

CHROMOSOMES AND DISEASE

Date: September 29, 2005 *
Time: 8:00 am- 8:50 am *
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Office Hours: by appointment

***Please consult the online schedule for this course for the definitive date and time for this lecture.**

The syllabus is meant to accompany the lectures and the material covered (other than that in italics) is considered important to your understanding of the subject.

Lecture Objective

Chromosome abnormalities are defined as changes resulting in visible alteration of the chromosomes. This lecture will focus on how we analyze a patient's chromosomal constitution and two broad categories of chromosomal abnormalities: numerical (triploidy, trisomy and monosomy) and structural (deletion, duplications and others) aberrations. Details of chromosome pairing and mis-pairing will be illustrated for your general appreciation of the process. You will not be expected to remember these details but rather the broad clinical implications of different chromosomal aberrations. We will also discuss prenatal diagnosis of chromosomal abnormalities and will touch on cancer cytogenetics.

Chromosomes and Disease

WHAT ARE CHROMOSOMAL DISORDERS?

A chromosomal disorder occurs when there is a change in the number or structure of the chromosomes. This change in the amount, or arrangement of, the genetic information in the cells may result in problems in growth, development and/or functioning of the body systems. The chromosomal abnormalities may occur either during the production of the egg or sperm or early after the baby's conception: a spontaneous occurrence for unknown reasons. Chromosomal abnormalities may also be inherited from a parent. There are two main types of chromosomal disorders: changes in chromosome **number** and changes in chromosome **structure**

Chromosome disorders: major category of human disease, represent 1% of live births, *2% of pregnancies in women older than 35, 50% of all spontaneous first trimester abortions, responsible for more than 100 human syndromes* and are more common than all Mendelian single-gene disorders

Studying Human Chromosomes

Clinical Cytogenetics: study of chromosomes, their structure and inheritance

Karyotype: chromosome constitution (46,XX or 46,XY)

Mitotic chromosomes can be seen in any dividing cell but most conveniently in lymphocytes. Meiotic chromosomes are difficult to study in humans

Chromosomes are identified by their size, position of the centromere and their banding pattern.

Chromosome nomenclature: short arms are labeled "p" (petit); long arms are labeled "q" (queue); **proximal** = segment of the chromosome arm that is closest to the centromere; **distal** = portion of chromosome arm that is most distant from the centromere (closest to telomere)

Each chromosome is divided into **regions**, labeled p1,p2,p3.... and q1,q2,q3.... counting outwards from the centromere. Regions are divided into **bands** and **sub-bands** labeled p11.1, 012.3, p13.5) (read as one-one.one, not eleven.one)

Centromere is designated '**cen**' and telomere '**ter**'

Studying Human Chromosomes

Chromosome identification is by size, centromere position and banding pattern

Chromosome nomenclature:

-proximal

-distal

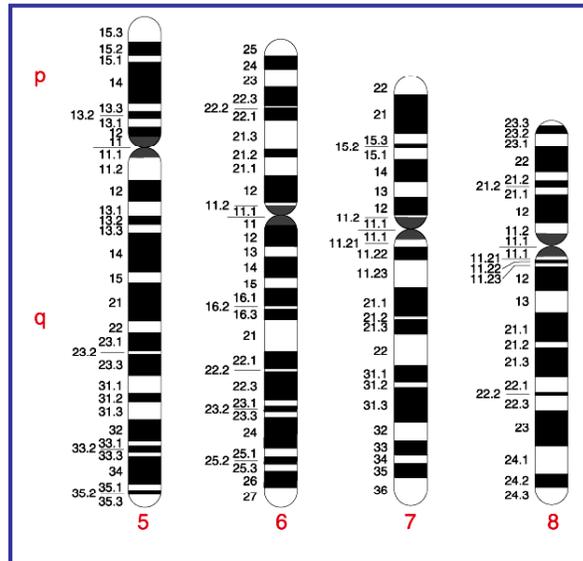
“p” (petit)

