

Early embryo:

X chromosomes

**Allele for
orange fur**

**Allele for
black fur**

**Cell division and
X chromosome
inactivation**

**Two cell
populations
in adult cat:**

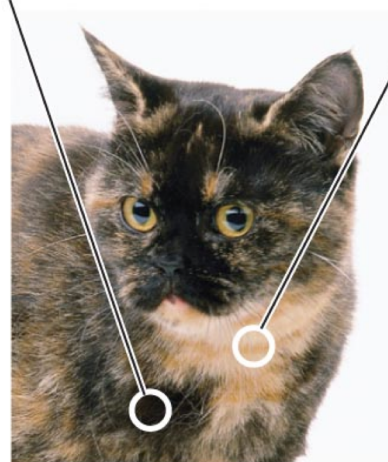
Active X

Inactive X

Active X

Black fur

Orange fur



Difference between calico and tortoiseshell

(a)



(b)



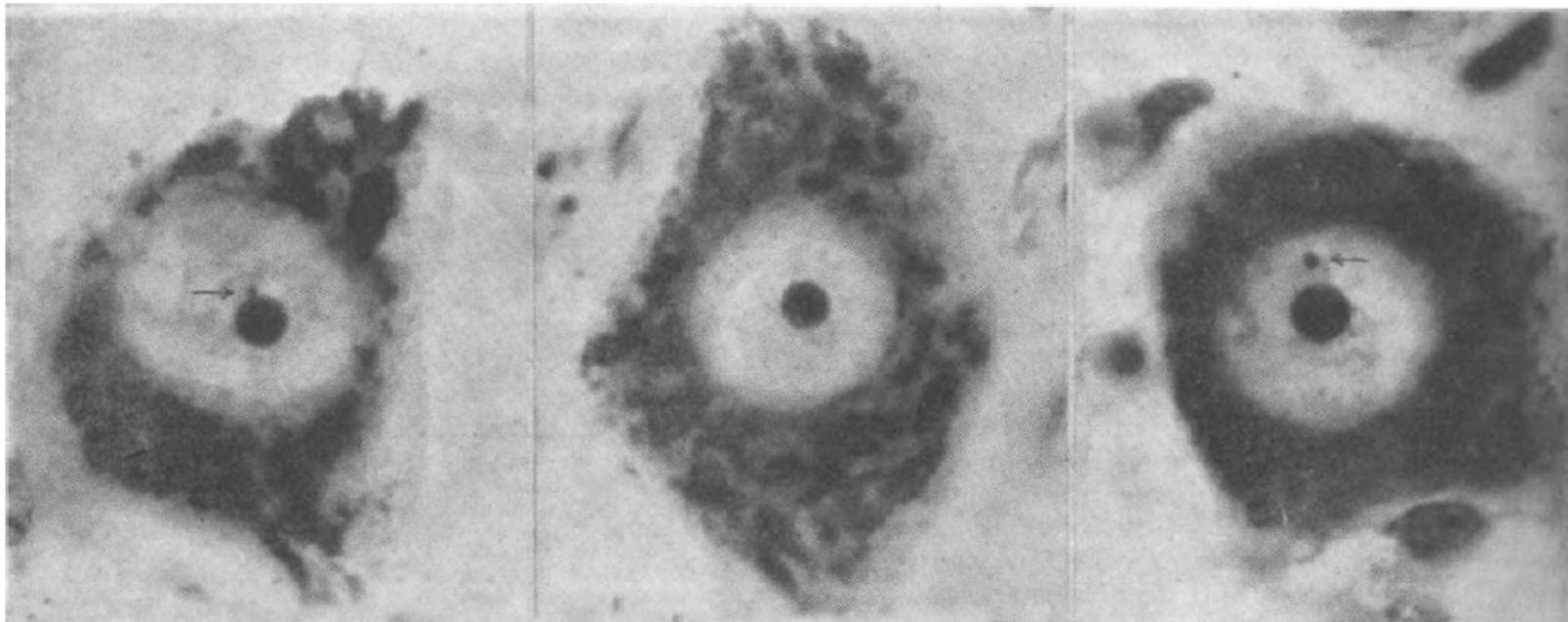
There is no melanocyte



Ewart BERTRAM

A Morphological Distinction between Neurones of the Male and Female, and the Behaviour of the Nucleolar Satellite during Accelerated Nucleoprotein Synthesis

Murray BARR
(1908-1995)



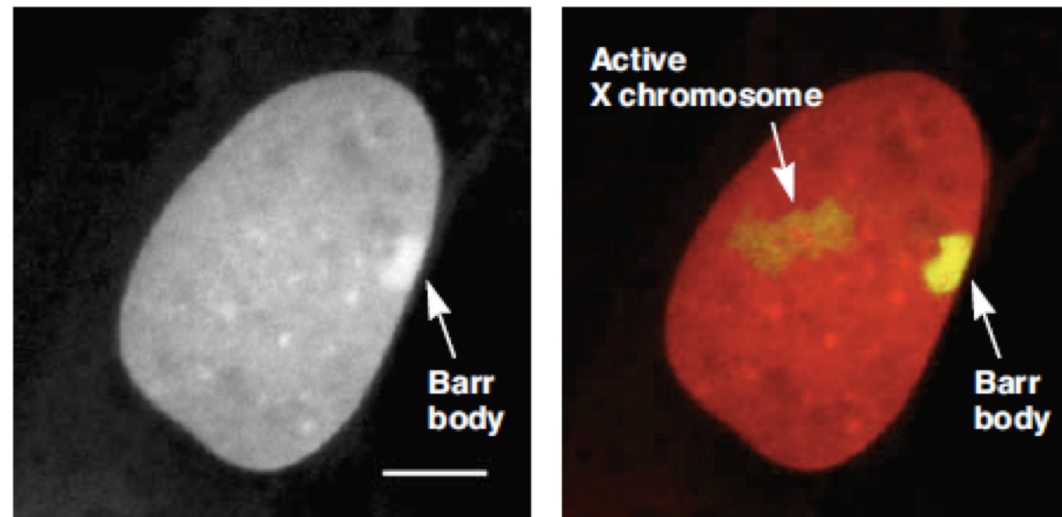
XX Fig. 1

XY Fig. 2

Fig. 3 XX

In 1949, Murray Barr and Ewart Bertram identified a **highly condensed structure** in the interphase nuclei of somatic cells in female cats that was not found in male cats.

This structure became known as the **Barr body**.



(a) Nucleus with a Barr body

In 1960, Susumu Ohno correctly proposed that the Barr body is a highly condensed X chromosome.

