

ORGANIZAÇÃO E FUNCIONALIDADE DO GENOMA HUMANO



Departamento de *Genética*

Nilce M. Martinez Rossi

Fenótipo = GENÓTIPO
+
Ambiente



O que é o genoma?

Projetos Genoma



articles

Initial sequencing and analysis of the human genome

International Human Genome Sequencing Consortium*

* A partial list of authors appears on the opposite page. Affiliations are listed at the end of the paper.

The human genome holds an extraordinary trove of information about human development, physiology, medicine and evolution. Here we report the results of an international collaboration to produce and make freely available a draft sequence of the human genome. We also present an initial analysis of the data, describing some of the insights that can be gleaned from the sequence.

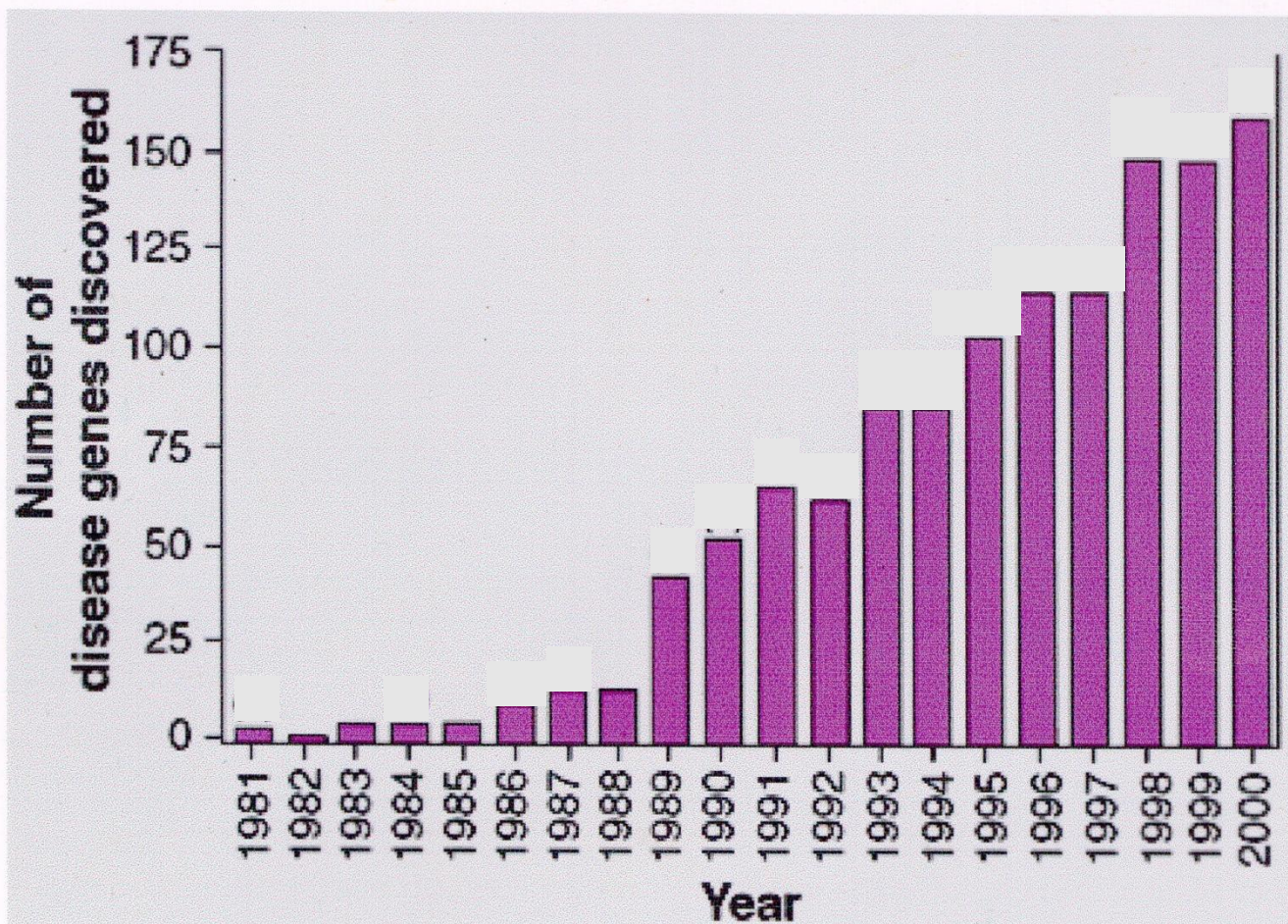
The rediscovery of Mendel's laws of heredity in the opening weeks of the 20th century^{1,2} sparked a scientific quest to understand the nature and content of genetic information that has propelled biology for the last hundred years. The scientific progress made falls naturally into four main phases, corresponding roughly to the four quarters of the century. The first established the cellular basis of

- coordinate regulation of the genes in the clusters.
- There appear to be about 30,000–40,000 protein-coding genes in the human genome—only about twice as many as in worms or fly. However, the genes are more complex, with more alternative splicing generating a larger number of protein products.
- The full set of proteins (the 'proteome') encoded by the human

