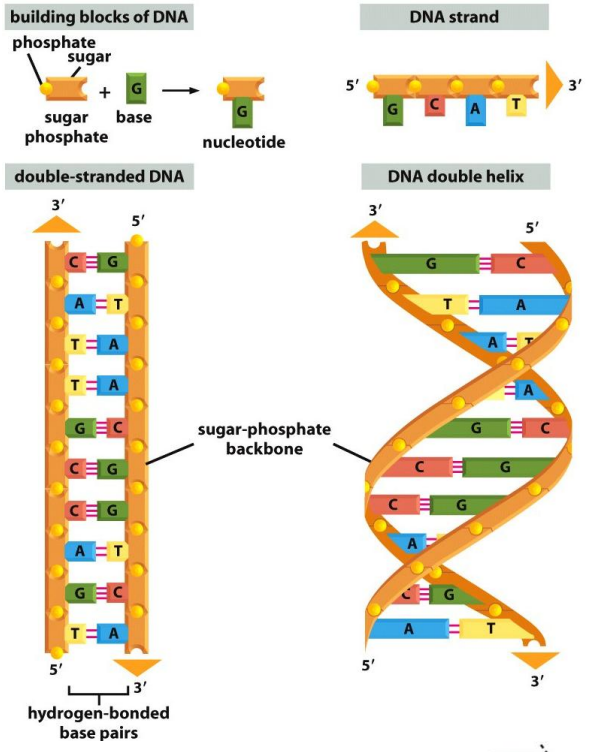
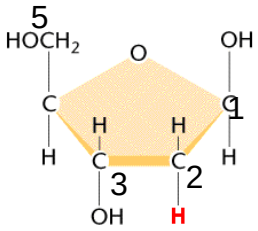
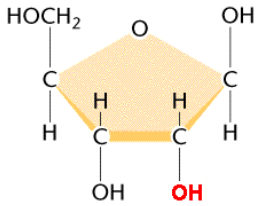
Basic molecular genetics

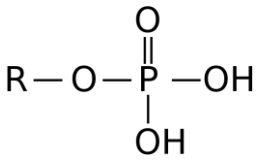
DNA base-pairing

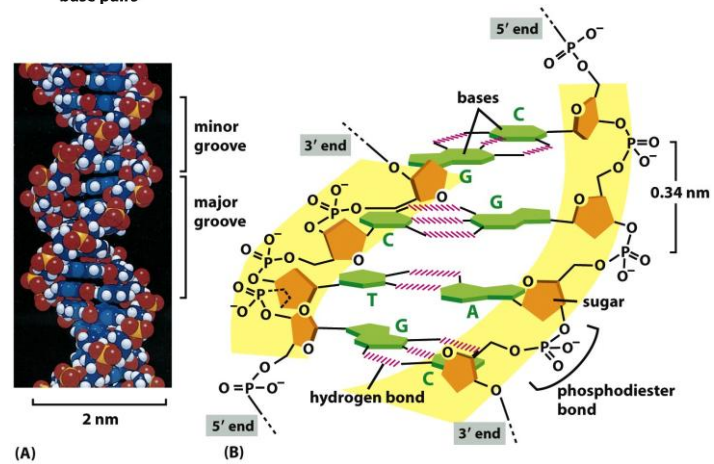


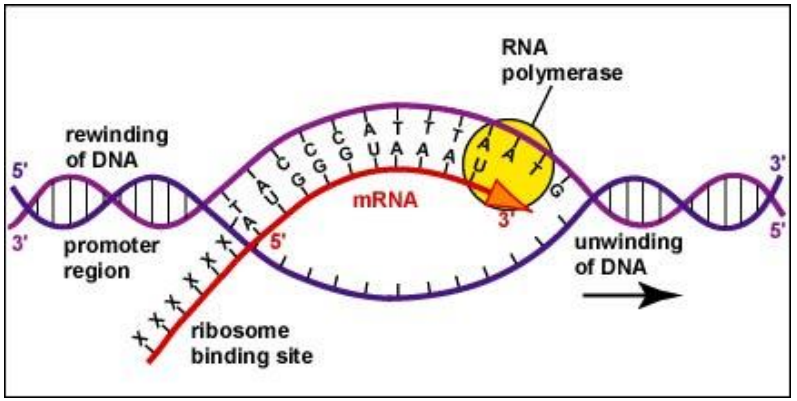
Deoxyribose - DNA is stable, RNA much less so







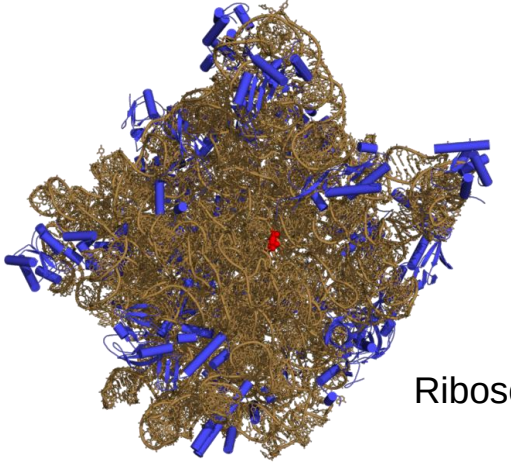




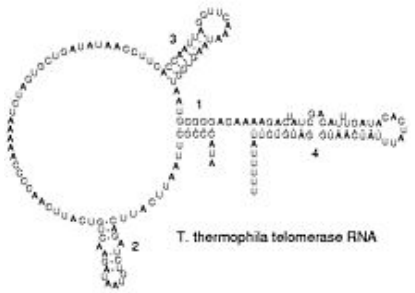
Transcription

Taq polymerase

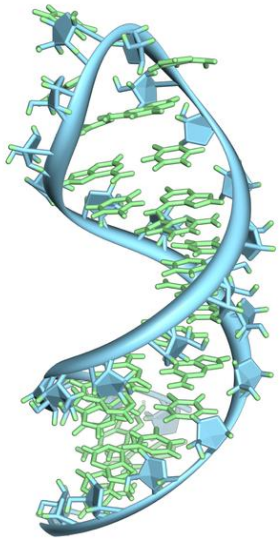
1. Discovered by Kary Mullis
2. A heat-stable DNA polymerase from *Thermus aquaticus*
3. A high-fidelity DNA polymerase
4. A DNA ligase