Barr body

(inactivated X chromosome)

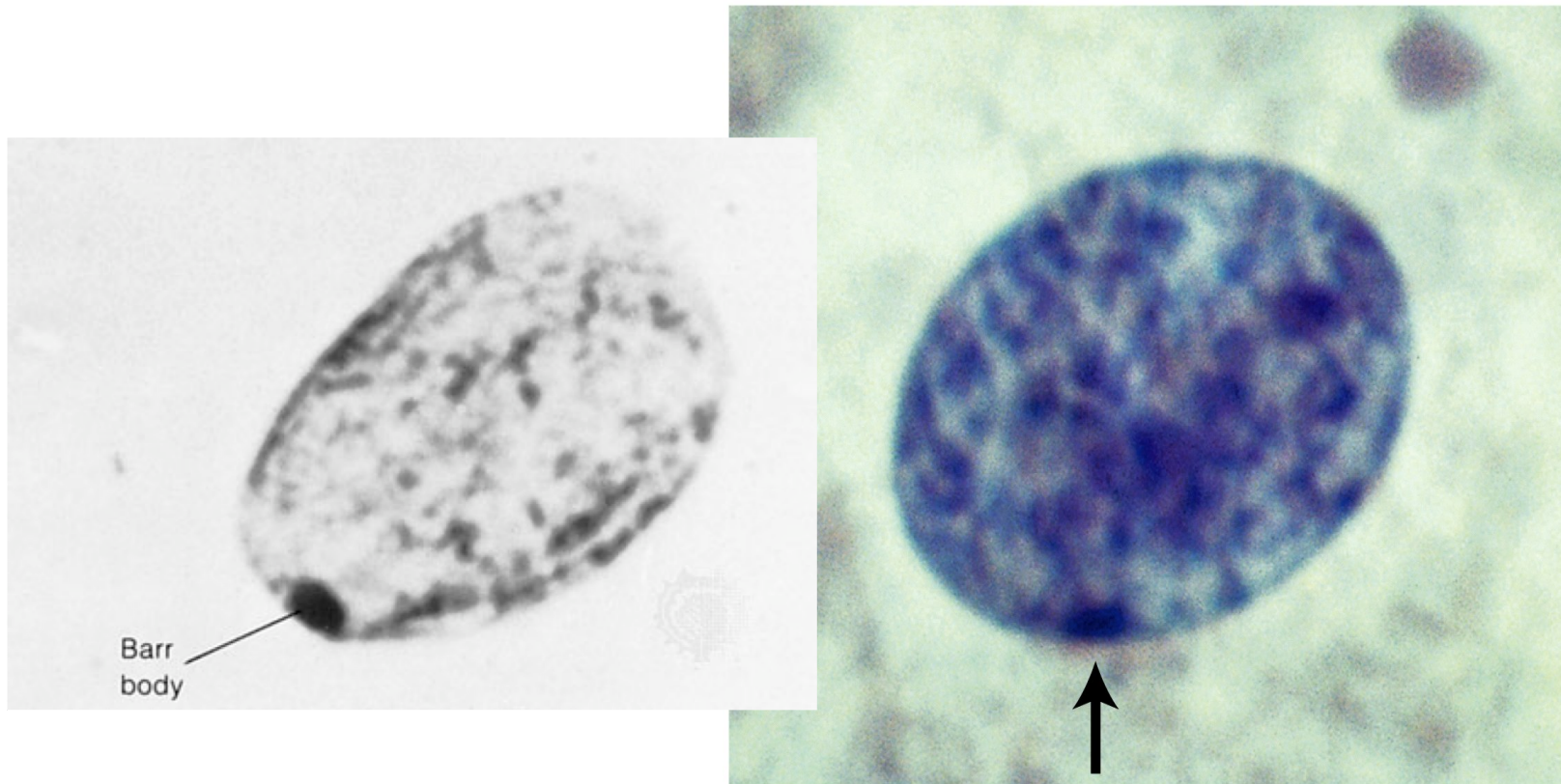
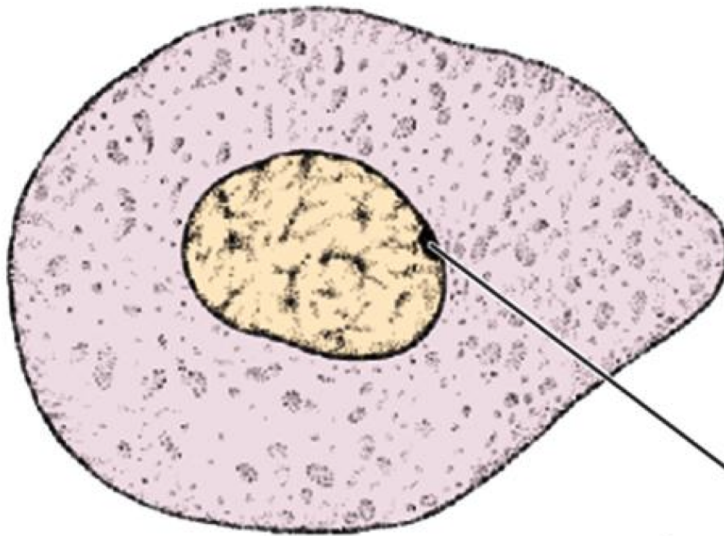


Figure 4-16a
Genetics: A Conceptual Approach, Third Edition
© 2009 W. H. Freeman and Company



Barr body

Buccal epithelium



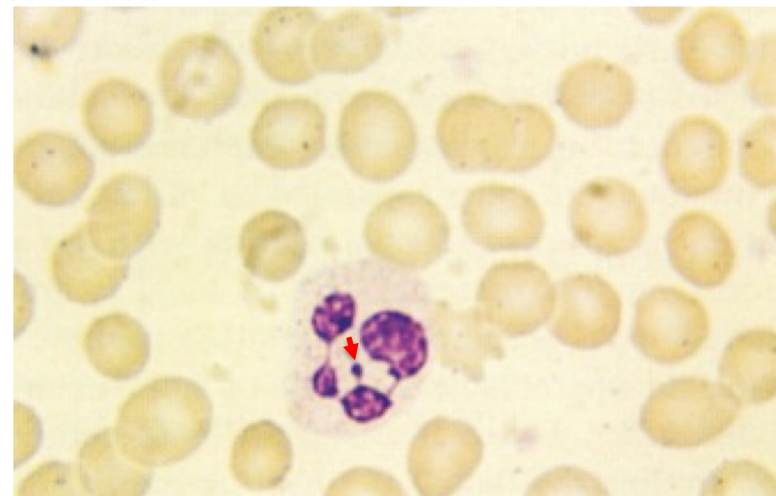
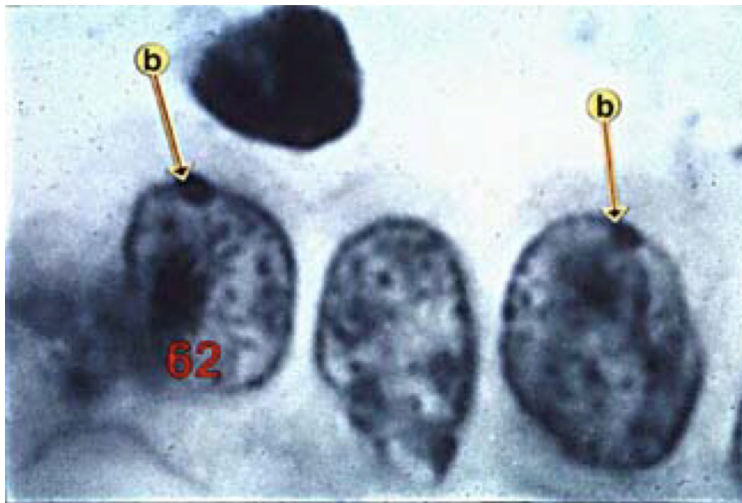
Polymorphonuclear leukocyte