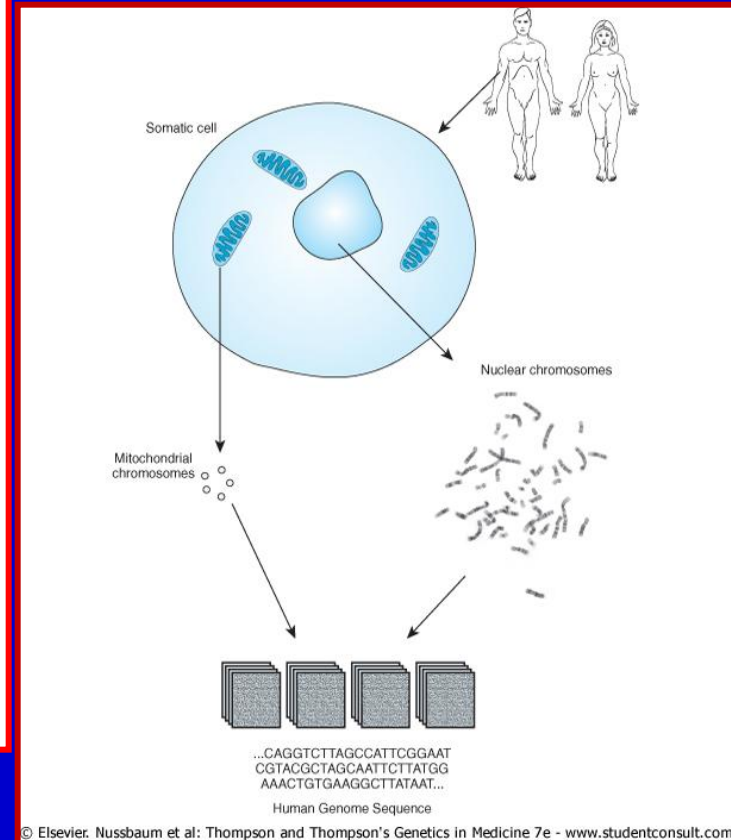
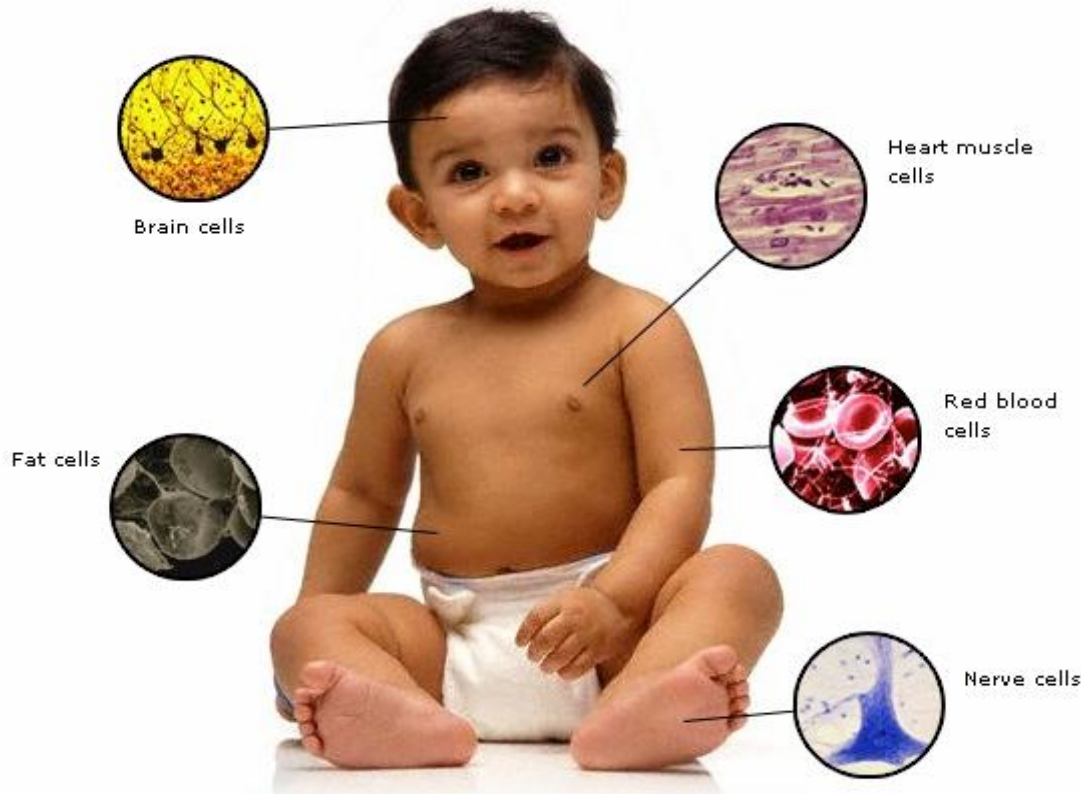
Genoma: sequencia de DNA de todos os cromossomos de um organismo

Evolução das descobertas da origem genética de doenças humanas

Pace of disease gene Discovery (1981-2000)



Onde estão os genes?