“q” (queue)

-regions are divided into bands

& sub-bands labeled

p11.1, p12.3, p13.5 etc



Chromosome Banding

G-banding- chromosomes are subjected to digestion with trypsin, then stained with Giemsa (A chemical dye that dyes DNA). Dark bands are known as **G bands** (condensed chromatin), pale bands are **G negative** (gene rich)

Chromosomal banding can detect structural changes in the chromosome on the order of 1-10 Mb scale

Molecular Cytogenetics Techniques

Can be used to identify chromosomal rearrangements or to diagnose chromosome number

- 1. Fluorescence In situ Hybridization (FISH):** Gene-specific probes to examine presence/absence of particular DNA sequence
- 2. Chromosome Paint:** mixture of single-copy DNA sequences that are specific for a chromosome and hybridize along the length of the chromosome
- 3. Spectral Karyotyping (SKY):** 24 individual chromosome painting probes labeled with different fluorescent dyes and used as a total genome chromosome

paint. Fluorescent signals are then analyzed by imaging software. The computer assigns a different color to each of the 24 different fluorescent spectra generated by the individual chromosome painting probes. The approach allows for a dramatic evaluation of the karyotype in a single experiment.

Types of Chromosomal Abnormalities

A chromosomal abnormality can be a numerical deviation from the diploid number (46, XX or 46, XY) or a structural rearrangement such as an **inversion**, **translocation** or **deletion**. The abnormalities may involve one or more than one autosome, sex chromosomes or both.

Most chromosomal aberrations are produced by **misrepair** of broken chromosomes, **improper recombination** during meiosis/mitosis (unequal crossing over) or **mal-segregation** of chromosomes during meiosis/mitosis

If a chromosome abnormality is present in all cells of the body it is called a **constitutional abnormality**, if it is present in only certain cells or tissues it is called a **somatic or acquired abnormality**. The latter can lead to a **mosaic** individual (one who possesses two or more genetically different cell lines derived from a single zygote).

-Numerical abnormalities: There can be extra copies of the autosomes or the sex chromosomes. **Aneuploidy** represents an abnormal chromosomal number due to an extra (**trisomy**) or missing (**monosomy**) chromosome.

Extra copy of a numbered chromosome (an autosome): The most common example of a disorder seen in newborn babies that is due to an extra copy of an autosome is called Down syndrome. *In 1866, a physician named John Langdon Down who worked with people with intellectual disabilities, observed that a number of his patients were so similar in appearance that they might easily be mistaken for children of the same parents. In a classic description, he recorded that these individuals possessed a broad, flat face, a thick tongue and a small nose and were intellectually impaired to a variable degree. This was a time in history when many new lands and races were being discovered and described for the first time. Thus the name suggested for people with this specific disability was Mongolism.* Today the disorder is more appropriately referred to as Down syndrome or trisomy 21. The disorder results from an abnormality in the chromosome number in some, or all, of the cells of an individual, specifically involving chromosome number 21. Individuals with this disorder have 47 chromosomes in their cells instead of 46. This is because there are three copies of chromosome number 21 instead of the usual two. Therefore Down syndrome is also called trisomy 21 as trisomy means "3 bodies". Other relatively common abnormalities of autosomes include trisomy 13 (3 copies of chromosome number 13 instead of the usual 2) and trisomy 18 (3 copies of chromosome number 18). Babies born with these chromosomal disorders in all the cells of their body have

a range of severe disabilities and do not usually survive past infancy or early childhood.

Extra copy of a sex chromosome (an X or Y): Having extra copies of either the X or Y chromosomes (the sex chromosomes) may also cause problems. An example is Klinefelter syndrome, which is due to an extra X chromosome in a male and is described as 47,XXY. Even though there are 2 copies of the X chromosome, the presence of a Y chromosome makes a person a male, regardless of the number of X chromosomes. Other boys have 48 chromosomes where there are 3 copies of the X chromosome in addition to the Y chromosome (48, XXXY). Other chromosomal disorders include triple X syndrome (girls with three copies of the X chromosome -XXX) and boys who have two copies of the Y chromosome (XYY syndrome).