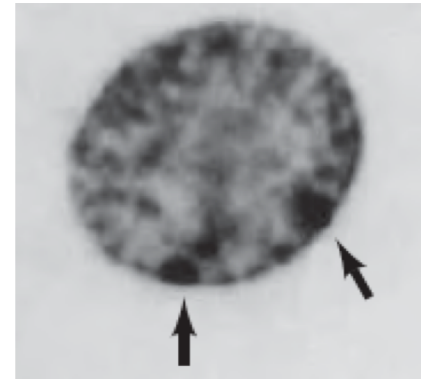
Blood sample

drumstick

Sex chromatin



Triple X (47, XXX)



2 Barr bodies
(2 inactivated X)

One lady with this anomaly was **Ewa Klobukowska**, a 1964 Olympic sprint bronze medallist who was the first woman to fail the sex chromosome test during the 1967 European Cup.

“One chromosome too many to be declared a woman for the purposes of athletic competition” was the statement, and she was obliged to return all her medals. She gave birth to a baby the following year.

➞ 47,XXX women have normal children !!

Feminity test : (competition)

Normal woman :



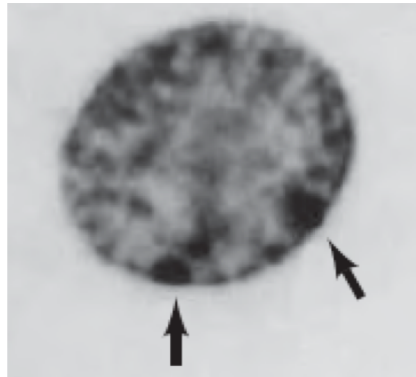
Barr body
= inactivated X chromosome

46, **X** X



47, **X** **X** X

Ewa Klobukowska
Bérénice



Official document attesting gender.

Complete androgen insensitivity

Certificat de féminité pour les sportives :

Miss/Mrs. PATINO CERTIFICATE No. 5341

First name /Prénomial Maria José Martínez

at/de Spain (Country/Pays)

born/née (date)

On the occasion of the/A l'occasion des
World Championships
(Games de Championships/
Jeux au Championnat)

held at/à Helsinki on le 1983
the above mentioned athlete underwent an
approved medical test. The result of which
was sex chromatin positive. This satisfies
the I.A.A.F. requirements for competition in
Women's athletic events.

L'athlète mentionnée ci-dessus a subi un
examen médical approuvé, et la chromatine
sexuelle s'est révélée positive. Ceci répond
aux règlements de la F.I.A.A. pour concourir
aux preuves féminines.

[Signature]
Athlete's signature/Signature de l'athlète

[Signature]
General Secretary, I.A.A.F.
Secrétaire Général, F.I.A.A.

I hereby confirm that this Certificate is
issued in accordance with the report of
this official Medical Panel of the
mentioned Games/Championnats.

Je certifie par la présente que ce
certificat est délivré conformément au
rapport du Comité médical officiel des
Jeux/Championnats mentionnés ci-contre.

46,XY female

L'athlète mentionnée ci-dessus
a subi un examen médical approuvé
et la **chromatine sexuelle** s'est
révélée positive.



Maria PATINO

Tested female in 1983 in Helsinki ;
failed the gender test in 1985

Santhi Soundarajan

Doha
2006

46,XY female



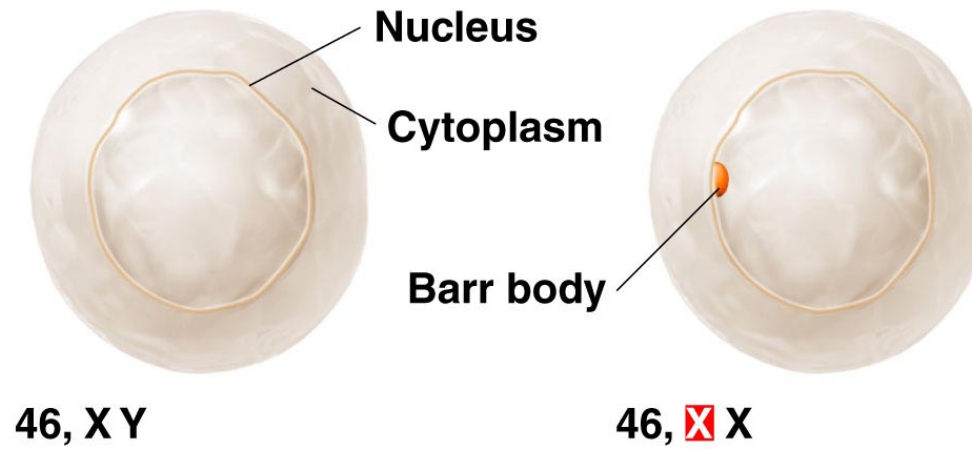
She had to give back her silver medal (800 m).

Is it fair ???

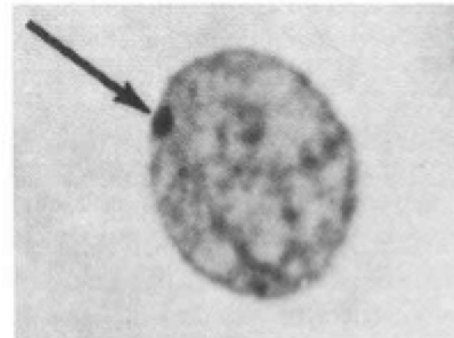


Gender test (competitions)