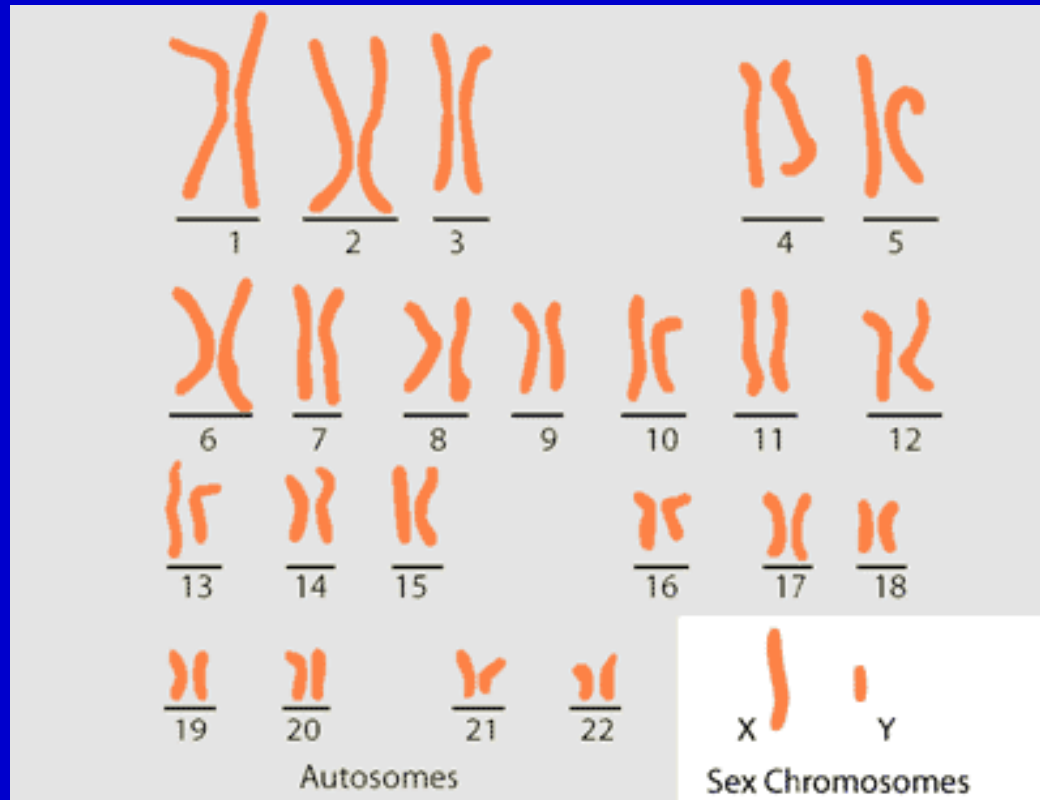


© Elsevier. Nussbaum et al: Thompson and Thompson's Genetics in Medicine 7e - www.studentconsult.com

Cromossomos nucleares e mitocondriais

Cromossomos Humanos

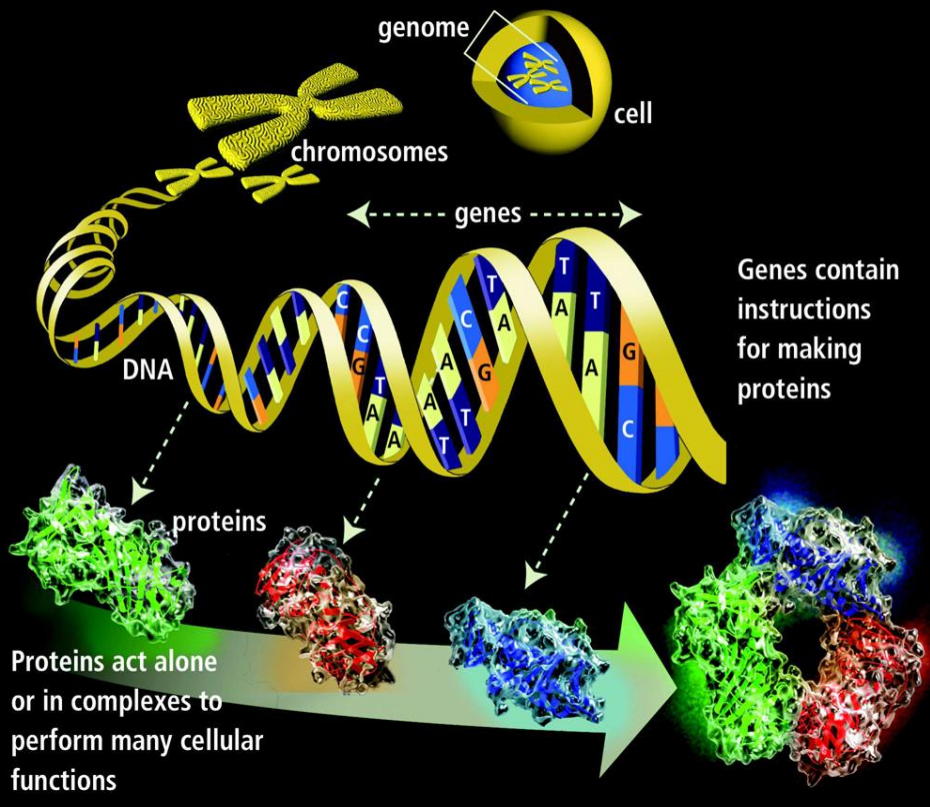
Cariótipo Humano



The Human Genome

The Human Genome is the total of the genetic information that is held in each human cell. It is usually made up of 46 chromosomes: 22 pairs of autosomes and 1 pair of sex chromosomes, which are usually X and X for females and X and Y for males.

