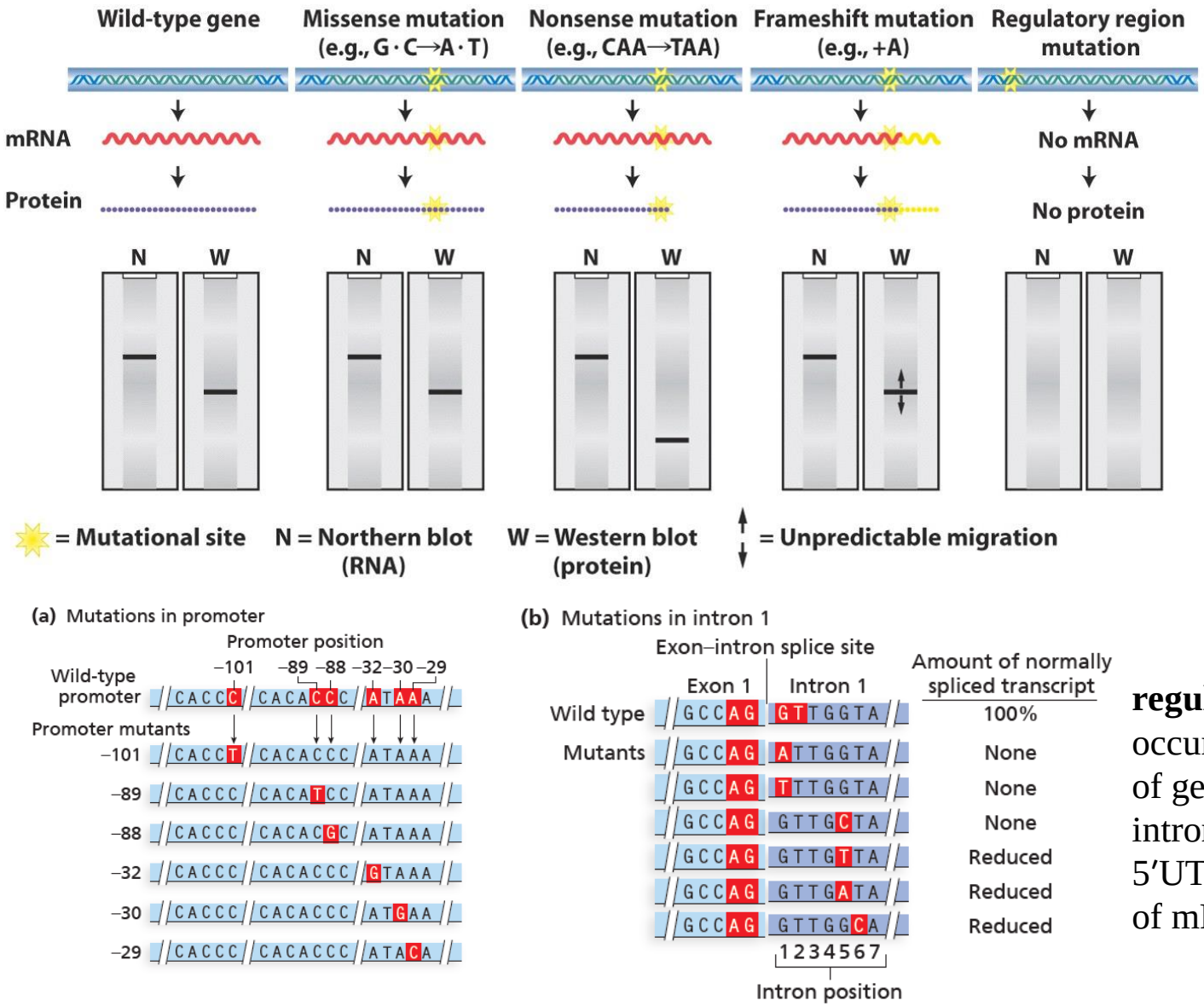


regulatory mutations, occur in noncoding regions of genes, such as promoters, introns, and regions coding 5′UTR and 3′UTR segments of mRNA.

Figure 11.4 Regulatory mutations of the human b-globin gene. (a) These base-pair substitution mutations in the promoter reduce, increase or complete elimination of transcription of the gene. (b) These base-pair substitutions in intron 1 reduce or eliminate normal pre-mRNA splicing.