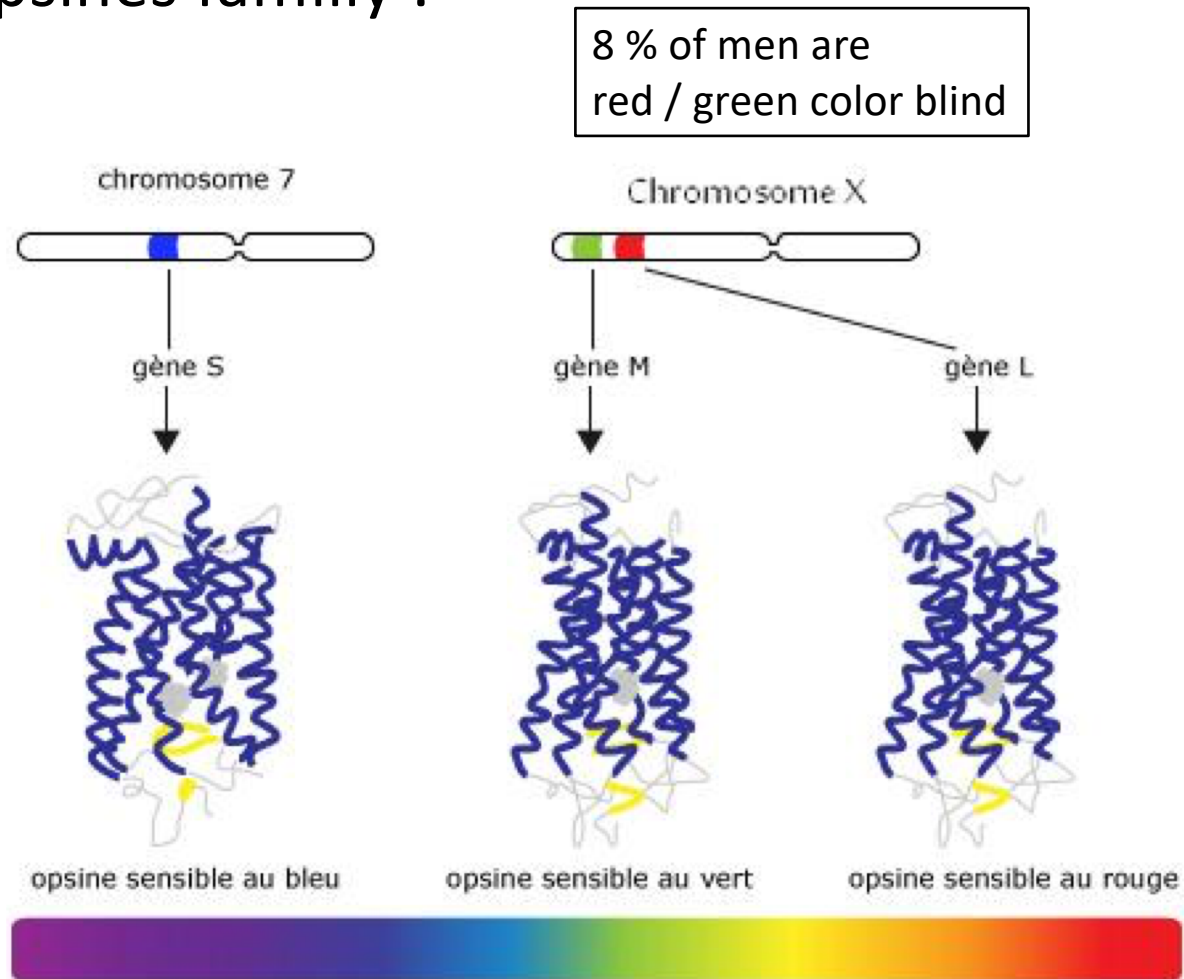
Normal woman :



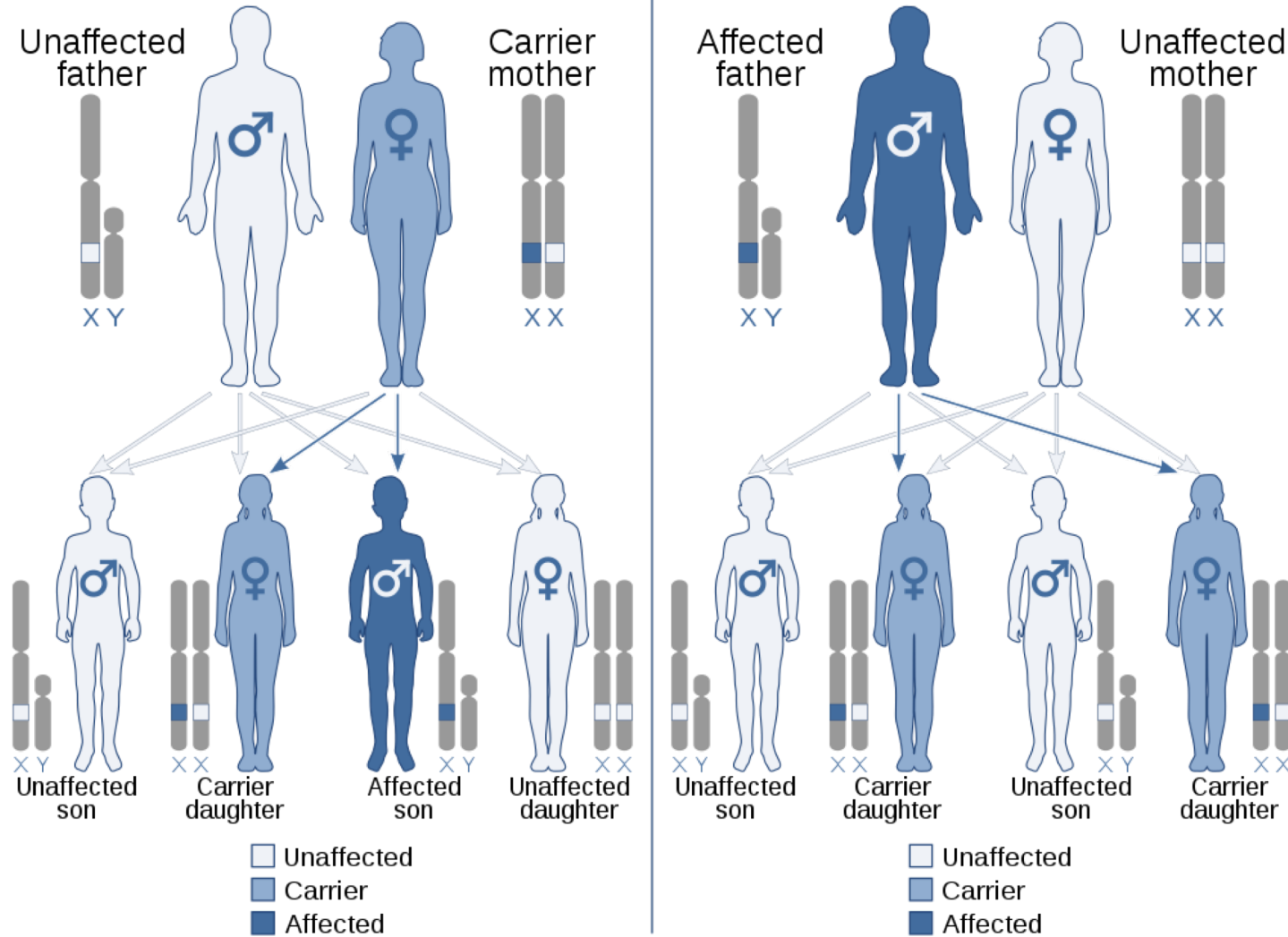
No Barr body



The opsines family :