DNA a molécula da vida



Cada célula tem:

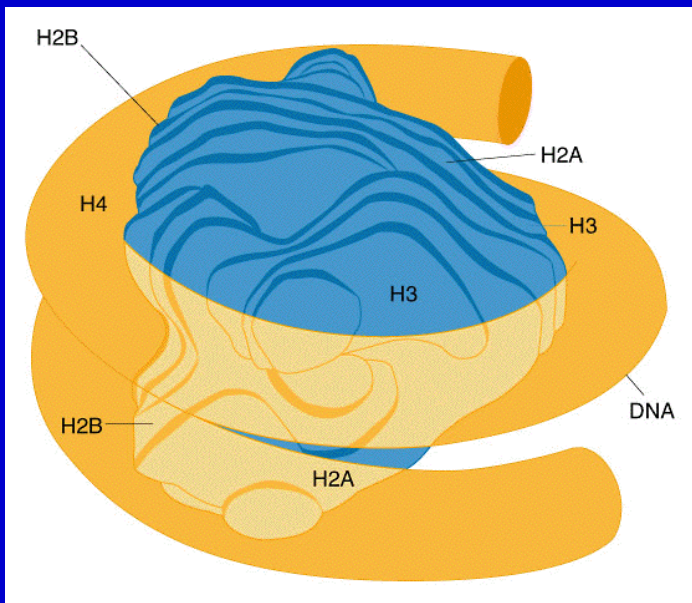
46 cromossomos humanos

2 metros de DNA

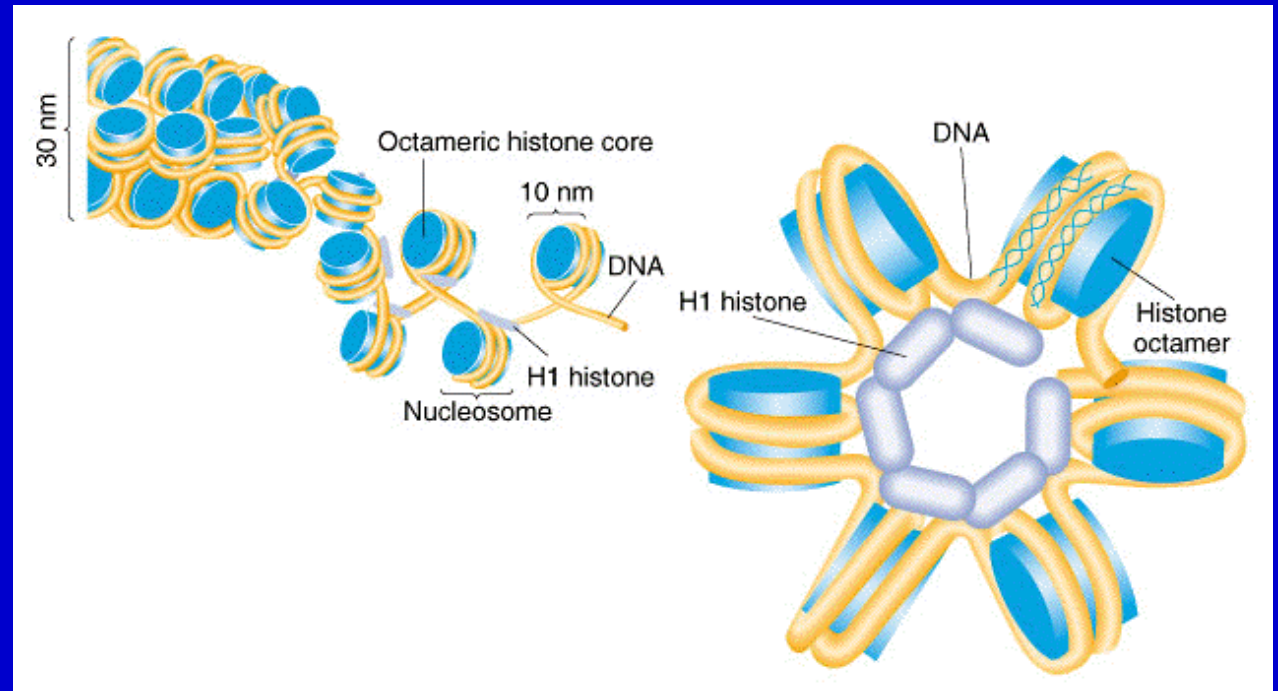
3 bilhões de subunidades de DNA (bases A, C, T, G)

~ 25.000 genes que codificam para RNA e proteínas responsáveis pelas funções biológicas