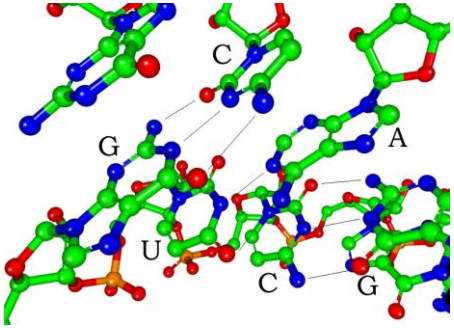
Ribosomal RNA



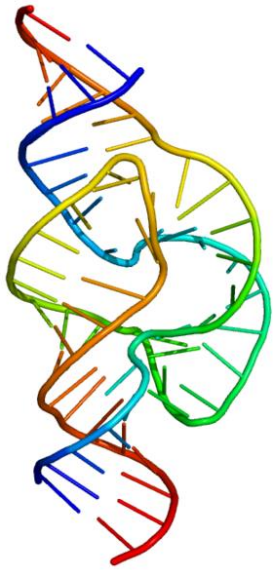
Telomerase RNA



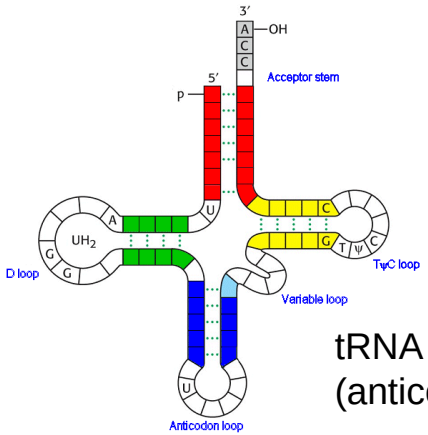
Pre-mRNA



Watson-Crick base pairs in siRNA



Hammer-head ribozyme

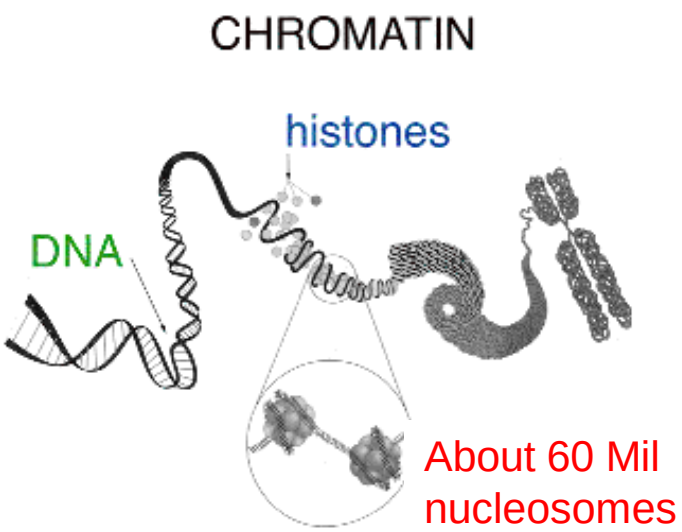


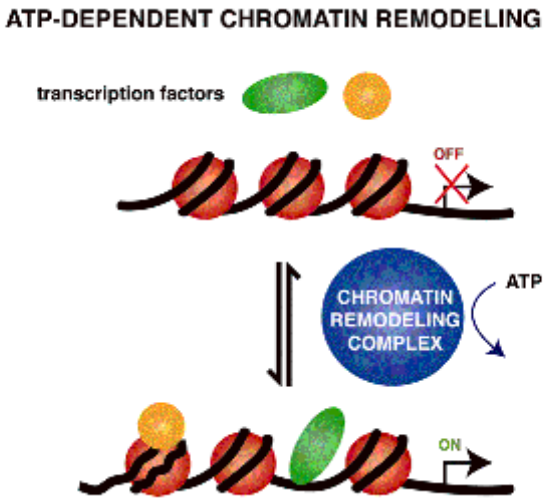
tRNA
(anticodon site and activating enzyme site)

During the three stages of transcription:

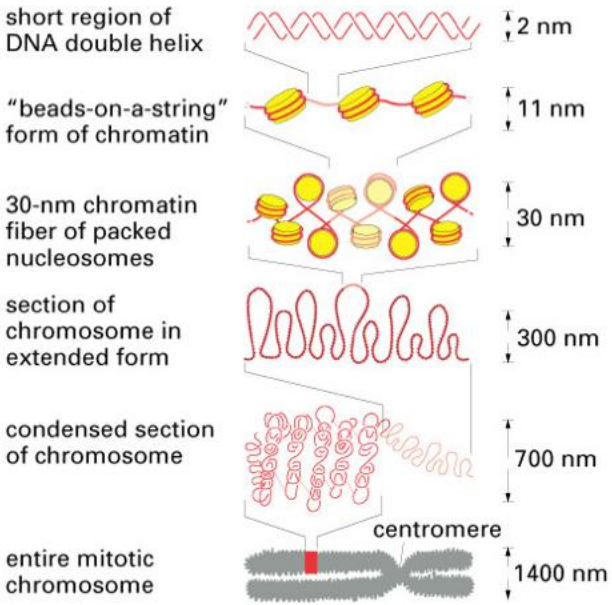
1. DNA polymerase forms a transcription bubble
2. Once the RNA strands have been transcribed, the double helix is restored
3. Polymerase advances from 3’ to 5’ down the template strand
4. The 5’ end of the RNA strand exits the RNA polymerase through a channel

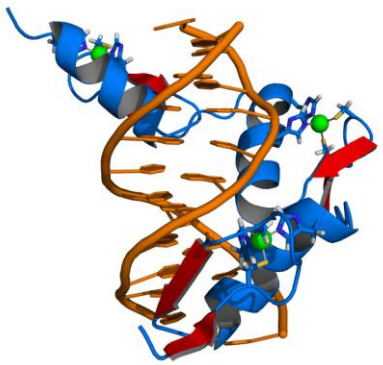
Histones are highly alkaline proteins found in eukaryotic cell nuclei that package and order the DNA into structural units called nucleosomes.





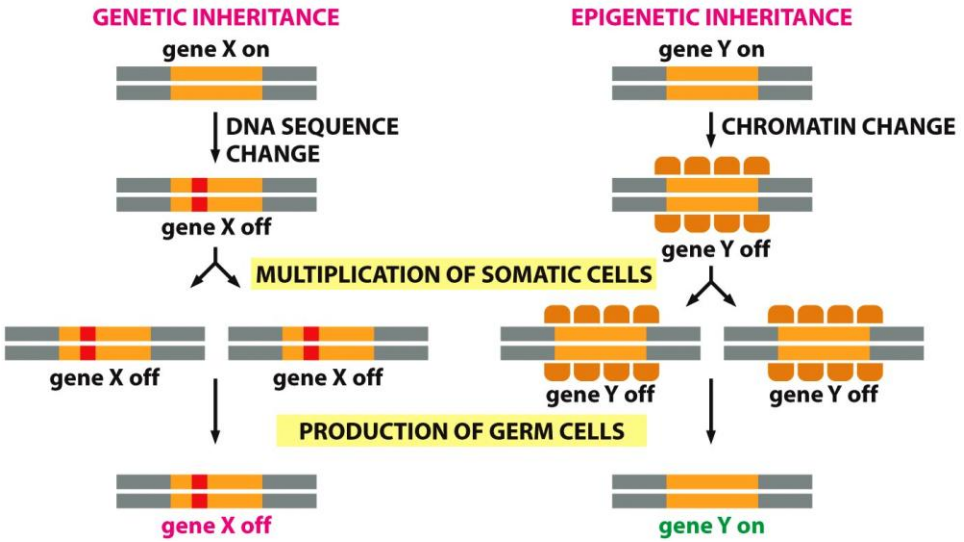
Histones and their regulation (methylation and acetylation)

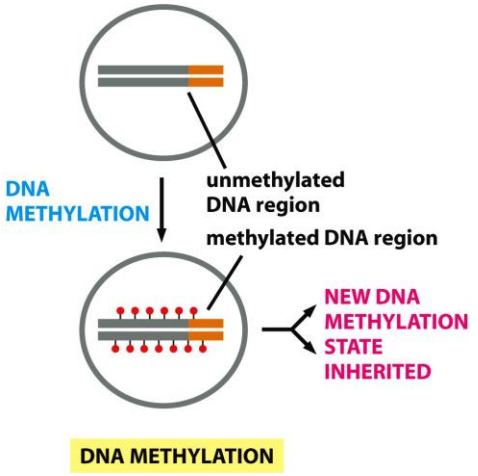


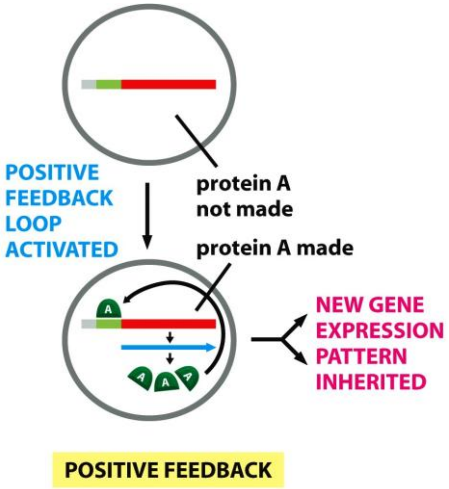


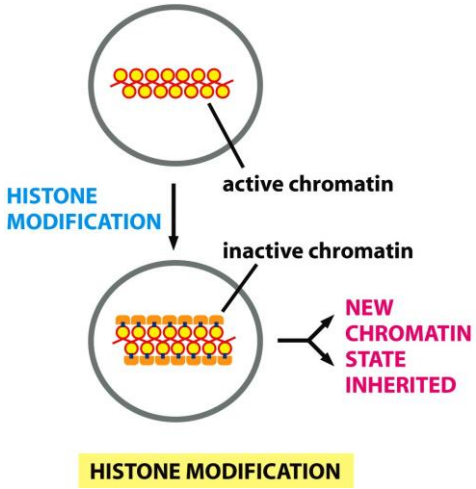
Transcription factor interactions

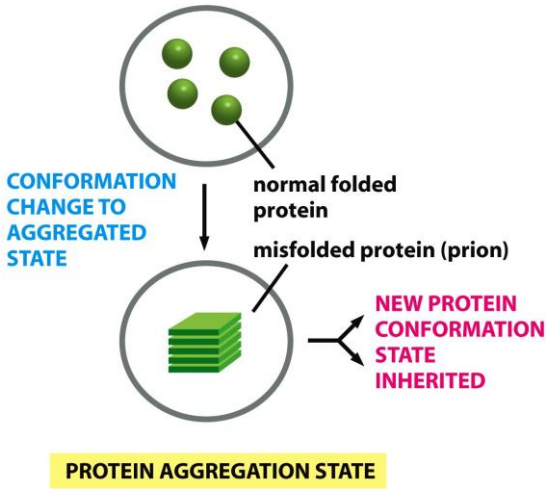
Cell memory based on change in protein structure rather than DNA sequence is a form of **epigenetic** inheritance.