X-linked recessive

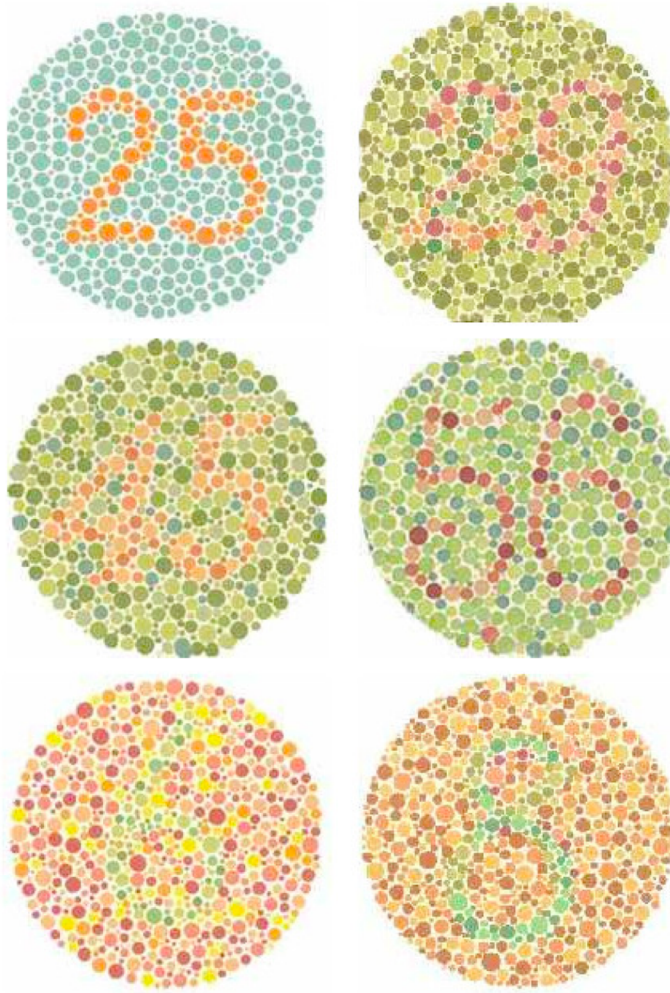


Recessivity is defined only in women.

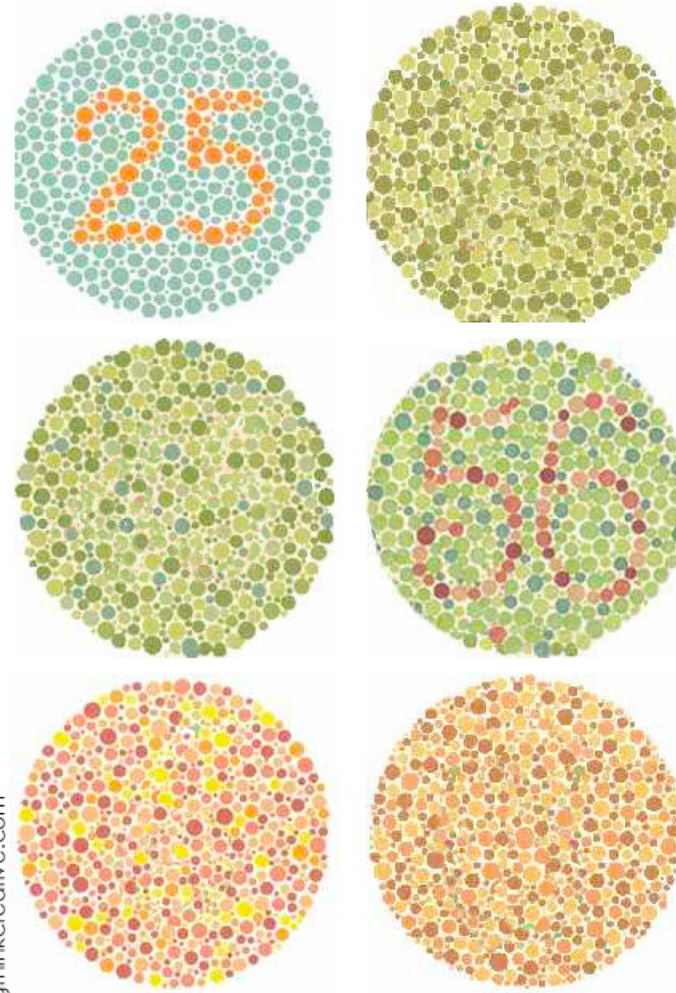
Note: a few carriers may be mildly affected due to skewed X-inactivation.