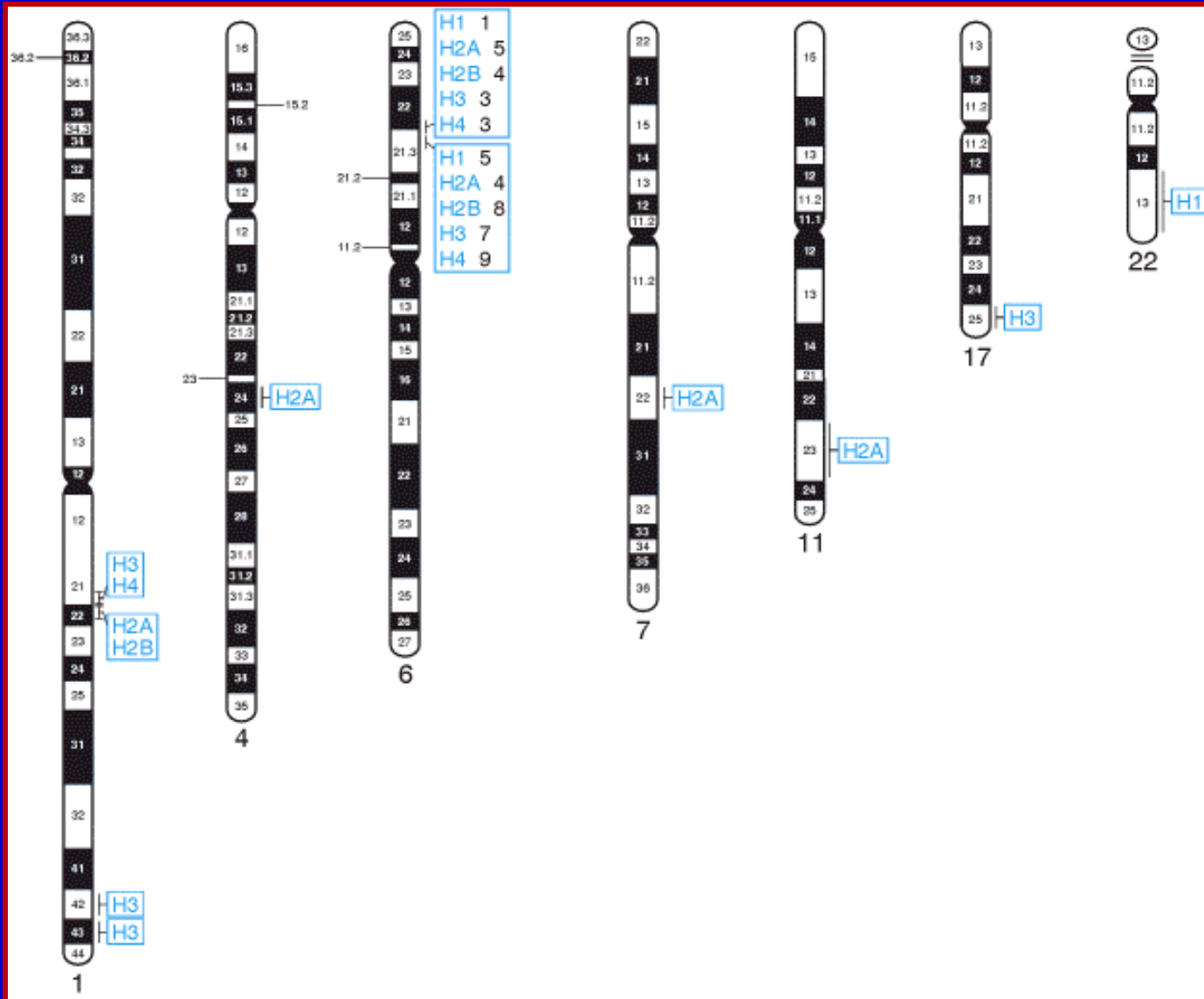
Níveis de empacotamento do DNA



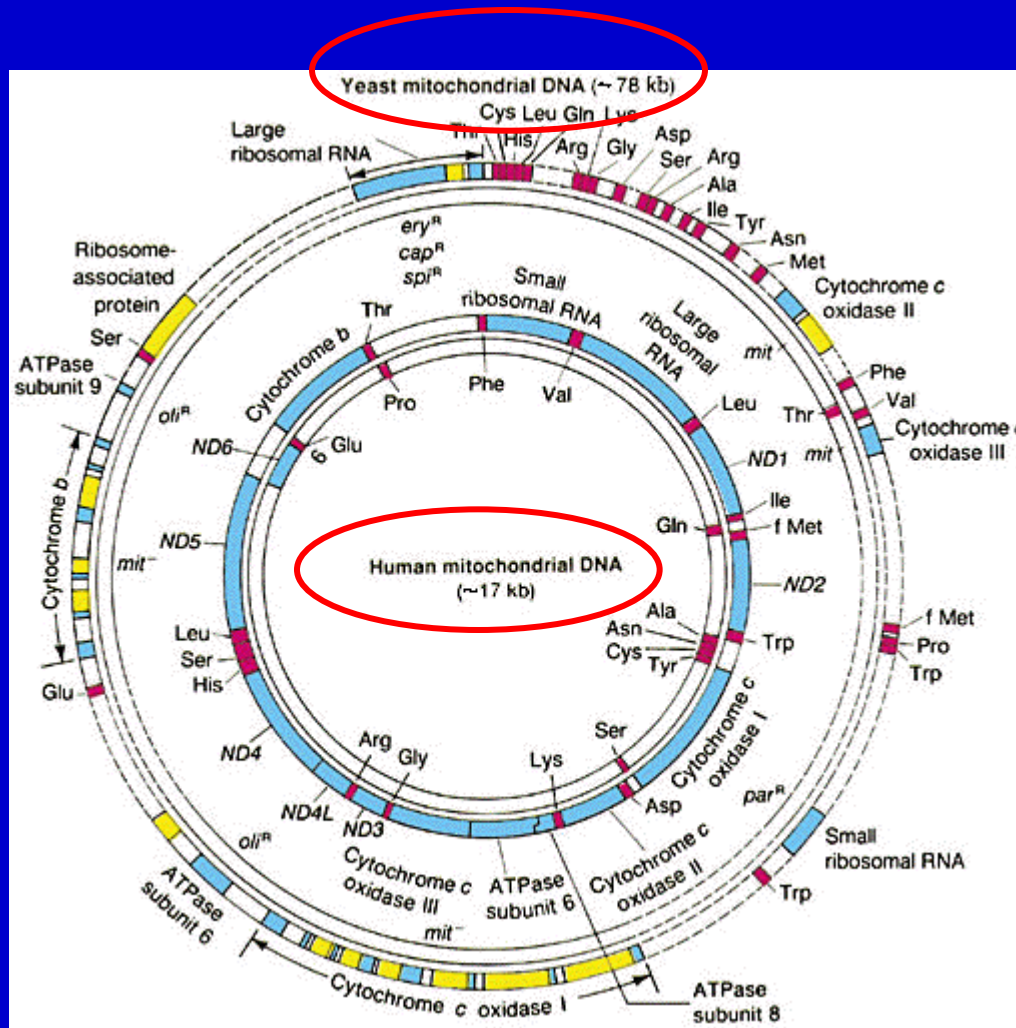
Nucleossomo



Genes Humanos envolvidos na síntese de histonas



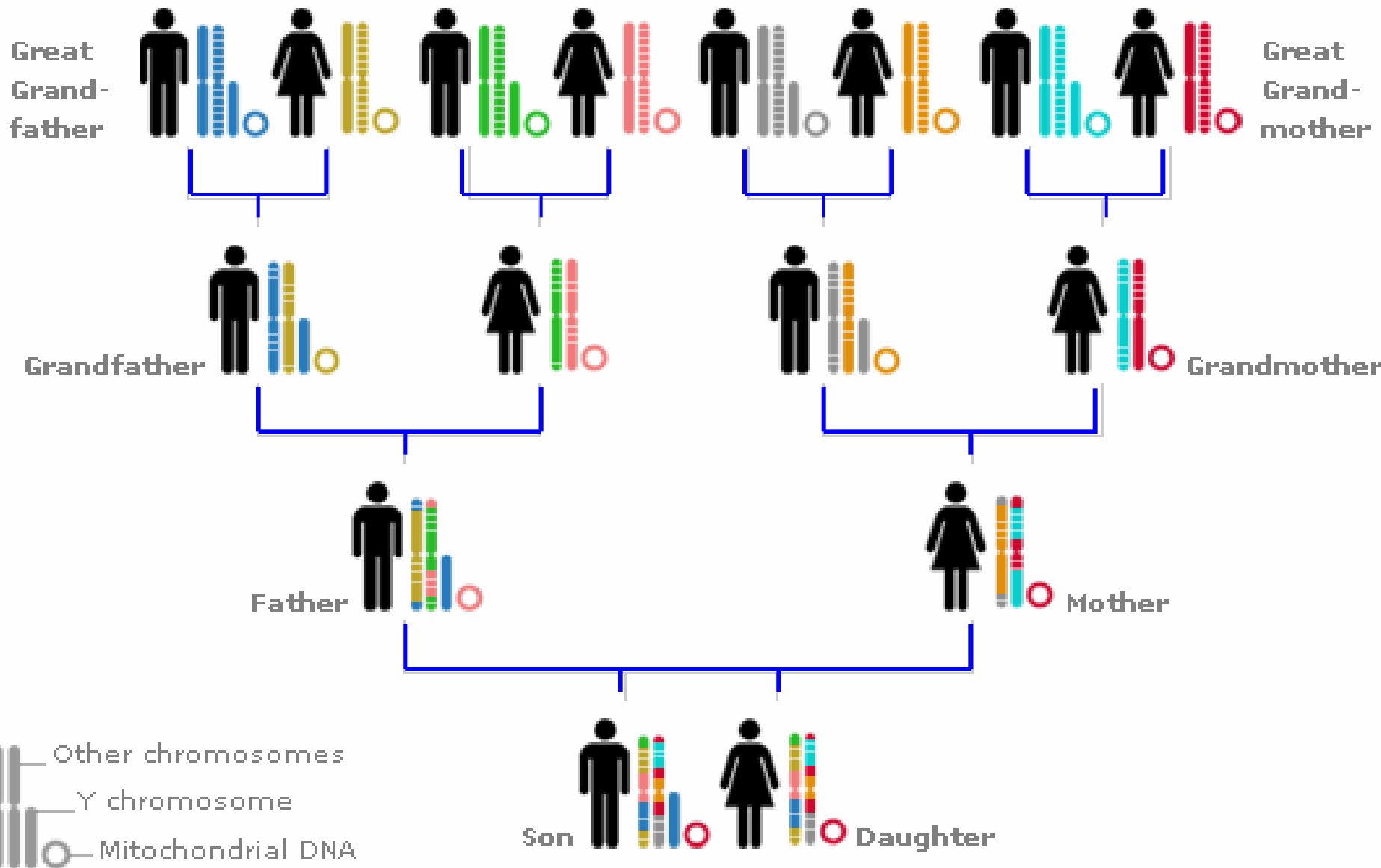
DNA mitochondrial



Codifica para algumas proteínas da cadeia de transporte de elétrons e tRNA, rRNA para a síntese protéica mitocondrial

37 genes mitocondriais humanos

Herança mitocondrial e do cromossomo Y



Principais tipos de doenças genéticas

- **Monogênica:** anemia falciforme, fibrose cística, albinismo
- **Multifatorial (combinação gênica que gera predisposição):** hipertensão
- **Transtornos cromossômicos:** síndrome de Down