Genetic Variation II

- Describe the functional effect of mutations
- Give examples of loss of function, gain of function, haploinsufficiency, dominant negative effects,
- Genotype-phenotype correlations

Different mutation classes

- A gene cannot be restricted to a coding sequence because it also contains sequences necessary for its expression, i.e. the promoter, the 5 and 3 untranslated regions (5UTR and 3UTR), The polyadenylation signal, and intronic sequences that have to be very precisely excised to reconstitute the exact coding sequence of the messenger RNA (or several coding sequences if the gene is alternatively spliced).
- **Therefore, a mutation in a gene will have different effects depending on its site and its nature.**

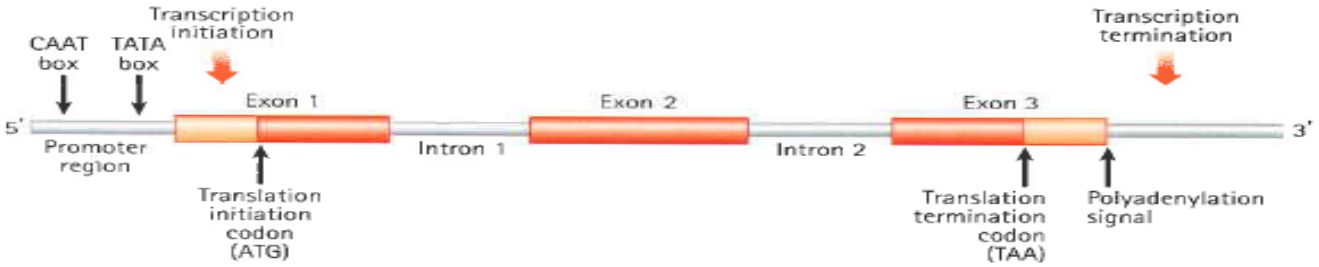
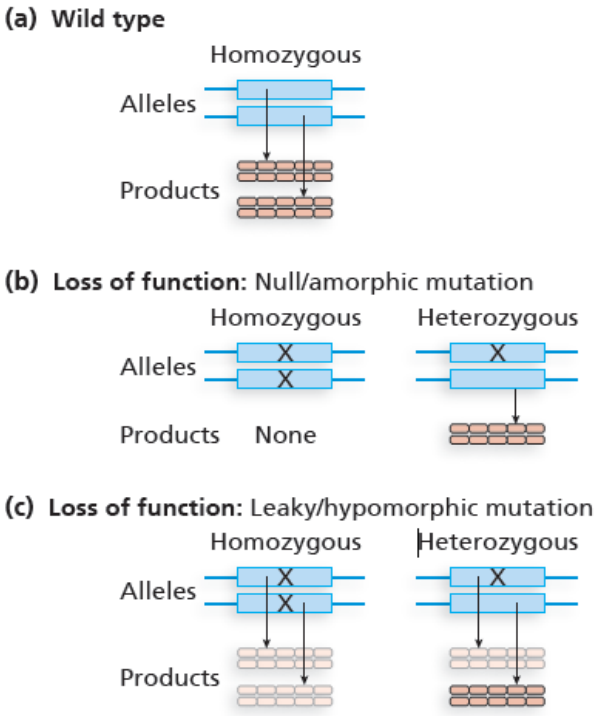


Fig. 2.6
Representation of a typical human structural gene.

Different mutation classes

- The loss of function mutations causes
  - A decrease or a loss of the gene product (quantitative mutations) OR
  - A decrease or a loss of the activity of the gene product (qualitative mutations);