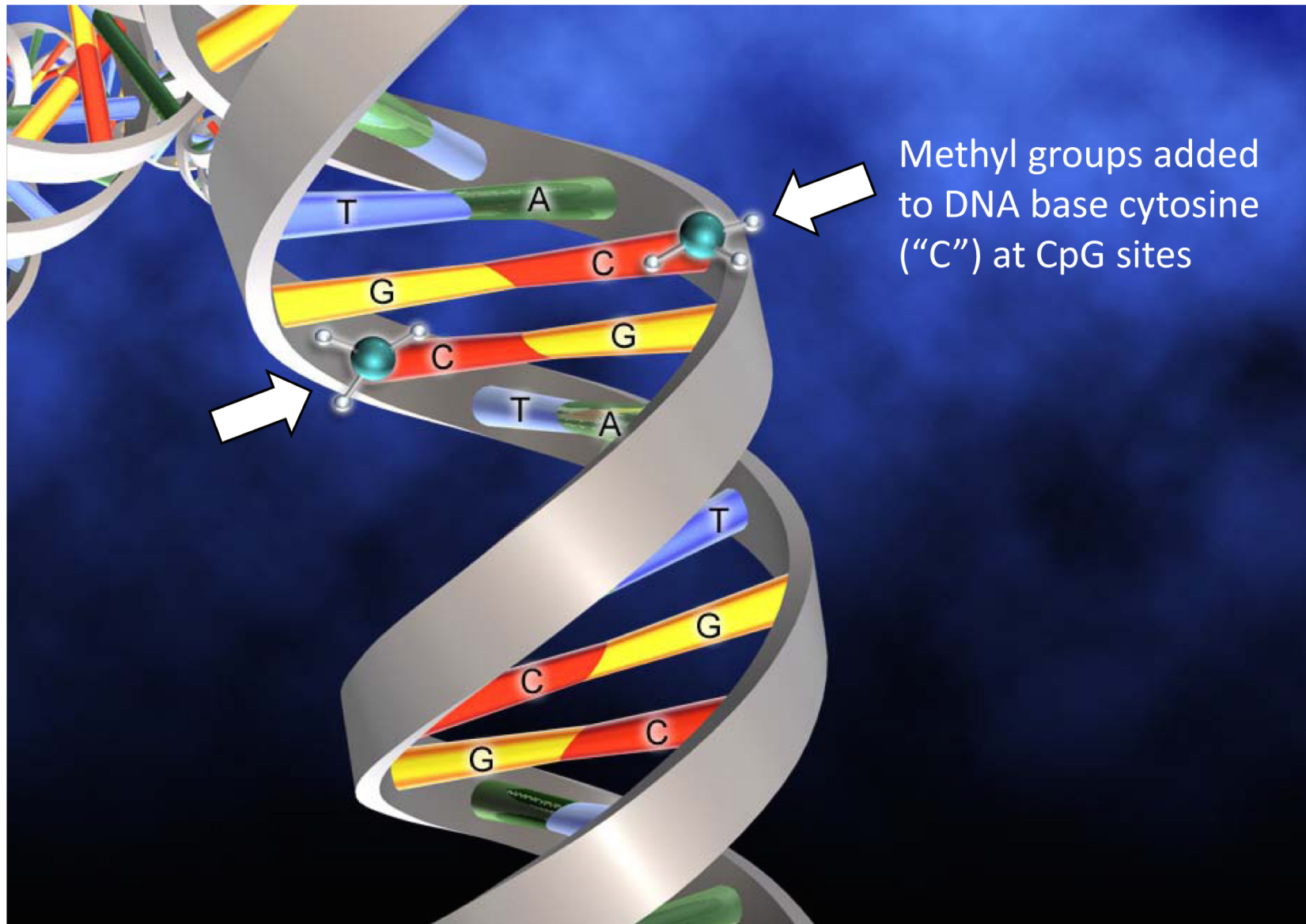
Ishihara Test For Color Blindness

What People With Regular Vision See

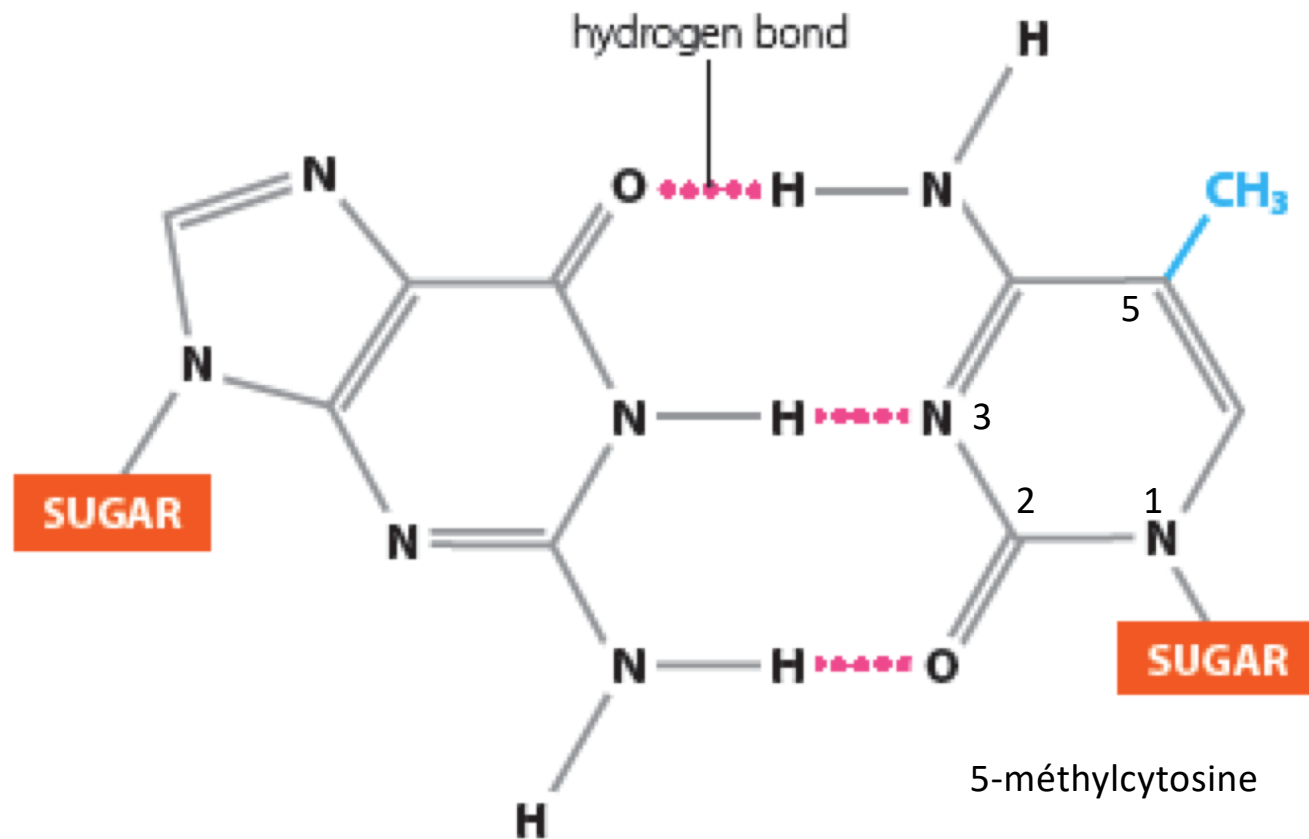


What Red-Green Color Blind People See