When there are fewer copies of particular chromosomes than usual: An example of this is seen with the sex chromosomes. For example, girls born with Turner syndrome have 45 chromosomes in their cells instead of 46. This is because there is only one copy of the X chromosome instead of the usual two copies. Cytogeneticists refer to this disorder as **monosomy X** ("monosomy" means "one body") and it is written as 45,X. In most cases, the loss of a whole chromosome is incompatible with life and will result in miscarriage or stillbirth.

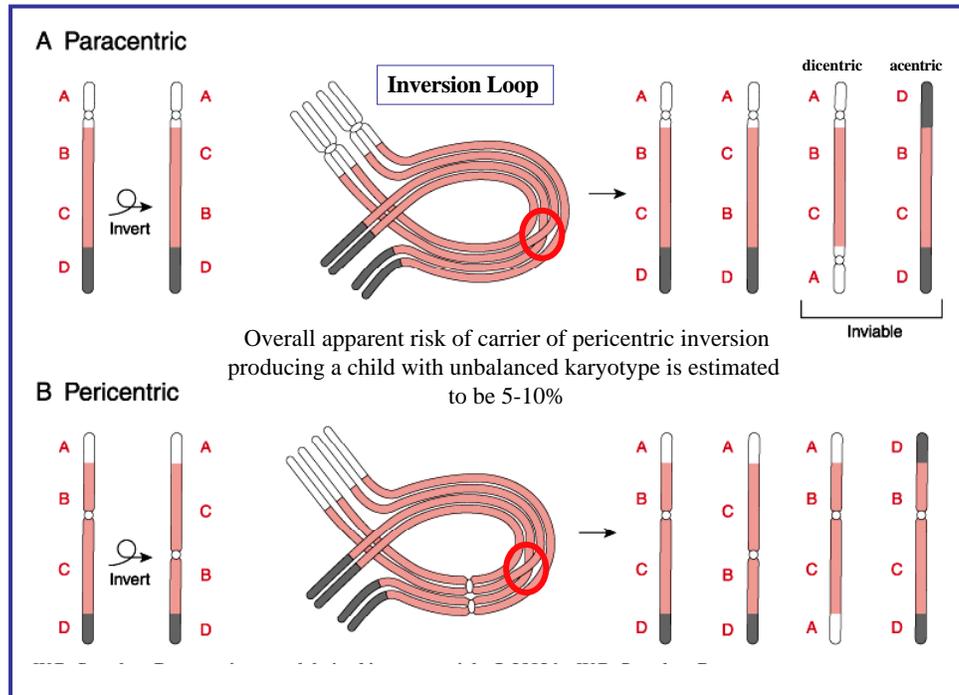
Euploid: exact multiple of the haploid chromosome number (1n,2n,3n,4n etc..).
Triploid (3n) usually results from fertilization by more than one sperm. These individuals can survive to term but do not survive. **Tetraploid** usually results from failure of completion of a zygotic cleavage. These fetuses do not survive.

Structural Rearrangements

Result from chromosome breakage, followed by reconstitution in an abnormal combination. The frequency is high (1/375 newborns).

A **balanced rearrangement** will still have a normal complement of chromosomal material and such a rearrangement often does not have a phenotypic effect because all of the chromosomal material is present even though it is rearranged. However, it is possible that the rearrangement breaks or disrupts a gene, leading to a mutation.

An example of such a rearrangement is an **inversion**. This is where a segment between two chromosomal breaks invert. A **pericentric inversion** includes the centromere and a **paracentric inversion** does not include the centromere. However carriers of these types of rearrangements have a high frequency of unbalanced gametes resulting in an increased risk of producing abnormal offspring.