An allele is

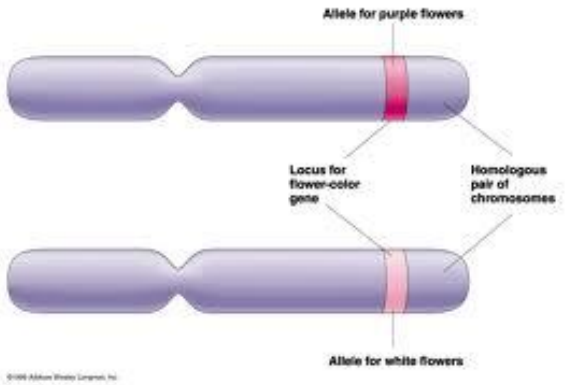
1. One of two or more forms of a gene
   or a genetic locus (generally a group of genes)

2. For blood group antigens, A, B, O are alleles

3. The word is a short form of allelomorph used to describe
   variant forms of a gene detected as different phenotypes

4. All are true

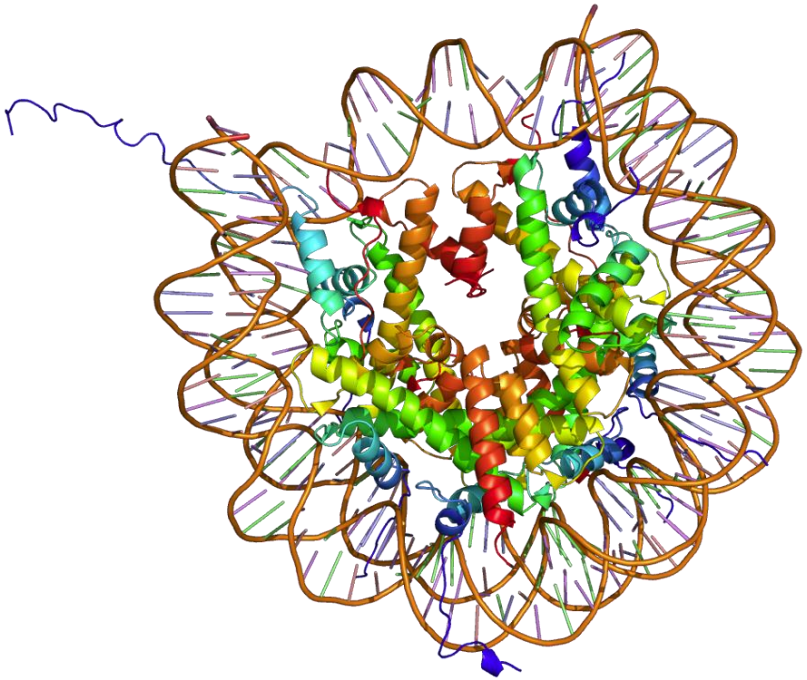
5. None are true

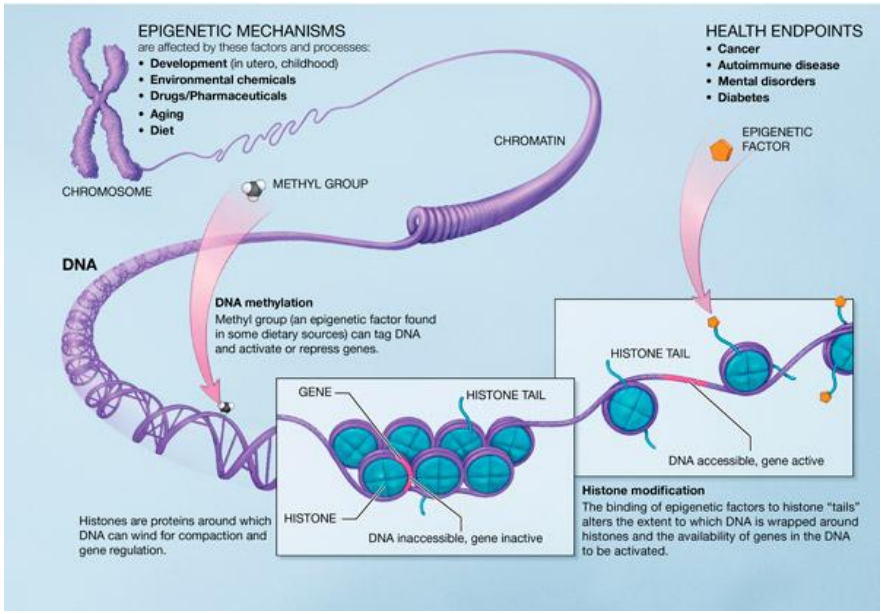


Genetics is the science of genes, heredity, and variation in living organisms

Molecular genetics deals with molecular structure and function of genes

Epigenetics is the study of heritable changes in gene expression or cellular phenotype caused by mechanisms other than changes in the underlying DNA sequence. Examples of such changes are DNA methylation and histone modification, enhancers, suppressors, mircoRNAs, long-noncoding RNAs, and imprinting





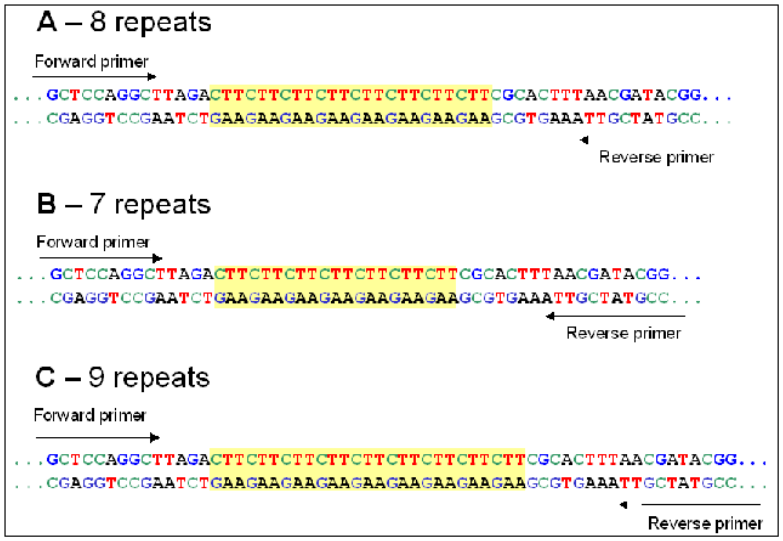
Genomische Prägung (engl. genomic imprinting) bezeichnet das Phänomen, dass die Expression von Genen davon abhängen kann, von welchem Elternteil das Allel stammt. Dieses Vererbungsschema steht im Widerspruch zur klassischen mendelschen Vererbung.

Histones are alkaline proteins in eukaryotic cells that

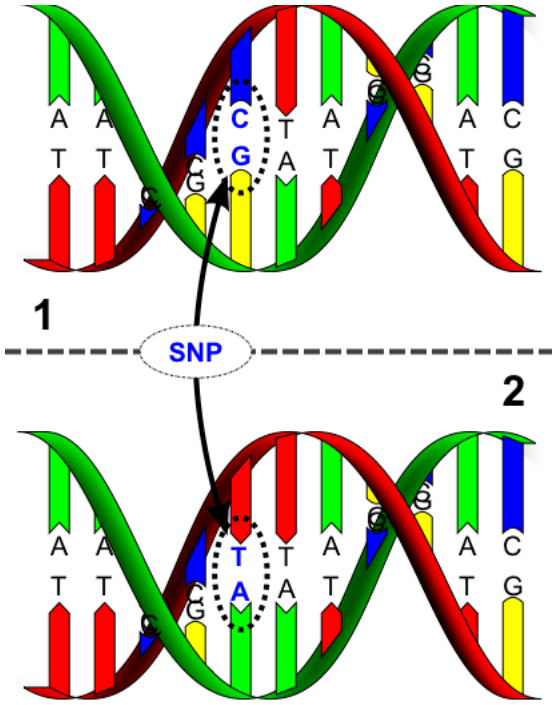
1. Condense 1.8 M DNA into 90 µM
2. Exist as four major families (H1/H5, H2A, H2B and H3
3. Are responsible for posttranscriptional modifications
4. Are methylated on specific alanines

Die Histon-Proteine wurden 1884 vom deutschen Mediziner und Physiologen Albrecht Kossel entdeckt. Der Begriff Histon stammt aus der deutschen Sprache. Unklar ist, ob sich der deutsche Begriff vom griechischen *histanai* oder *histos* herleitet.