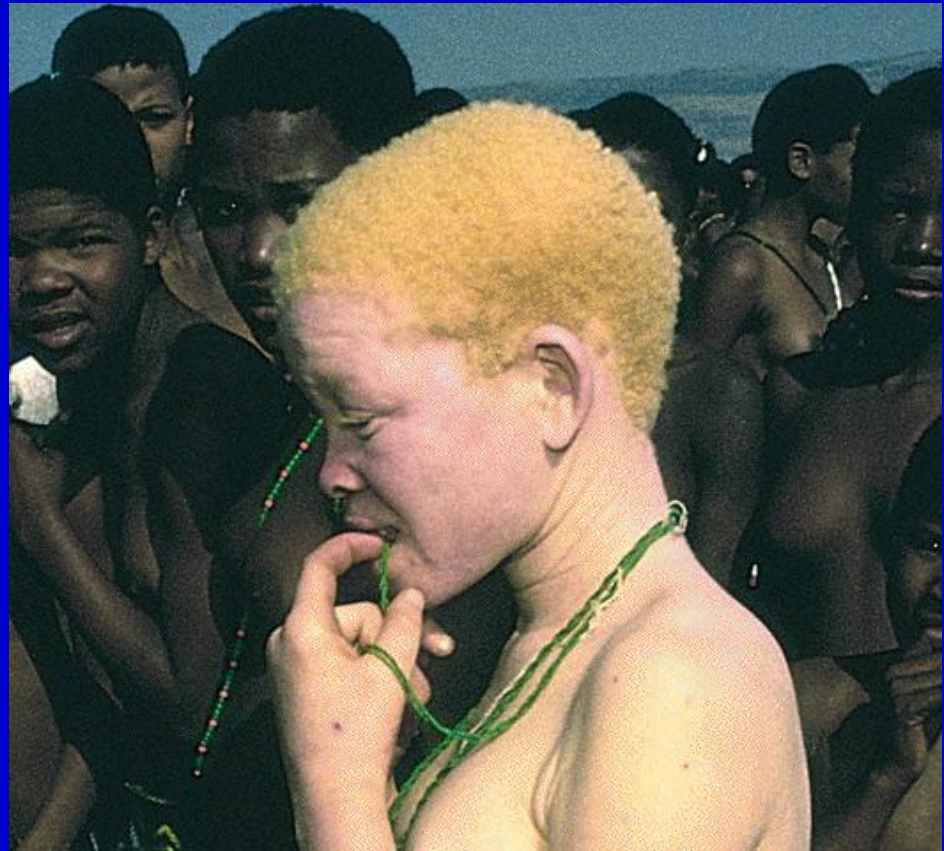
Exemplos de doenças monogênicas



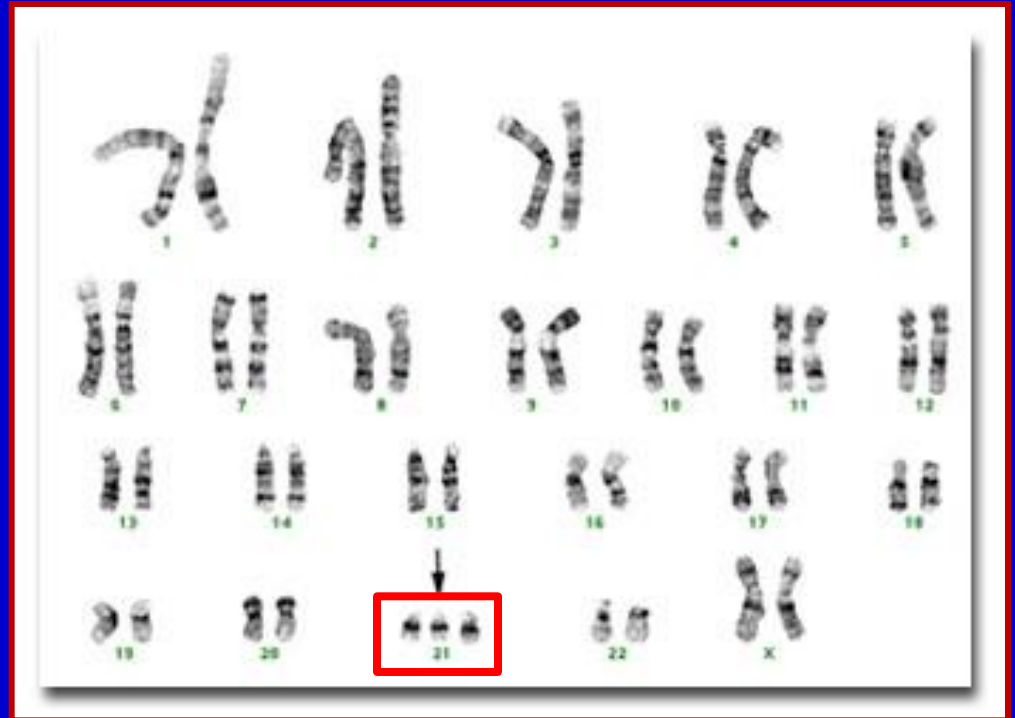
Acondroplasia:

substituição de uma glicina por arginina no domínio transmembranar do receptor do fator de crescimento fibroblástico 3

Albinismo: ausência ou defeito de uma enzima envolvida na produção de melanina.