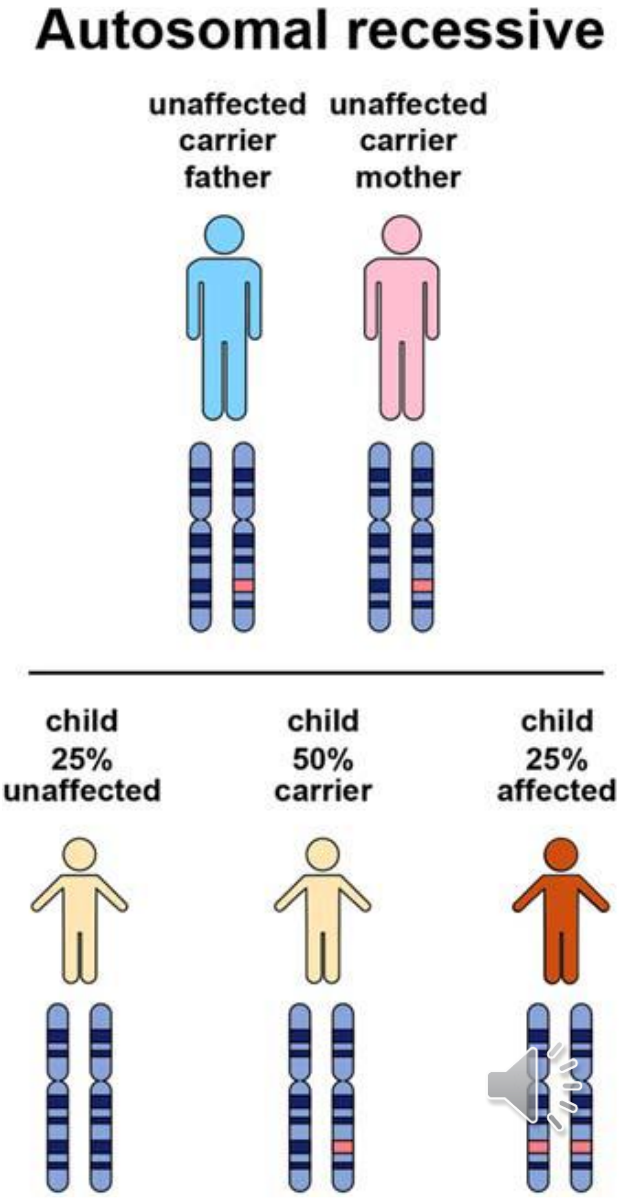


The expression of the products of wild-type alleles produces wild-type phenotype

Null alleles produce no functional product. Homozygous null organisms have mutant (amorphic) phenotype due to absence of the gene product (e.g. nonsense or stop-gain mutations, frameshift and splice-altering mutations).

Leaky mutant alleles produce a small amount of wild-type gene product. Homozygous organisms have a mutant (hypomorphic) phenotype. (e.g. nonsynonymous or missense mutations).

- Loss of function mutations frequently have a recessive effect compared with the functional gene effect. In heterozygous: the wild-type allele having an activity sufficient to compensate for the loss of the mutated gene.
- This is the case for most of the recessive diseases (β–thalassemia, cystic fibrosis, haemophilia, Duchenne/Becker myopathy).



Dominant-negative variants

A dominant-negative variant, a mutated protein disrupts the function of the normal protein. This often happens when proteins work in groups or complexes.

Dominant-negative variants can complicate genetic disorders, as the presence of one mutant allele can significantly disrupt normal protein function even if the other allele is normal. This contrasts with simple recessive mutations, where one normal allele is often sufficient to maintain normal function.

Dominance and recessivity are explained by molecular pathology

Example: Fibrillin-1 Gene (FBN1):

Normal Function: Fibrillin-1 is a protein that is essential for the formation of elastic fibers found in connective tissue.

Dominant-Negative Variant: In Marfan syndrome, mutations in the FBN1 gene can lead to the production of defective fibrillin-1, which disrupts the integrity of connective tissues, resulting in symptoms like long limbs, flexible joints, and cardiovascular problems.

Mechanisms of dominant disease

- haploinsufficiency, dominant negative, or gain-of-function effects
- Haplo-insufficiency: Loss-of-function mutations in the heterozygous state in which half normal levels of the gene product result in phenotypic effects.

Figure 9.30 Haploinsufficiency: Some loss-of-function mutant alleles are dominant to wild-type alleles. The *GLI3* gene is haploinsufficient.
heterozygotes *GLI3* exhibit polydactyly (extra fingers and toes).

The mutant allele has novel function that produces a mutant phenotype in homozygous and heterozygous organisms and may be more severe in homozygous organisms.