Microsatellites, also known as Simple Sequence Repeats (SSRs), short tandem repeats (STRs), or variable numbers of tandem repeats (VNTRs) are repeating sequences of 2-6 base pairs of DNA. Microsatellites are typically co-dominant. They are used as molecular markers in genetics, for kinship, population and other studies. They can also be used to study gene duplication or deletion. Microsatellites are also known to be associated with human disease, especially neurodegenerative disorders and cancer. One common example of a microsatellite is a (CA)n repeat, where n varies between alleles.



A single-nucleotide polymorphism (SNP, pronounced snip) is a DNA sequence variation occurring when a single nucleotide — A, T, C or G — in the genome (or other shared sequence) differs between members of a biological species or paired chromosomes in an individual. A SNP in which both alleles produce the same polypeptide sequence is called a synonymous polymorphism (sometimes called a silent mutation). If a different polypeptide sequence is produced by the polymorphism it is termed a replacement (nonsynonymous) polymorphism. A replacement polymorphism change may be either missense, which results in a different amino acid, or nonsense, which results in a premature stop codon. If a base-pair is missing, a "frame-shift" mutation results.



Microsatellites:

1. First launched by the Russians
2. Variable numbers of tandem repeats
3. Are translated into polypeptides
4. Single base-pair substitutions
5. Most useful for genetic association

Genetic linkage is the tendency of genes that are located proximal to each other on a chromosome to be inherited together during meiosis. Genes whose loci are nearer to each other are less likely to be separated onto different chromatids during chromosomal crossover, and are therefore said to be genetically linked.

Possible alleles

C

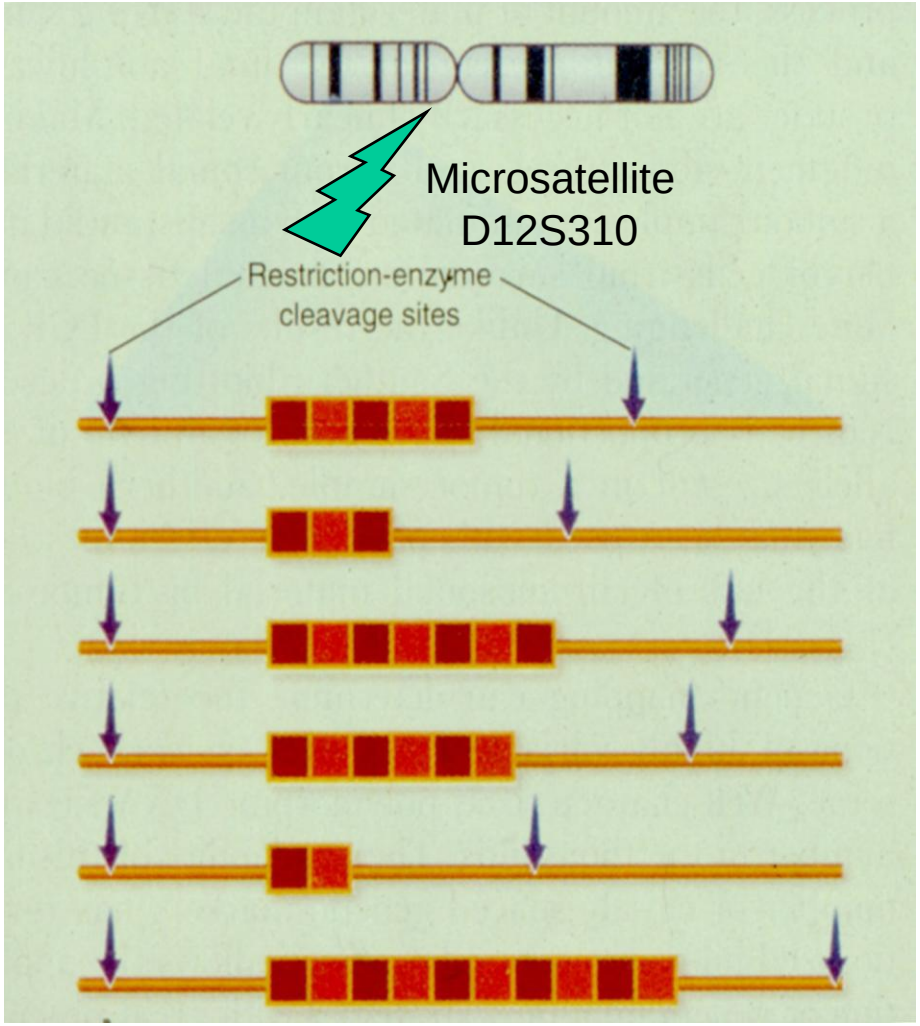
B

E

D

A

F



Linkage analysis, finding gene markers that are always inherited with the disease (we do not use RFLP anymore but the example is a good one)