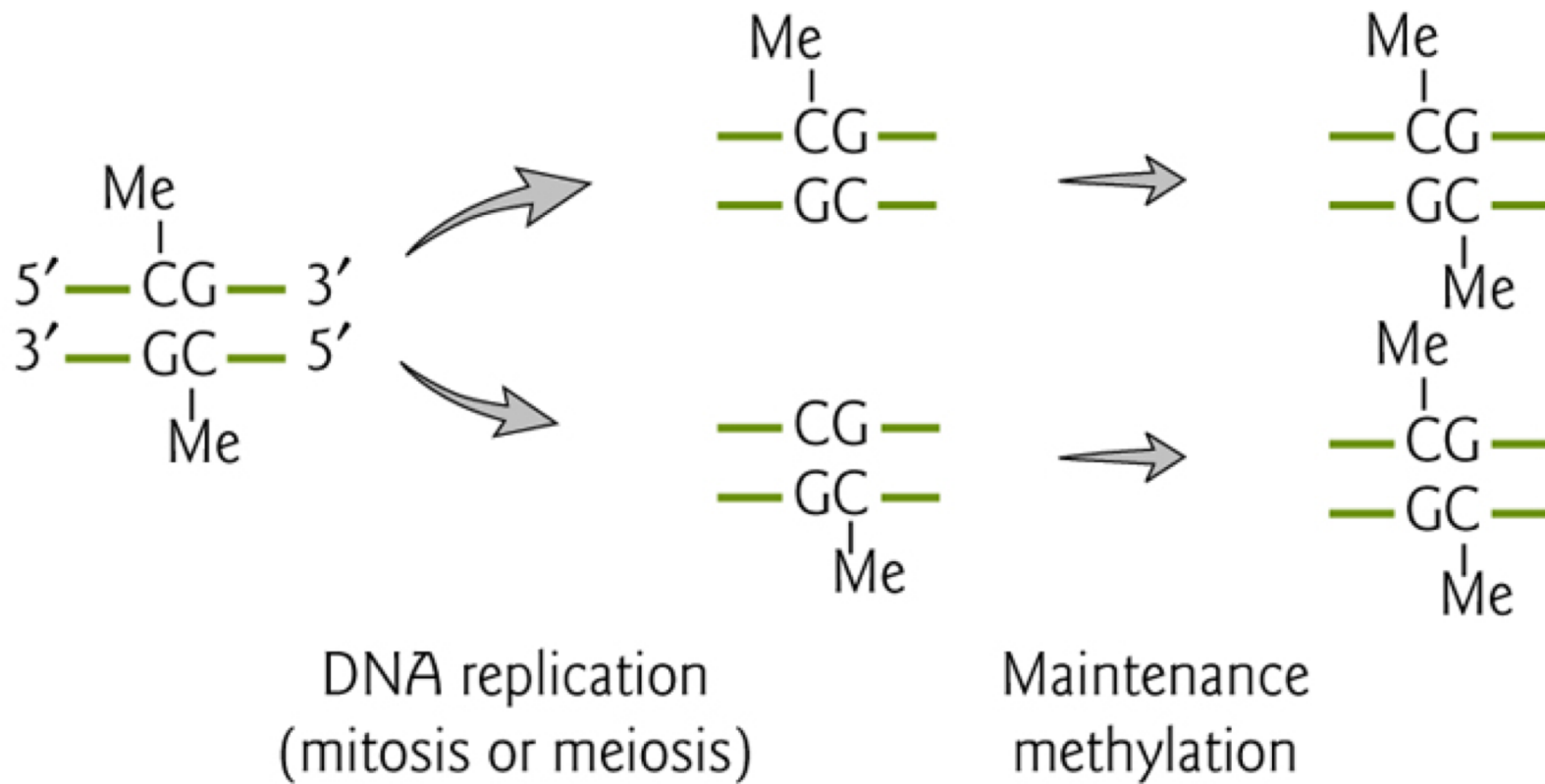


L'appariement n'est pas affecté par la méthylation :

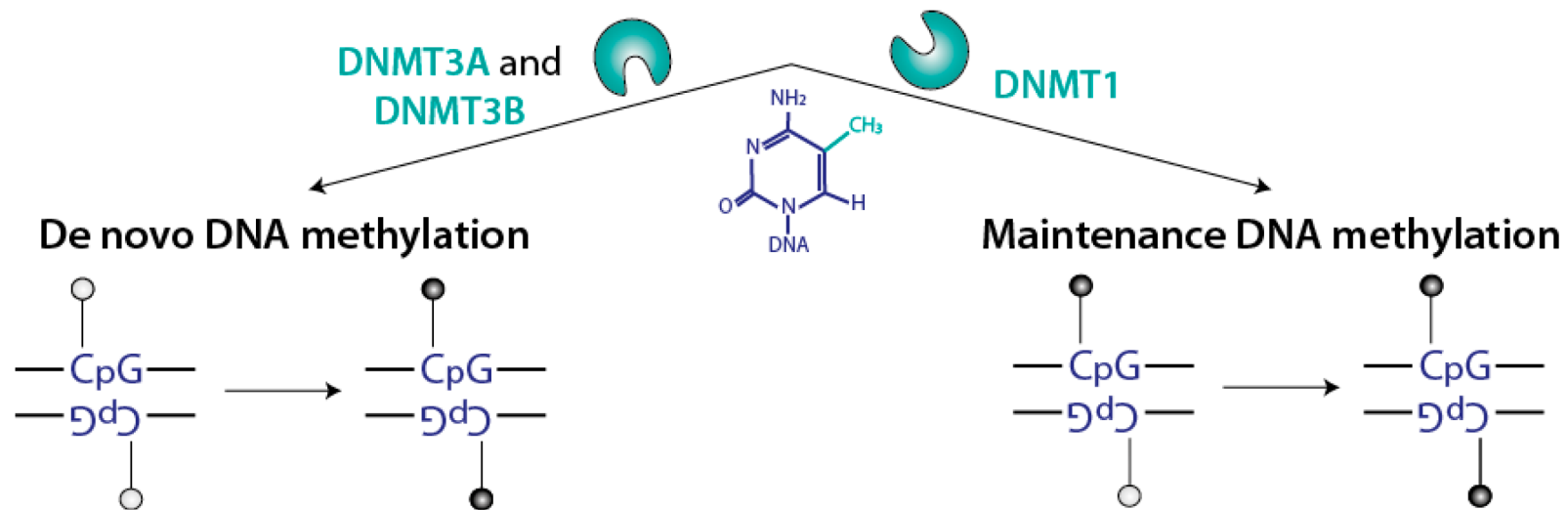


5-methylcytosine base-pairs with guanine in exactly the same way as unmodified cytosine.