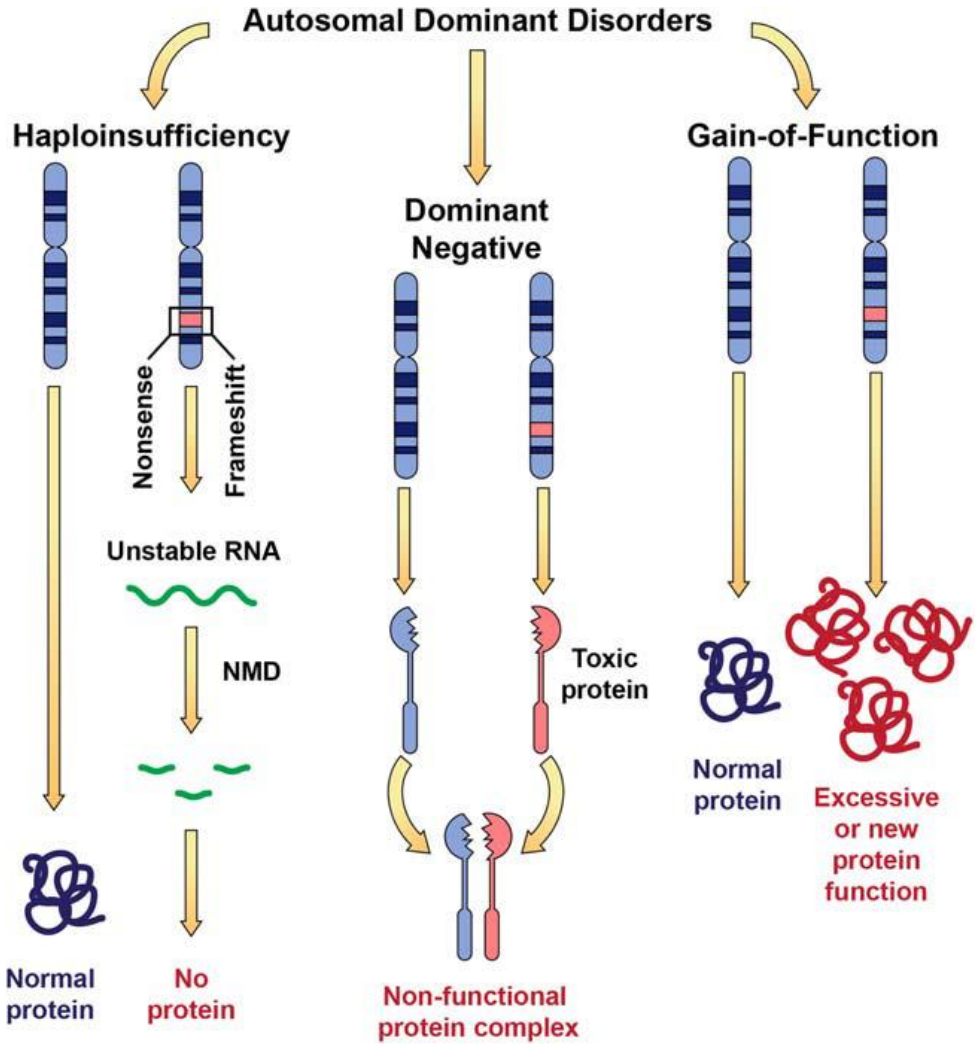
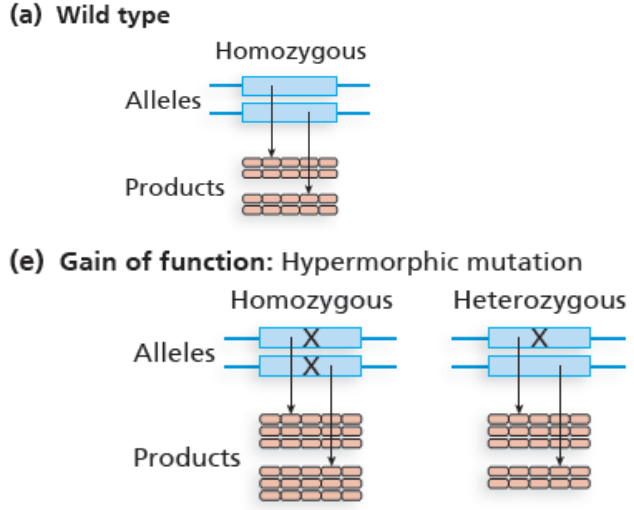


FIGURE 6.2

The three major mechanisms of disease in autosomal dominant disorders are illustrated: haploinsufficiency, dominant negative, and gain-of-function effects, where the mutated copy of a gene leads to no protein, a toxic protein, or an excessive or new protein function, respectively.

Gain of function mutations: Hypermrphic

- The gain of function mutations causes an increase in the amount of gene product (quantitative mutation) or an increase in its activity
  - Sometimes creates a new property, leading to a toxic product responsible for a pathological effect.



Excessive expression of the gene product leads to excessive gene action. The mutant phenotype may be more severe or lethal in the homozygous genotype than in the heterozygous genotype.

- Hypermorphic allele where the protein activity cannot be switched off.
- e.g. Receptor usually is only activated if bond by ligands. A receptor that transduce signals without ligands is Constitutively Active.

Gain of function mutations: Hypermrphic

Cell signaling often starts with an extracellular molecule (called ligand, peptides or chemicals) binding to its specific receptor protein. The conformational change of the receptor leads to the activation of the enzymatic activity of the receptor or its downstream binding proteins