Female

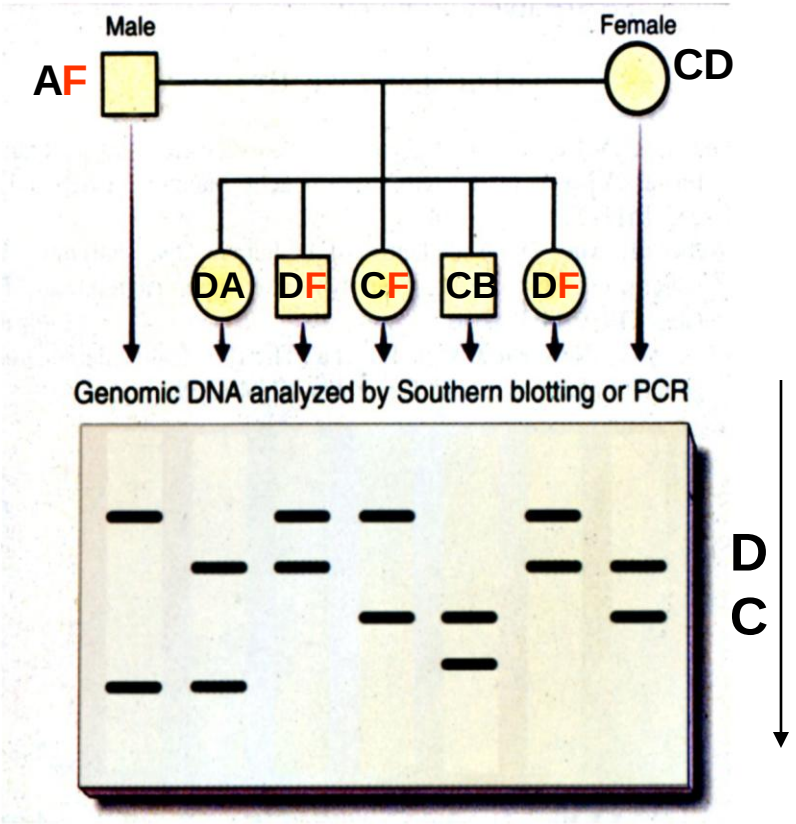
Male

Gel →

Slower { **F**

Faster { **A**

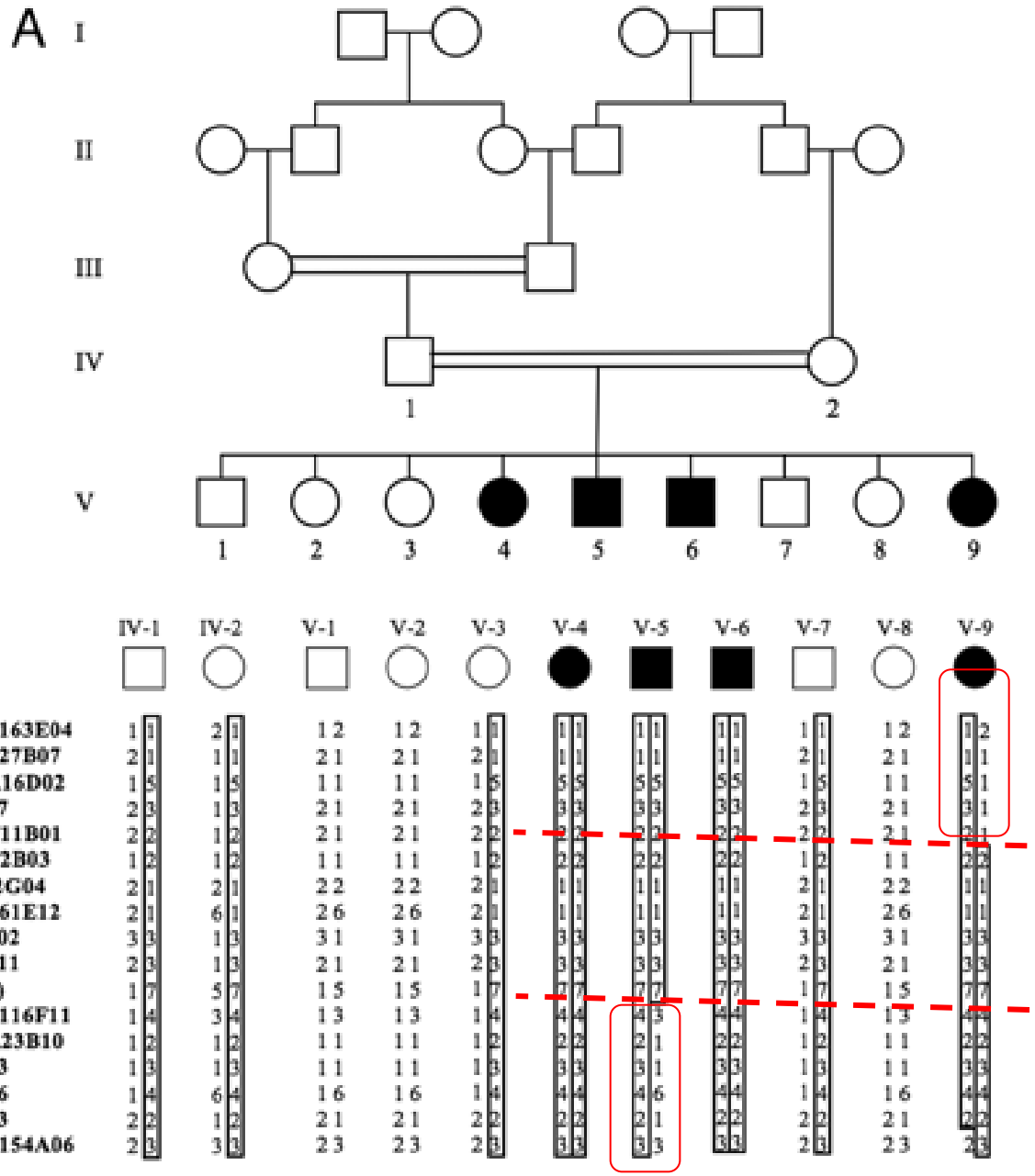
Disease gene is next to F



What is a haplotye?

1. A combination of alleles (DNA sequences) at adjacent locations (loci) on the chromosome that are transmitted together.
2. Collection of microsatellites on opposite chromosomes
3. Cannot be expressed in a Punnett square
4. All are true
5. None is true

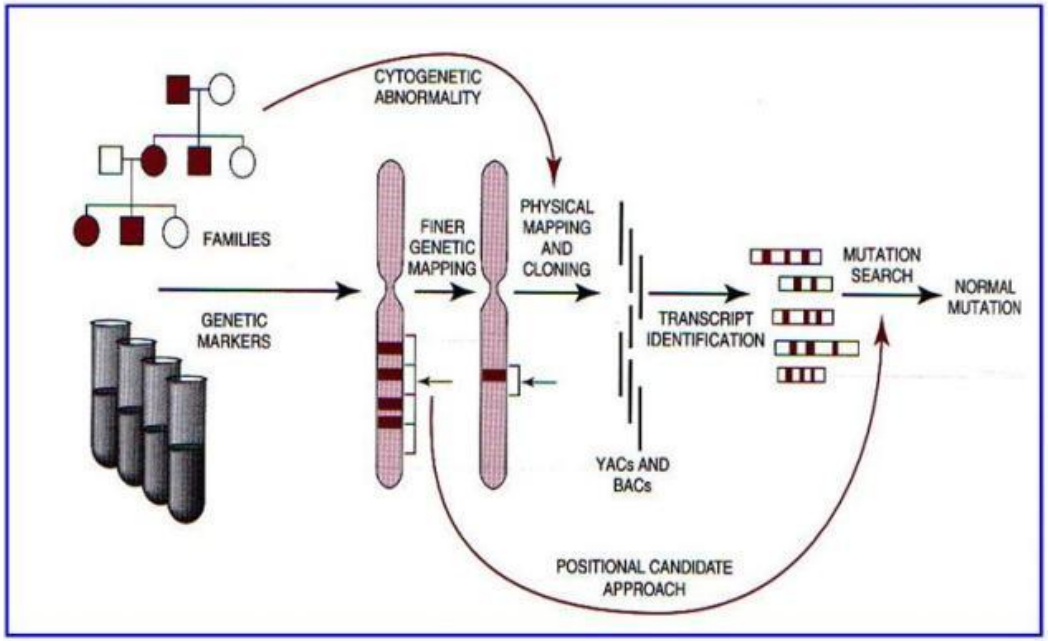
An efficient strategy for mapping human genes that cause recessive traits has been devised that uses mapped restriction fragment length polymorphisms (RFLPs) and the DNA of affected children from consanguineous marriages. The method involves detection of the disease locus by virtue of the fact that the adjacent region will preferentially be homozygous by descent in such inbred children.

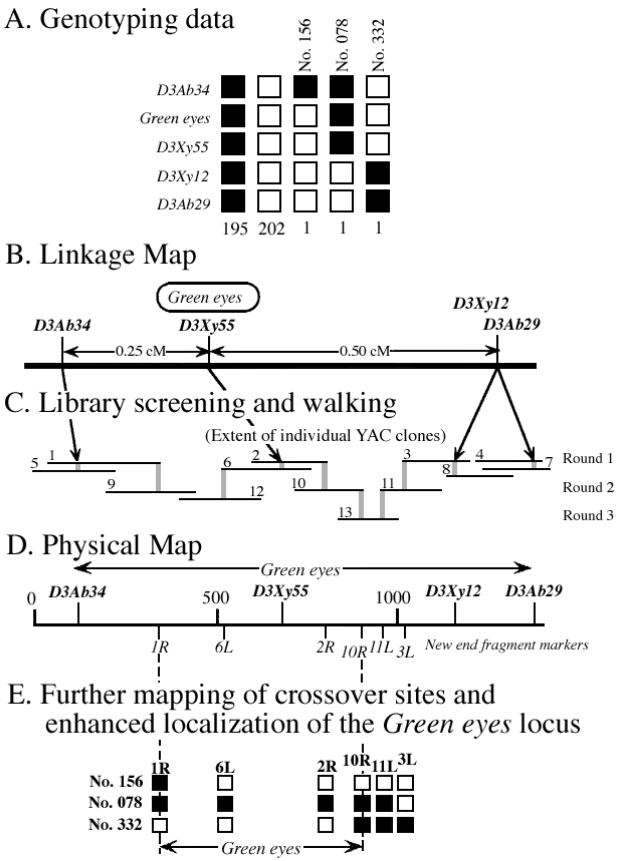


Nonsense mutations

1. Same as missense mutations but occur in introns
2. Point mutations that cause a stop codon
3. Employ a nonsense-mediated DNA decay pathway
4. Cause sickle-cell anemia

Positional cloning is a technique which is used in genetic screening to identify specific areas of interest in the genome, and then determine what they do. This type of genetic screening is sometimes referred to as reverse genetics, because researchers start by figuring out where a gene is, and then they determine what it does, in contrast with methods which start by determining the function of a gene and then finding it in the genome. Genes related to conditions such as Huntington's Disease and cystic fibrosis have been identified with this technique.