The above table should be considered supplementary and is presented for those who wish to envision the process of inversions in more detail

Translocations (trans = across; location = place) is the term used to describe a rearrangement of chromosome material involving two or more chromosomes. The most common type of chromosome translocation is called a reciprocal translocation because material is swapped between two chromosomes. Thus, translocations are another structural rearrangement that result from an exchange of chromosome segments between two (usually nonhomologous) chromosomes.

There are two main types of translocations. **Reciprocal translocations** represent breakage of nonhomologous chromosomes with exchange of the broken parts. Usually only two chromosomes are involved and the exchange is reciprocal so it is balanced. These are seen at a frequency of about 1/600 newborns. **What is the result of having a balanced translocation?** A "balanced" reciprocal translocation usually means that the person has the correct amount of genetic information for normal development. This is particularly so where a parent has the same translocation as their child and is unaffected. However, when a child is the only person in a family to have such a rearrangement, then it is not always possible to say that there will be no problem. A breakpoint may occur in an important gene whose function will be disrupted as a result of the break. In this case, there may well be symptoms or a problem for the person concerned. When a person carries an apparently "balanced" reciprocal translocation, there is an increased probability that there will be reproductive consequences. The nature of these consequences depends on the particular chromosomes involved and the size of the translocated material.

Although the translocations themselves are usually harmless, they are associated with a high risk of unbalanced gametes that can produce abnormal progeny depending on how the chromosomes segregate during meiosis. The translocations produce a **quadrivalent** at meiosis and then anaphase segregation can occur in one of three ways: **alternate** (produces balanced gametes), **adjacent 1** (homologous centromeres go to separate daughter cells and produce unbalanced gametes) and **adjacent 2** segregation (homologous centromeres go to the same daughter cells and produce unbalanced gametes).

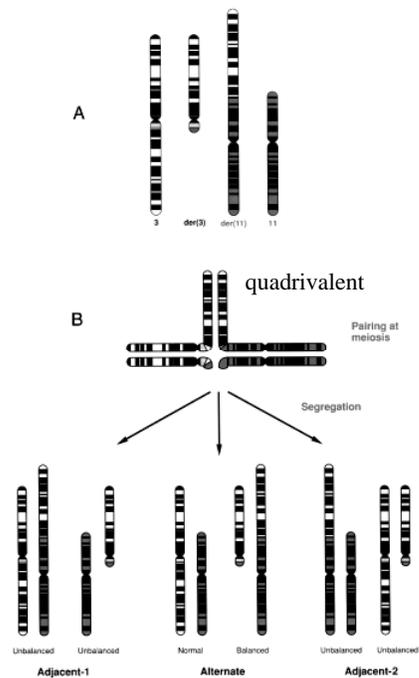
The diagram below should be considered supplementary and is presented for those who wish to envision the process of inversions in more detail

-translocation associated with high risk of unbalanced gametes

Pairing at meiosis forms **quadrivalent**

-anaphase segregation occurs in one of three ways

alternate (balanced)



Robertsonian Translocations

These only involve chromosomes numbers 13, 14, 15, 21 and 22. These chromosomes are different from the other chromosomes as their centromeres lie very near the tip of the chromosome, giving a chromosome with a long arm and a very tiny short arm. Translocation of these chromosomes involves loss of the short arms and fusion of two chromosomes at the centromere, resulting in one chromosome which consists of two long arms of either the same numbered chromosome or two different chromosomes and containing either one or both centromeres. There is a loss of the short arm material which seems to have little or no effect on the individual involved. Robertsonian translocations are relatively common and are found in about 1 in 1000 people in the general community. Balanced Robertsonian translocations generally do not result in physical or developmental problems for the carrier. However, there are reproductive risks for carriers of these translocations also.

HOW DO CHROMOSOME TRANSLOCATIONS OCCUR?