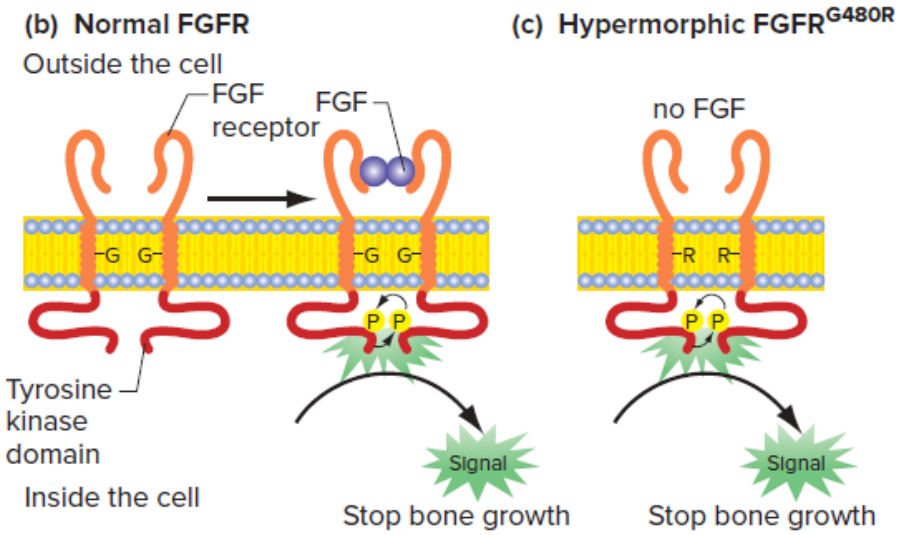
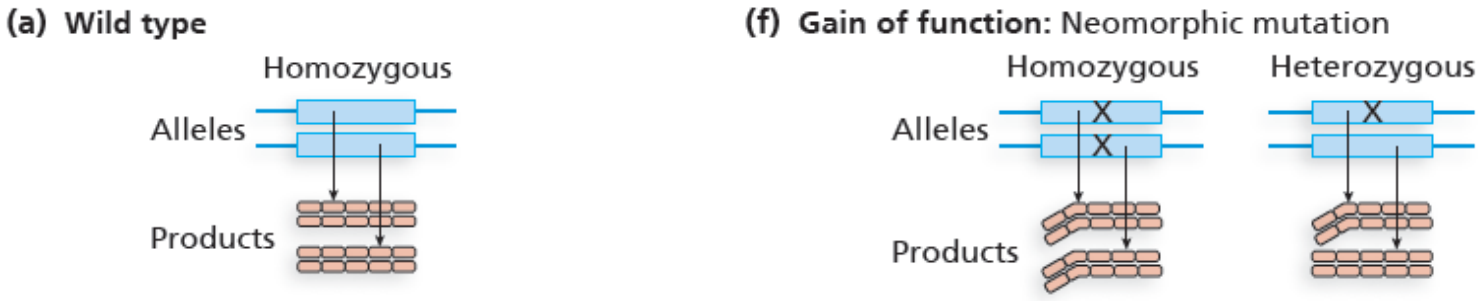


Figure 9.31: **(b)** FGFR3 encodes a dimeric transmembrane receptor that is normally activated only when it is bound to the hormone FGF. The tyrosine kinase domain of one activated FGFR3 subunit adds phosphate groups (P in yellow circles) to the other subunit and vice versa. These phosphorylations initiate a signal that ultimately stops bone growth. **(c)** Mutant FGFR3 (p.G480R) protein is always activated, whether FGF is present or not, leading to improper bone development.

Example: Achondroplasia: FGFR3: p.G480R

Example: myeloproliferative neoplasms (MPNs), particularly essential thrombocythemia: JAK2: p.V617F

Gain of function mutations: Neomorphic



- Gain-of-function mutations resulting from neomorphic ("new form") mutations acquire novel gene activities not found in the wild type and are usually dominant.
- Function of the gene is completely changed → new product is maybe being produced or the gene is being expressed in a new location/new time
- Homozygotes for a neomorphic allele may exhibit a more severely affected phenotype than do heterozygotes.

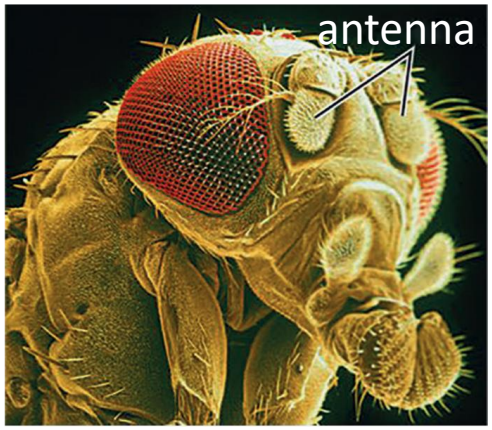
Genetic Analysis An Integrated Approach by John L. Bowman, Mark L. Sanders (z-lib.org)