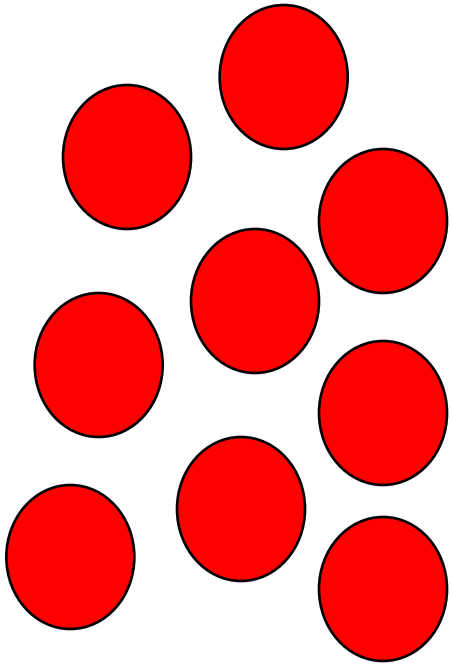


Epigenetics:

1. Involves genetic epidemiology
2. Involves changes in the DNA sequence
3. Involves phenotypes that do not persist after cell divisions
4. Genomic imprinting is an epigenetic process

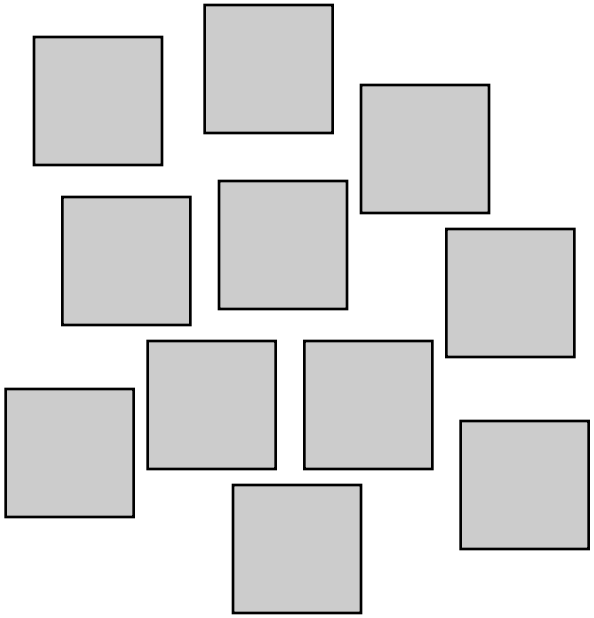
Genetic association is the occurrence, more often than can be readily explained by chance, of two or more traits in a population of individuals, of which at least one trait is known to be genetic. Studies of genetic association aim to test whether single-locus alleles or genotype frequencies (or more generally, multilocus haplotype frequencies) differ between two groups of individuals (usually diseased subjects and healthy controls). Genetic association studies are based on the principle that genotypes can be compared "directly", i.e. with the sequences of the actual genomes.

Cases



x100

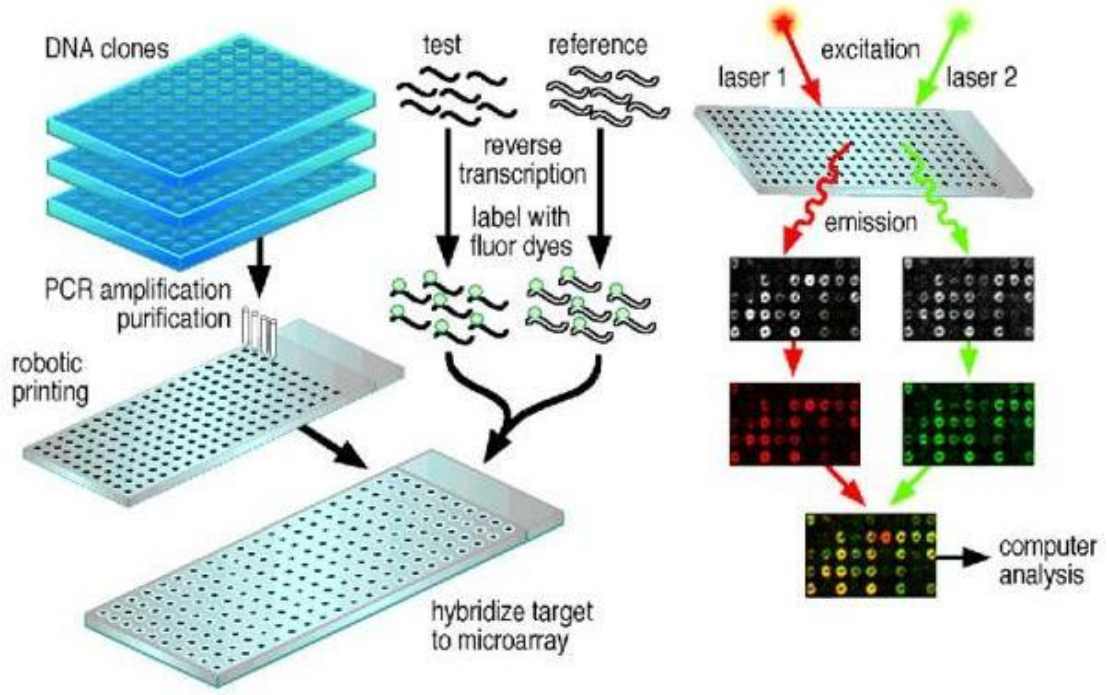
Controls



x100

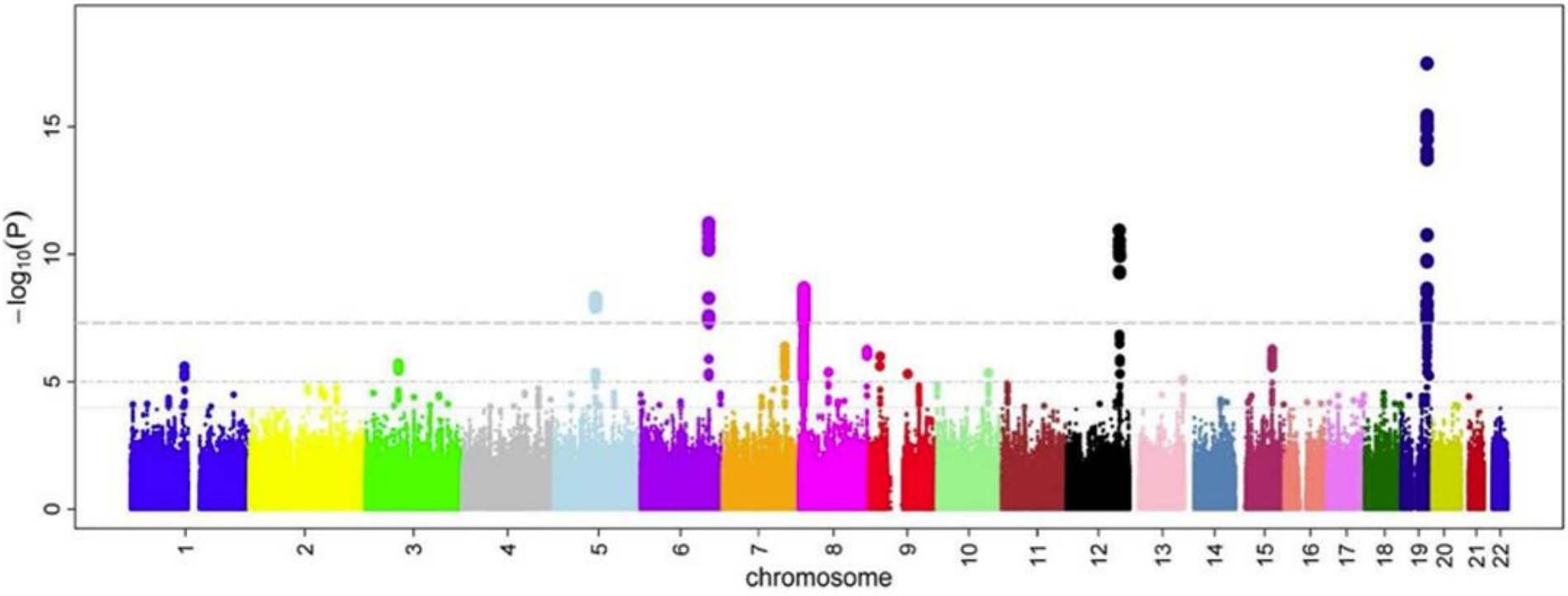
“Top-down” association study or “bottom-up” association study

SNP array is a type of DNA microarray which is used to detect polymorphisms within a population. A single nucleotide polymorphism (SNP), a variation at a single site in DNA, is the most frequent type of variation in the genome. For example, there are around 10 million SNPs that have been identified in the human genome. As SNPs are highly conserved throughout evolution[citation needed] and within a population, the map of SNPs serves as an excellent genotypic marker for research.



Macroarrays contain sample spot sizes of about 300 microns or larger and can be easily imaged by existing gel and blot scanners. The sample spot sizes in microarray are typically less than 200 microns in diameter and these arrays usually contains thousands of spots.