Autosomal reciprocal translocations are nearly always spontaneous, i.e. they occur at the time of conception and neither parent has a similar chromosome pattern. However, in some cases, one parent may have a balanced translocation themselves which can be passed to a child. *Robertsonian translocations* between the same chromosome pair are very rare and almost always occur as a spontaneous or one-off event, with no one else in the family having a similar chromosomal arrangement. Translocations between two different chromosomes are much more common and can be passed on through the generations as either balanced or unbalanced rearrangements. The most common one in humans is a fusion of 13q to 14q (1/1300 individuals).

Deletions

These abnormalities are due to loss of a chromosome segment that occur at a frequency of about 1/7,000 newborns. They can be terminal or interstitial deletions that result from breakage followed by fusion or they can result from unequal crossing over. Deletions can lead to **haploinsufficiency**, which is defined as the inability of a single copy of a gene to carry out the functions normally performed by two copies.

Examples of autosomal deletions syndromes:

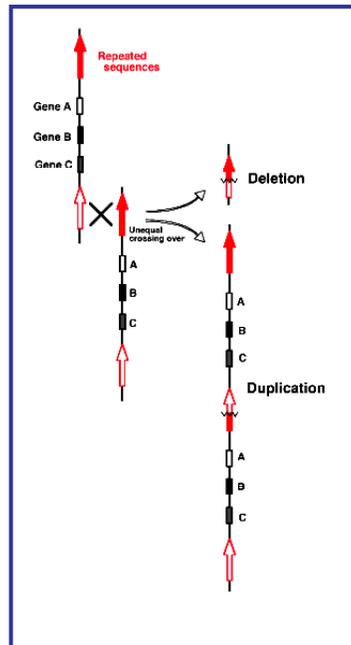
-contiguous gene syndrome: haploinsufficiency of multiple, contiguous genes within a deleted region

-Cri du chat syndrome: a large deletion of the short arm of chromosome 5. *This deletion accounts for 1% of all institutionalized mentally retarded patients. Their facial appearance is distinctive with microcephally, hypertelorism, epicanthal folds, low set ears, micrognathia, mental retardation and heart defects. The critical region has been defined as 5p15 with many of the responsible genes being located in 5p15.2*

-microdeletion syndromes: see that the size of the deletion is similar in many different patients. It turns out that the breakpoints are located in clusters of low-repeat DNA sequences. DiGeorge syndrome (craniofacial abnormalities, mental retardation, heart defects) is one of the more common deletions occurring in about 1/2000 to 1/4000 births.

-unequal crossing over: This can occur between misaligned sister chromatids that contain homologous copies of repeated DNA sequence. When this occurs it can lead to deletions and duplications

Unequal crossing over between misaligned sister chromatids



The above diagram should be considered supplementary and is presented for those who wish to envision the process of unequal crossing over and its results in more detail

Ring Chromosomes

These chromosomes can form when a normal chromosome undergoes two breaks and then the broken ends fuse to produce a ring structure. These chromosomes are rare and they produce mitotic instability. The two sister chromatids become tangled in an attempt to disjoin at anaphase. This results in more breakage and fusions.

Isochromosomes

These chromosomes have one arm missing and the other arm duplicated in a mirror image fashion. The result is monosomy for the missing arm and trisomy for the duplicated arm. These types of chromosomes are frequently seen in tumors.

You are not responsible for a detailed knowledge of nomenclature. It is reasonable for you to know that “del” means deletion, “inv” means inversion, etc.

Structural Abnormalities: Nomenclature

Deletion	46, XX, del(4)(p16.3) = terminal deletion with breakpoint at 4p16.3 46, XX, del(5)(q13q33) = interstitial deletion between 5q13 to 5q33
Inversion	46, XY, inv(11)(p11p15) = inversion between 11p11 to 11p15
Duplication	46,XX, dup(1)(q22q25) = duplication of 1q22 to 1q25
Insertion	46,XX, ins(2)(p13q21q31) = a rearrangement of one copy of chromosome 2 by insertion of segment 2q21-q31 into a breakpoint a 2p13
Ring	46XY, r(7)(p22q36) = ring chromosome from 7p22 to 7q36
Marker	47, XX, +mar = cell that contains an extra unidentified chromosome
Translocation, reciprocal	46,XX, (t2;6)(q35:p21.3) = a balanced reciprocal translocation with breakpoints in 2q35 and 6p21.3
Translocation, Robertsonian	45,XY,der(14;21)(q10;10) = a balanced carrier of a 14;21 Robertsonian translocation q10 is not a band but indicates the centromere, der = derivative