Gain of function mutations: Neomorphic

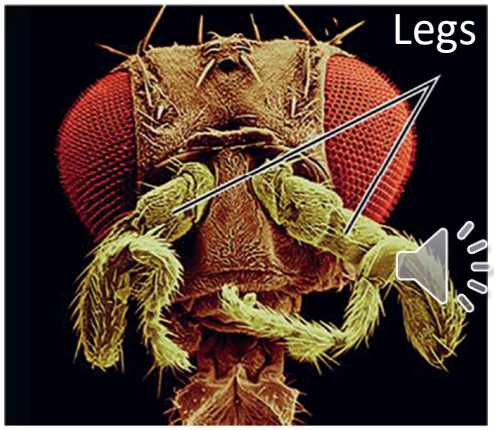
- Gain of Novel Function: Unlike loss-of-function mutations that reduce or eliminate the normal activity of a gene, neomorphic mutations lead to a new function.
  - Example: The BCR-ABL fusion protein in chronic myeloid leukemia (CML) gains abnormal tyrosine kinase activity.
- Altered Protein Activity: The mutated gene product (protein) may have a completely new activity not present in the wild-type protein.
  - Example: IDH1 and IDH2 Mutations in Gliomas
  - Normal Function: Isocitrate dehydrogenase 1 and 2 (IDH1 and IDH2) are enzymes involved in the citric acid cycle, converting isocitrate to α-ketoglutarate (α-KG).
  - Neomorphic Mutation: Mutations in IDH1 (e.g., R132H) and IDH2 (e.g., R172K) lead to a new enzymatic activity that converts α-KG to 2-hydroxyglutarate (2-HG), an oncometabolite.
  - Impact: 2-HG accumulation disrupts cellular metabolism contributing to oncogenesis. This leads to altered gene expression and promotes the growth of gliomas and other cancers.

Gain of function mutations: Neomorphic

- Ectopic Expression: The gene may be expressed in different tissues or at different times compared to its normal expression pattern, potentially leading to abnormal effects.
  - The gene is expressed in tissues or developmental stages where it is normally inactive.
  - Example: Antennapedia mutation, where a gene that normally specifies leg development is aberrantly expressed in the head, leading to the development of legs instead of antennae.



Normal fly



mutant fly

Genetic Analysis, 3rd edition
Philip Meneely
Genes, Genomes, and Networks in Eukaryotes

Genetic heterogeneity can be explained by molecular pathology

Mutations of different genes can produce the same, or very similar, abnormal phenotypes. This phenomenon is known as genetic heterogeneity

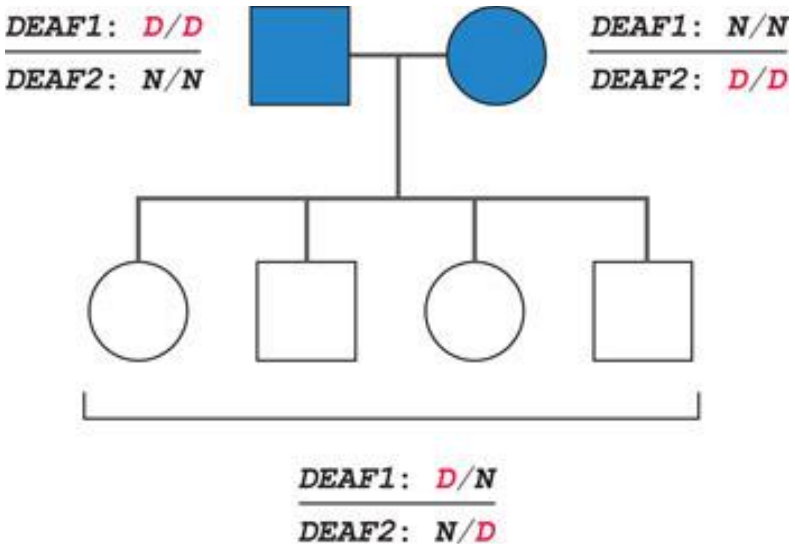
Heterogeneous traits have the same phenotype but are caused by mutations in different genes

locus heterogeneity: one disease/several genes

- The simplest way to consider a Mendelian disease is as a monogenic disease, meaning a disease for which every patient is affected in the same gene (one disease/one gene).
  - E.g. phenylketonuria and cystic fibrosis.
  - The disease is monogenic
- Different genes within the genome can cause the same clinical condition. This can complicate genetic diagnosis and research because identifying a single causative gene might not explain the genetic basis for a condition in all patients.
- Locus heterogeneity explains how parents who are both affected with the same common recessive disorder produce multiple unaffected children.

Inter-locus heterogeneity: one disease/several genes

- Deafness in humans can be caused by mutations in ~ more than 60 different genes



- **Hypertrophic Cardiomyopathy:** Over 20 genes involved.
- Retinitis pigmentosa
  - A common cause of visual impairment due to photoreceptor degeneration associated with abnormal pigment distribution in retina.
  - Known to occur in AD, AR, and X-linked forms

Allelic heterogeneity: one gene/several diseases

- Allelic heterogeneity occurs when different mutations arise within the same gene locus.
- These mutations can involve various types of genetic alterations, including point mutations, insertions, deletions, and structural rearrangements.