CpG Methylation

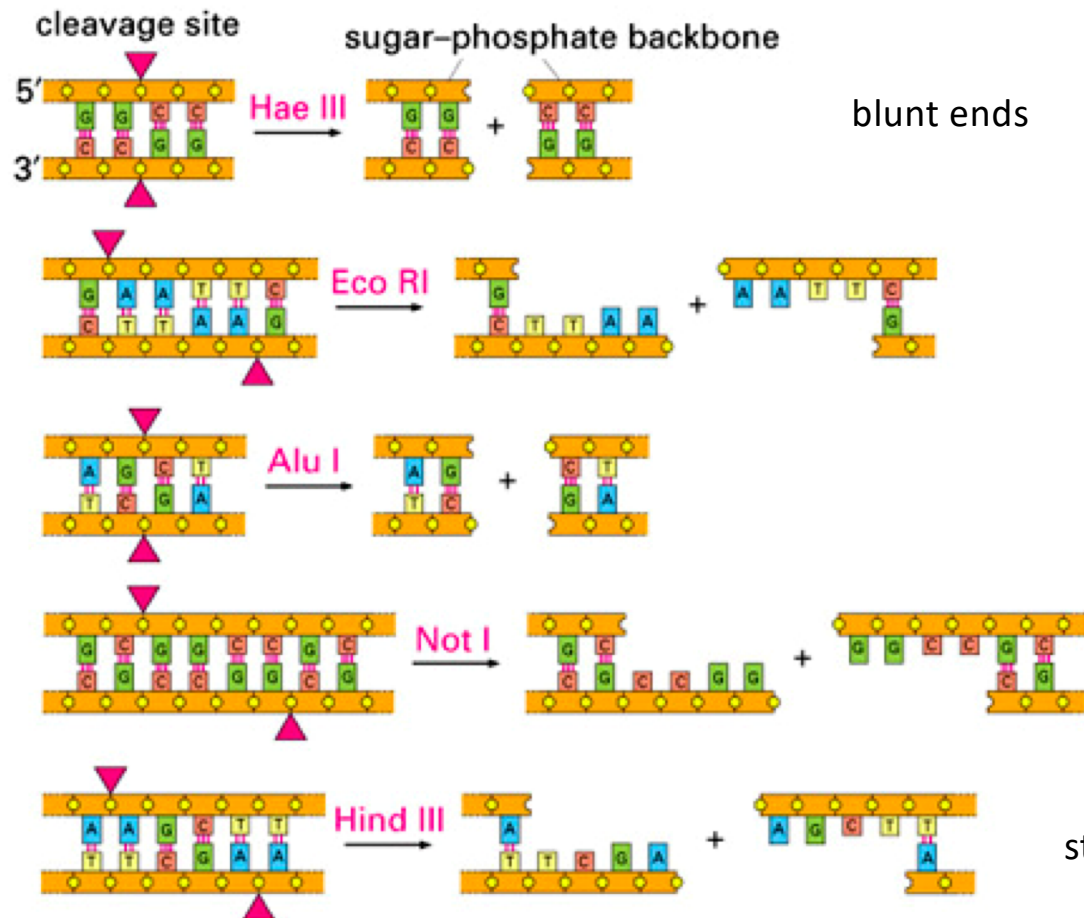


CpG Methylation



DNMT = **D**Na **M**ethyl **T**ransferase

DNA can be cut by restriction nucleases



- cut double-stranded DNA
- at particular sites: recognition sites
- highly specific sequences of
4 nucleotides: frequent cutters
6 nucleotides: medium cutters
8 nucleotides: rare cutters
- from bacteria; defense mechanism against phages

Isoschizomers

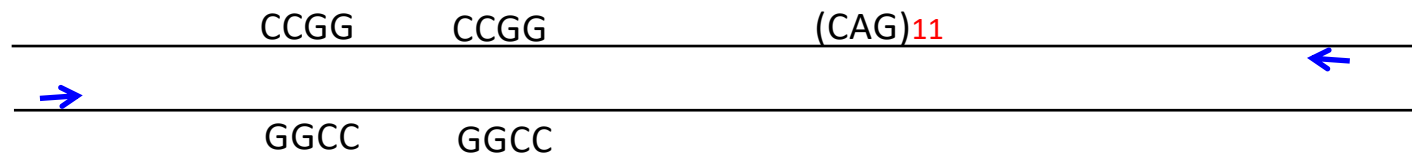
	cut	no cut
Hpa II recognizes	5'- C C G G - 3' 3'- G G C C - 5'	5'- C ^m C G G - 3' 3'- G G ^m C C - 5'
Msp I recognizes	5'- C C G G - 3' 3'- G G C C - 5'	5'- C ^m C G G - 3' 3'- G G ^m C C - 5'
	cut	cut

Hpa II and Msp I recognize the same restriction site : they are isoschizomers.

However there is one important difference :

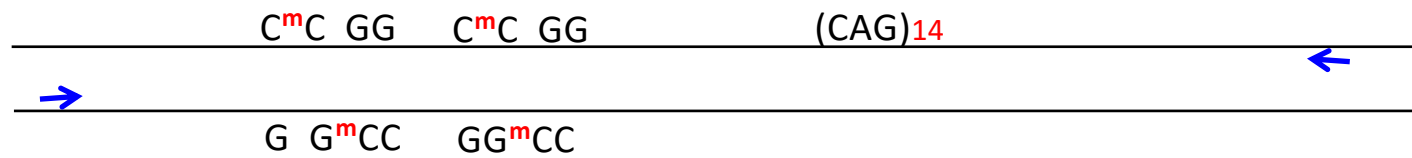
Hpa II is **sensitive** to DNA methylation

Msp I is **insensitive** to DNA methylation



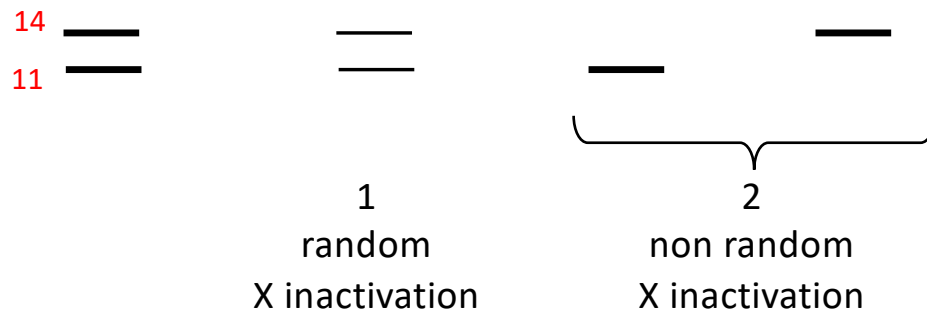
The active X
is not methylated

microsatellite

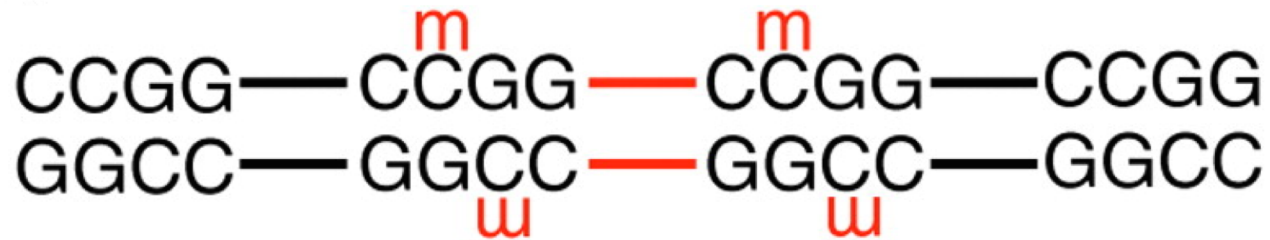
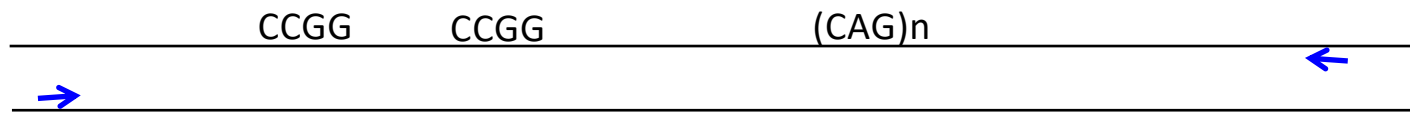


The inactive X
is methylated

HUMARA test (PCR)



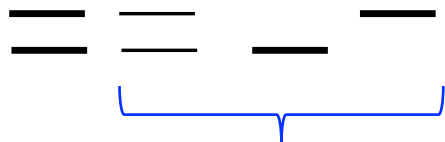
Only the DNA from the **inactive X**
can be amplified by PRC.



Digestion with *HpaII*
 (blocked by CG
 methylation)

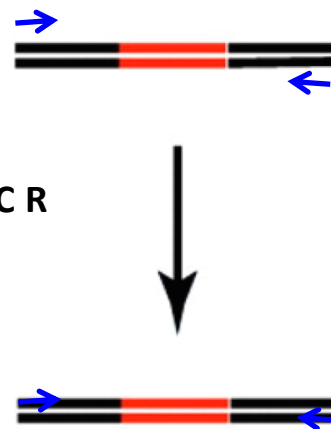
Digestion with *MspI*
 (not blocked by CG
 methylation)

HUMARA test



after HpaII digestion

P C R



No amplification product