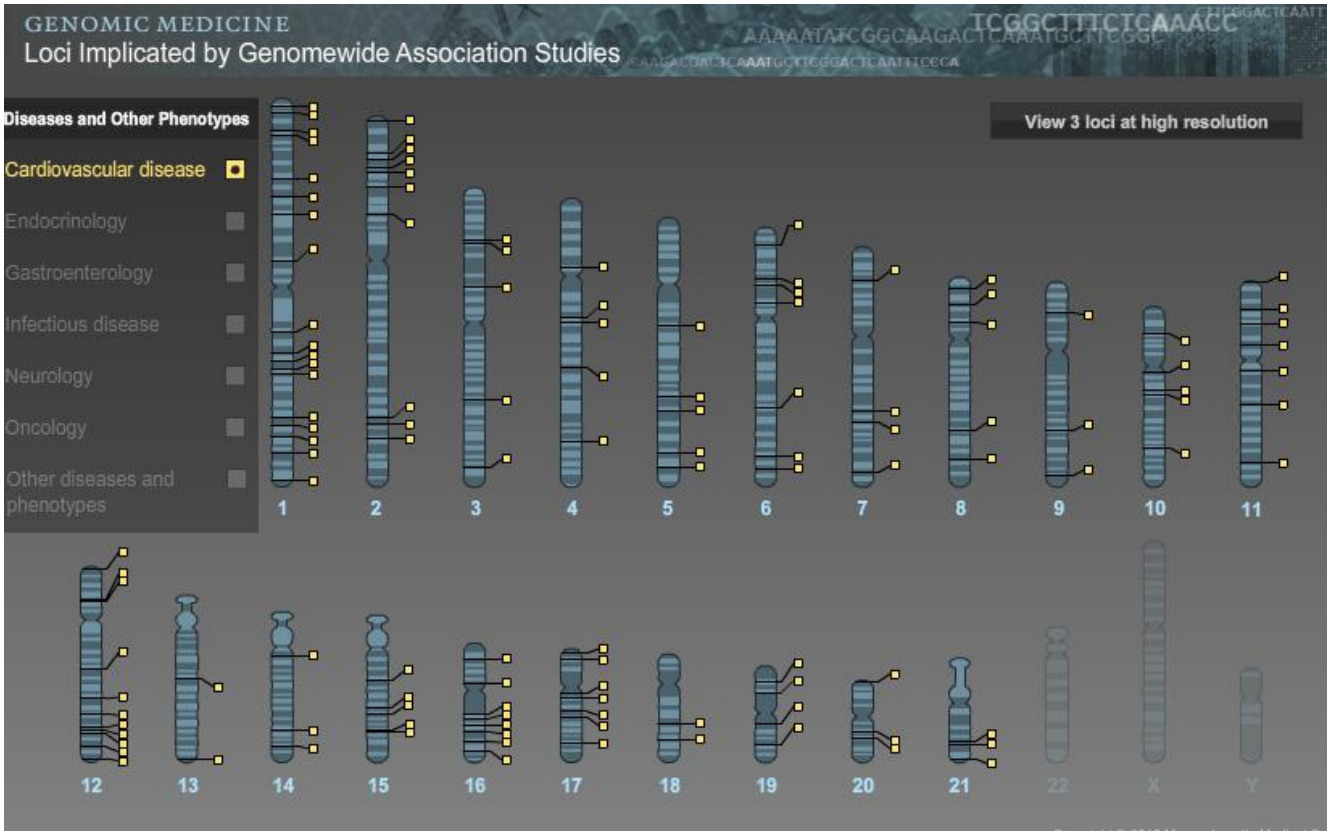
In genetic epidemiology, a genome-wide association study (GWA study, or GWAS), also known as whole genome association study (WGA study, or WGAS), is an examination of many common genetic variants in different individuals to see if any variant is associated with a trait. GWAS typically focus on associations between single-nucleotide polymorphisms (SNPs) and traits like major diseases. These studies normally compare the DNA of two groups of participants: people with the disease (cases) and similar people without (controls). Each person gives a sample of DNA, from which millions of genetic variants are read using SNP arrays.



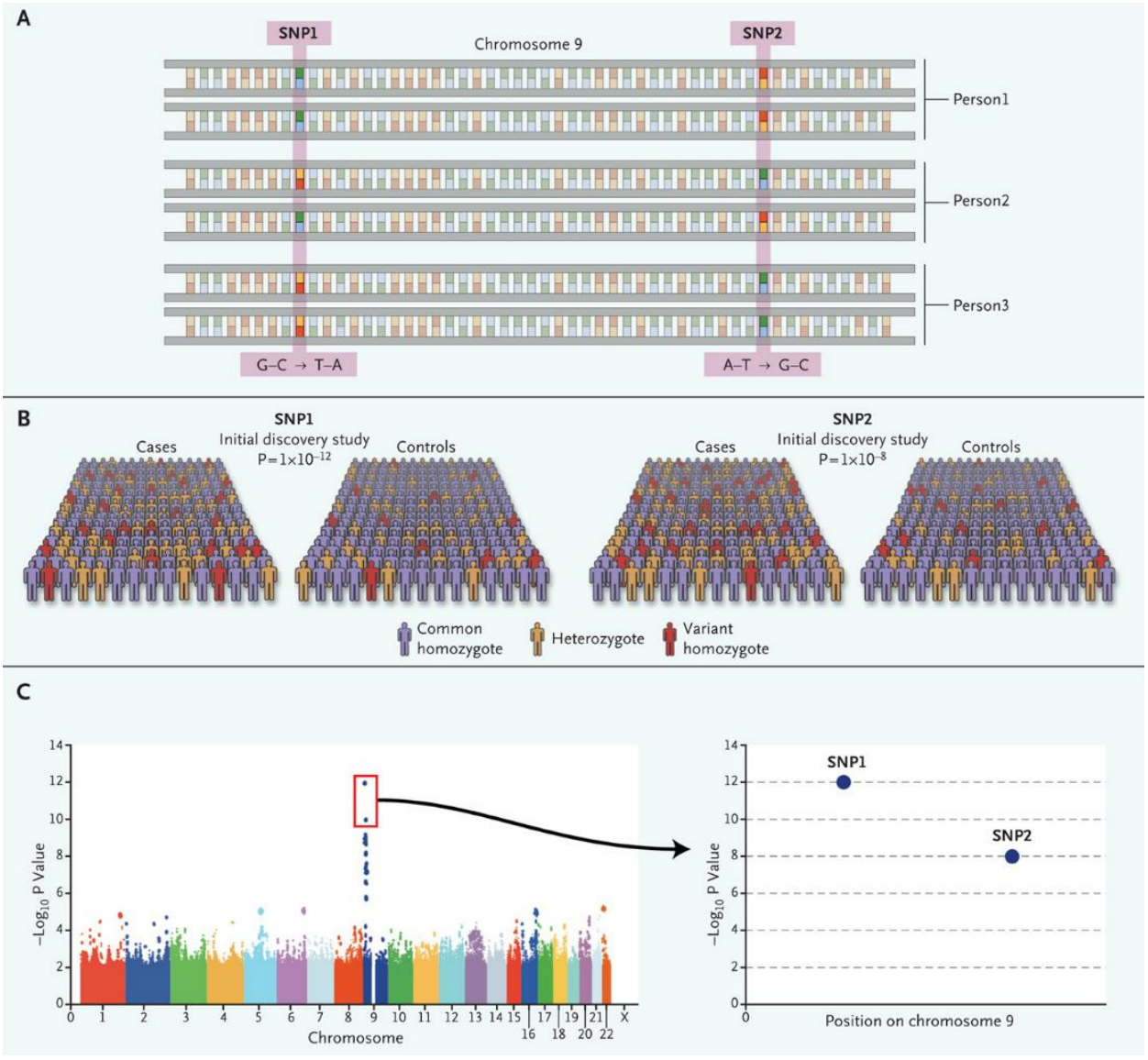
Genetic linkage:

1. Is independent of homologous recombination
2. Helps answer the question “where is it”
3. Requires microsatellites and not SNPs
4. Is fundamental to mitosis

Genomewide association studies — in which hundreds of thousands of single-nucleotide polymorphisms (SNPs) are tested for association with a disease in hundreds or thousands of persons have revolutionized the search for genetic influence on chomplex traits. Such conditions, in contrast with single-gene disorders, are caused by many genetic and environmental factors working together, each having a relatively small effect and few if any being absolutely required for disease to occur. Although complex conditions have been referred to as the geneticist's nightmare,3 in the past 5 years genomewide association studies have identified SNPs implicating hundreds of robustly replicated loci (i.e., specific genomic locations) for common traits.