Allelic Heterogeneity in Cystic Fibrosis (CF):

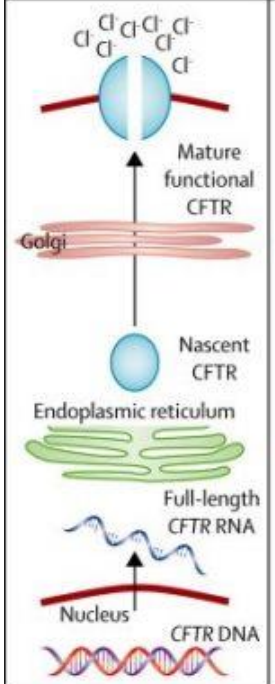
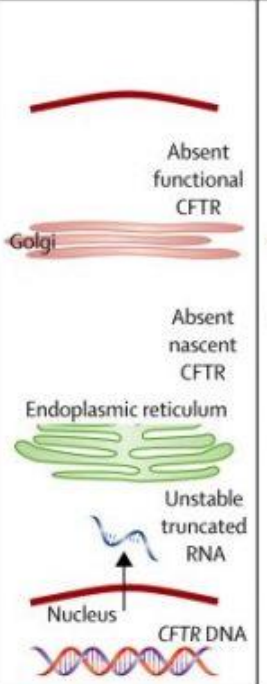
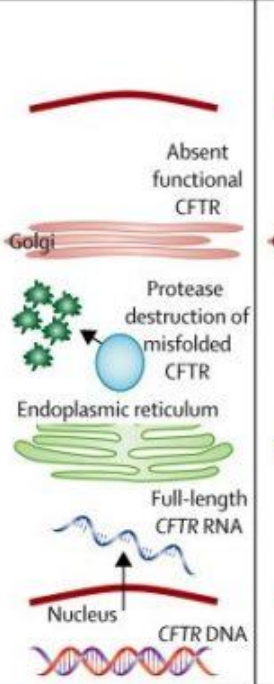
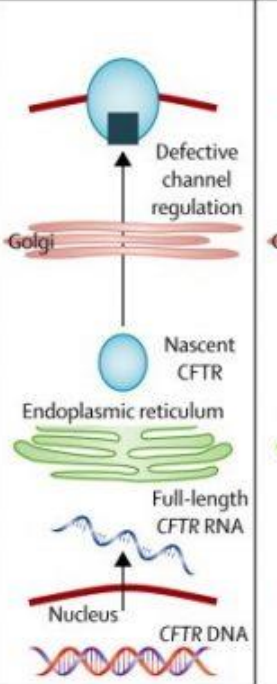
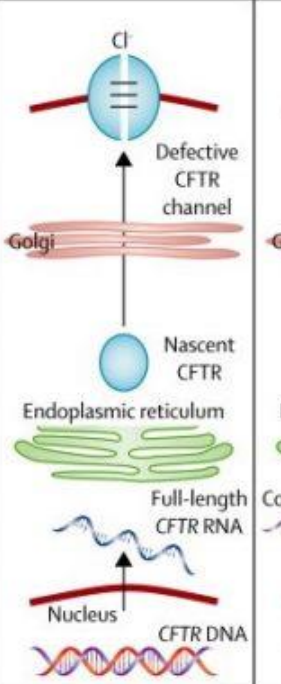
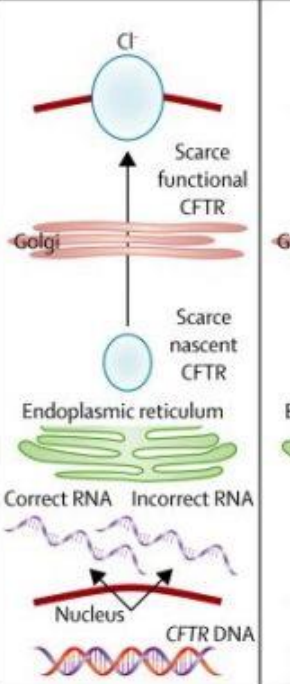
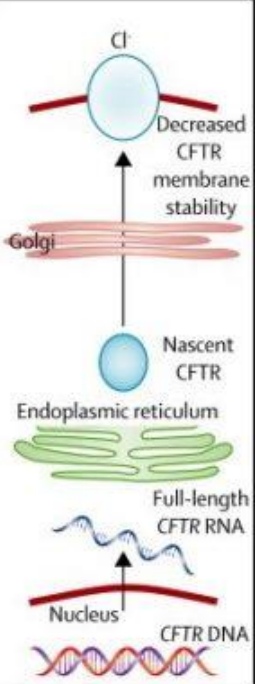
- CF is primarily caused by mutations in the Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) gene located on chromosome 7.
- The CFTR gene encodes a protein involved in the regulation of chloride and sodium ion transport across cell membranes.