Sex Chromosome Abnormalities

These abnormalities represent some of the most common of human genetic disorders (1/400-1/500 births). They can be numerical or structural. Phenotypes tend to be less severe than those associated with comparable autosomal disorders.

Sex	Disorder	Karyotype	~frequency
Male	Klinefeltersyndrome	47,XXY	1/1,000
		48, XXXY	1/25,000
		Other (48,XXYY; 49,XXXYY)	1/10,000
	47, XYY syndrome XX males	47, XYY 46 X,X	1/1,000 1/20,000
Female	Turner Syndrome	45, X	1/5,000
	Trisomy X	47, XXX	1/50,000
	XY females	46, XY	1/20,000

You should know that Klinefelter Syndrome is 47XXY and that Turner Syndrome is 46X

Klinefelter syndrome was *first described in 1942 by Dr Harry Klinefelter* and is an example of an sex chromosome abnormality. It is the result of a mistake in cell division either at the time of conception or soon after. Boys with Klinefelter syndrome have a 47,XXY chromosome complement. *In some very rare cases, males may have three (48,XXXY) or four (49,XXXXY) X chromosomes. About 80% of boys with Klinefelter syndrome will have 47,XXY in all cells of the body. About 6% will show a normal chromosome complement i.e. 46,XY in some cells and 46,XXY in the others (mosaicism). In 5% of cases there is a 46,XX/47,XXY mosaic pattern and in the remainder, three or four X chromosomes or other arrangements of an X chromosome.* The incidence of Klinefelter syndrome does not appear to be related to either the age of the mother or the father at the time of conception.

Turner syndrome is another example of a sex chromosome abnormality. *In 1938 Henry Turner, an American doctor, was one of the first people who noticed a pattern in some women who had decreased height and a lack of secondary sex characteristics i.e. breast development, menstruation and sexual hair growth. Some 20 years later, in 1959, it was discovered that women with this pattern of symptoms were missing all or part of an X chromosome. Turner syndrome is not*

common, but neither is it extremely rare. About 1 girl in every 2000 will have this condition. *Approximately 1.5% of all babies conceived have Turner syndrome but only 2-3% of these babies will survive to full term. About 50% of women with Turner syndrome will have an XO chromosome pattern in their cells when they are looked at under the microscope. In about 20% of girls with Turner syndrome, there will be two types of chromosome pattern in the cells. One type will be the usual 46,XX cell line which is common to all females. The other cells will have 45,X. This is called a chromosomal mosaic pattern. The remainder of the girls (about 30%) will have one of several possible arrangements of the second X chromosome. There can be missing portions of the short (p), or the long (q) arm of the chromosome, or the chromosome can be arranged in a ring form. In some cases, the two long arms of the X chromosomes are joined together in an isochromosome Xq. Very rarely, a cell line which contains part of the Y chromosome may exist in a person with Turner syndrome. All these patterns can lead to varying symptoms of Turner syndrome.*

Incidence of Chromosomal Abnormalities in Newborn Surveys	
Type of Abnormality	
Sex chromosome aneuploidy	
Males (47,XXY; 47, XYY; other)	
Females (45X; 47, XXX; other)	
Autosomal aneuploidy	
Trisomy 21	
Trisomy 18	
Trisomy 13	
Other aneuploidy	
Structural abnormalities	
Balanced rearrangements	
Unbalanced rearrangements	
All Chromosomal Abnormalities	1/154 births

It is reasonable that you know the general incidence of Chromosomal Abnormalities in newborn surveys (above, approximately 1/150)