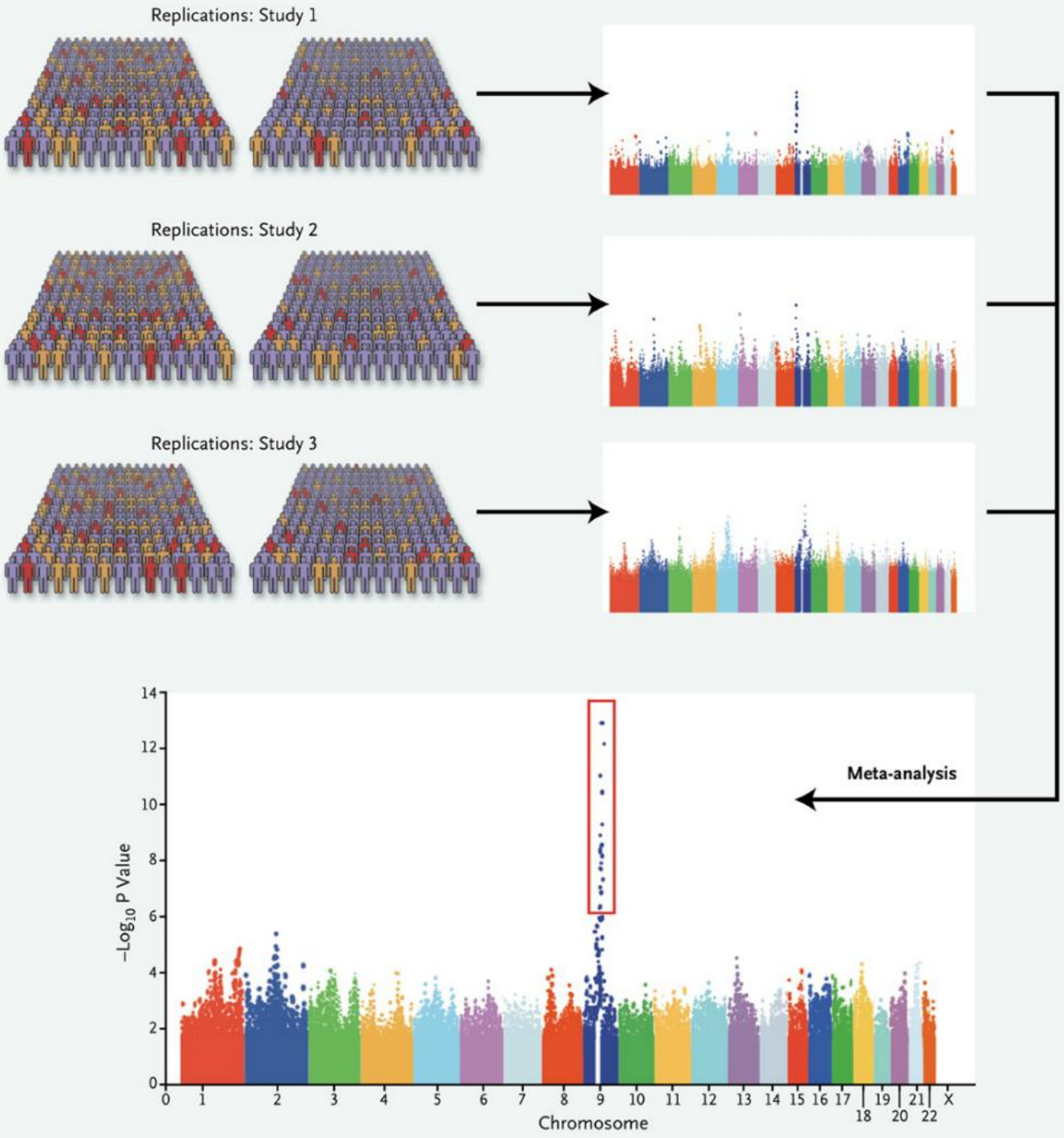
Manolio TA. N Engl J Med 2010



Panel A depicts a small locus on chromosome 9, and thus a very small fragment of the genome. In Panel B, the strength of association between each SNP and disease is calculated on the basis of the prevalence of each SNP in cases and controls. In this example, SNPs 1 and 2 on chromosome 9 are associated with disease, with P values of 10−12 and 10−8, respectively. The plot in Panel C shows the P values for all genotyped SNPs that have survived a quality-control screen, with each chromosome shown in a different color. The results implicate a locus on chromosome 9, marked by SNPs 1 and 2, which are adjacent to each other (graph at right), and other neighboring SNPs.

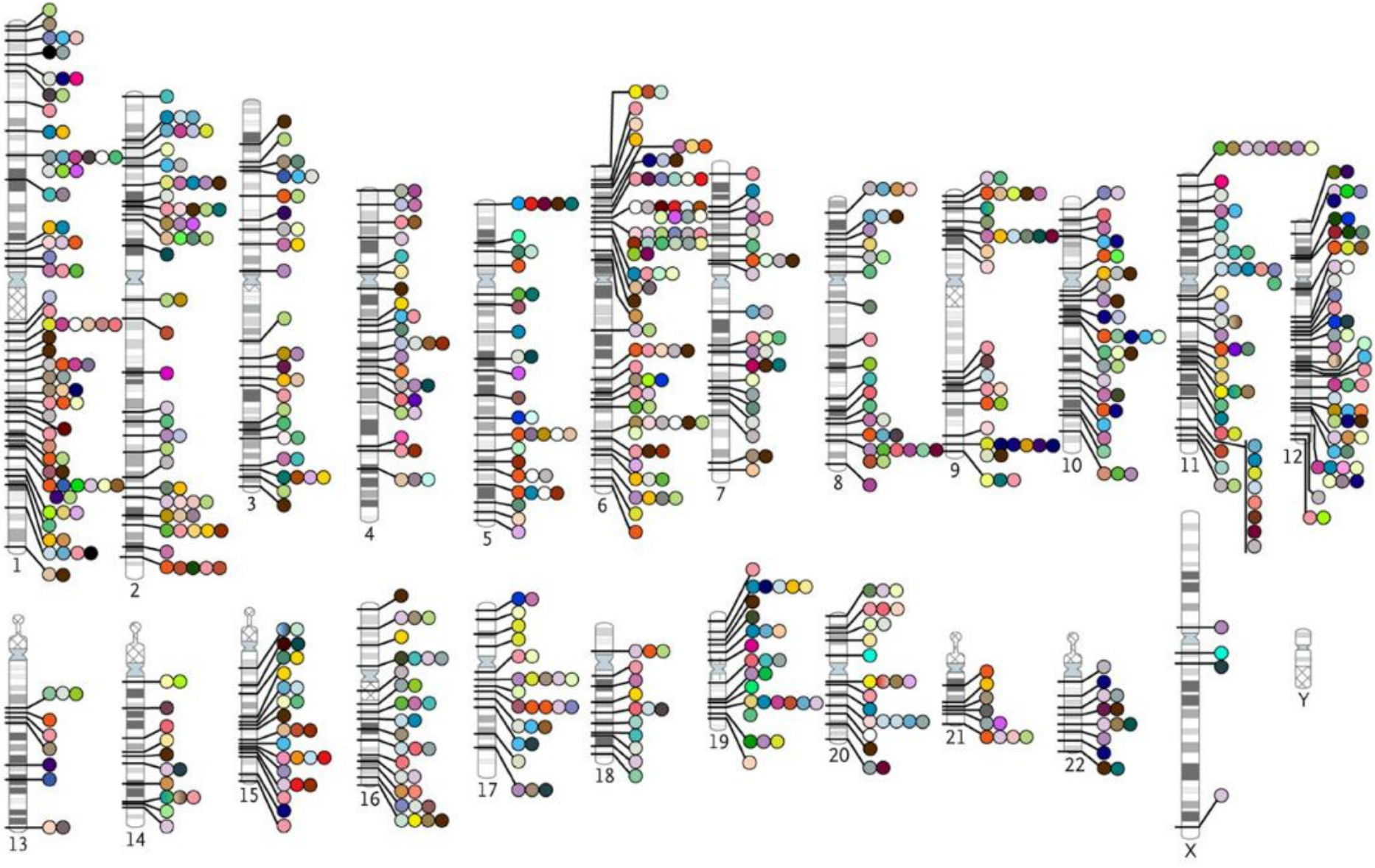
Non-synonymous SNPs

1. Are very common in GpC islands
2. Cause no change in amino-acid sequence
3. Are found in the 3’ UTR regions
4. Can change the protein sequence



The results of genomewide association studies can be evaluated in a meta-analysis, which combines the results of multiple studies to improve the power for detecting associations. In this example, the results of three studies, none of which may show genomewide significance individually, are combined in a meta-analysis to reveal a strong, significant signal on chromosome 9.

Circles indicate the chromosomal location of nearly 800 single-nucleotide polymorphisms (SNPs) significantly associated ($P<5\times10^{-8}$) with a disease or trait and reported in the literature (545 studies published through March 2010 yielded the associations depicted). Each disease type or trait is coded by color.



GWAS studies

1. Are very expensive

2. Have catapulted epidemiologists into molecular genetics

3. Have found fewer genes and mechanisms than is generally maintained

4. Are politically correct and very trendy but may be overrated

5. All are true