Allelic heterogeneity: one gene/several diseases

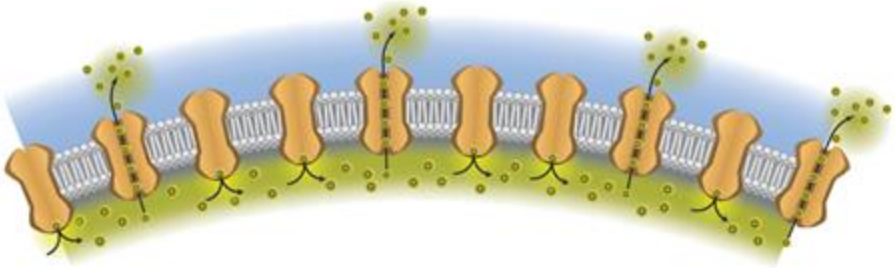
Example: >2000 different mutations in CFTR in cystic fibrosis.

- Some mutations cause: Only the abnormality of the male reproductive tract.
- Some mutations cause: Pancreatic insufficiency, severe progressive lung disease and congenital absence of vas deferens in males (classic form)
- Some mutations cause: lung disease but normal pancreatic function.

CF classification

Normal	I	II	III	IV	V	VI
						
CFTR defect	No functional CFTR protein	CFTR trafficking defect	Defective channel regulation	Decreased channel conductance	Reduced synthesis of CFTR	Decreased CFTR stability
Type of mutations	Nonsense; frameshift; canonical splice	Missense; aminoacid deletion	Missense; aminoacid change	Missense; aminoacid change	Splicing defect; missense	Missense; aminoacid change
Specific mutation examples[11]	Gly542X Trp1282X Arg553X 621+1G→T	Phe508del Asn1303Lys Ile507del Arg560Thr	Gly551Asp Gly178Arg Gly551Ser Ser549Asn	Arg117His Arg347Pro Arg117Cys Arg334Trp	3849+10kbC→T 2789+5G→A 3120+1G→A 5T	4326delTC Gln1412X 4279insA

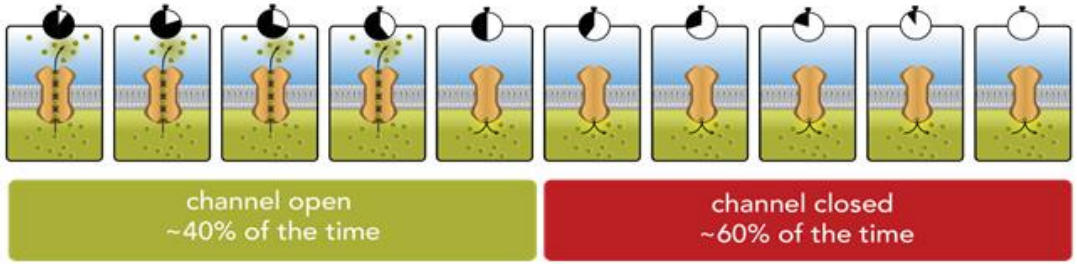
Normal CFTR quantity at the cell surface



- CFTR **quantity** is determined by:
  - –CFTR synthesis
  - –CFTR processing and trafficking
  - –CFTR surface stability

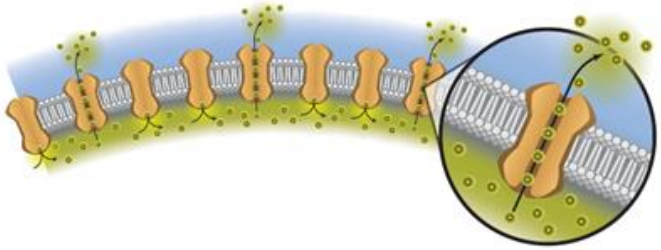
Normal CFTR channel-open probability

Status of a single channel over time[1*]



- **Channel-open probability:** the fraction of time that a single CFTR protein channel is open and transporting ions

Normal CFTR channel conductance



- **Channel conductance:** rate at which ions move through open channels

Clinical and genetic heterogeneity

- Many diseases present a clinical heterogeneity (phenotypic heterogeneity) characterized by the severity of the symptoms, by the severity or the way the disorder evolves.
- One part of the clinical heterogeneity can be explained by allelic heterogeneity, by the fact that different mutations can have variable phenotypic effects, either in their type or in their strength.
- Another part of the clinical heterogeneity, notably between affected individuals within the same family, can be explained by the effect of modifying genes that can increase or decrease the effect of pathological mutations in the gene principally involved, without being responsible for the appearance of the pathology.
- Finally, the effect of the environment on the appearance of the disease and on its clinical variability should not be neglected.