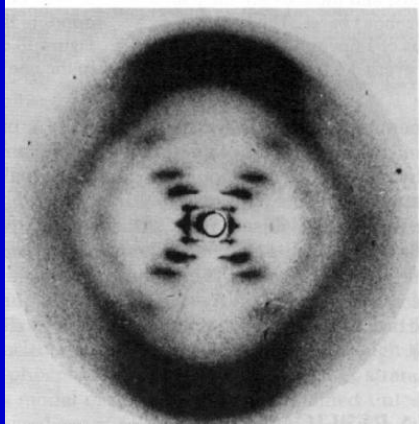


Exemplo de transtorno cromossômico



Síndrome de Down

Características do DNA



Difração de raio X do DNA

Table 11-1 Molar Properties of Bases* in DNAs from Various Sources

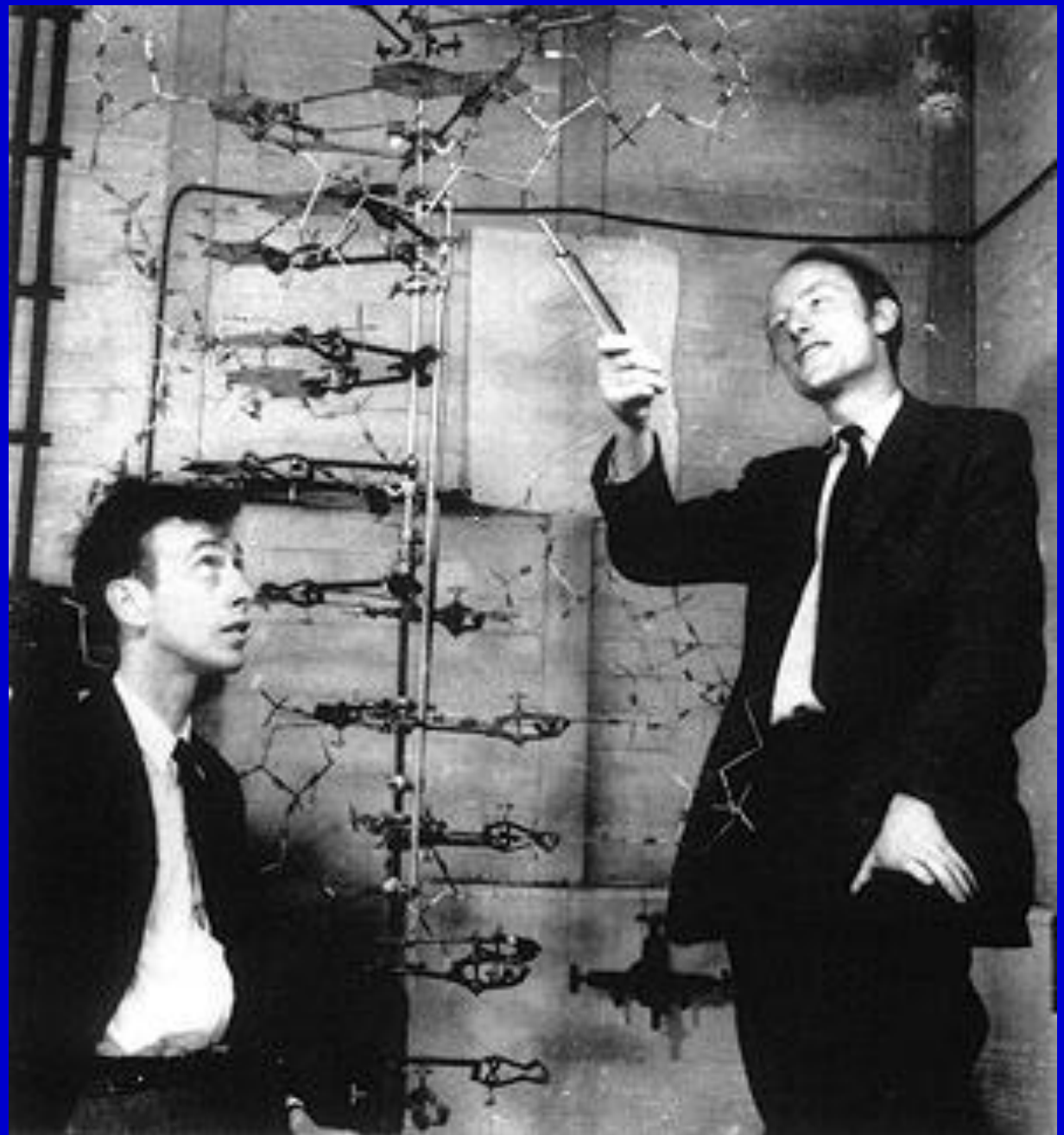
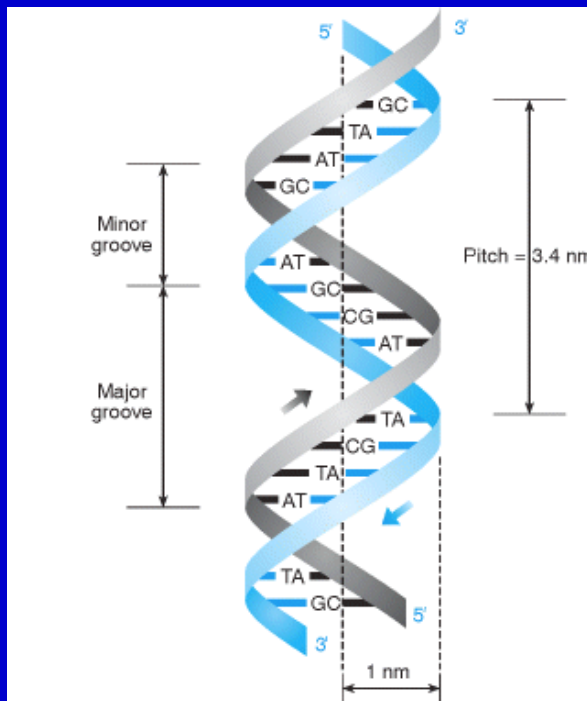
Organism	Tissue	Adenine	Thymine	Guanine	Cytosine
<i>Escherichia coli</i> (K12)	—	26.0	23.9	24.9	25.2
<i>Diplococcus pneumoniae</i>	—	29.8	31.6	20.5	18.0
<i>Mycobacterium tuberculosis</i>	—	15.1	14.6	34.9	35.4
Yeast	—	31.3	32.9	18.7	17.1
<i>Paracentrotus lividus</i> (sea urchin)	Sperm	32.8	32.1	17.7	18.4
Herring	Sperm	27.8	27.5	22.2	22.6
Rat	Bone marrow	28.6	28.4	21.4	21.5
Human	Thymus	30.9	29.4	19.9	19.8
Human	Liver	30.3	30.3	19.5	19.9
Human	Sperm	30.7	31.2	19.3	18.8

*Defined as moles of nitrogenous constituents per 100 g-atoms phosphate in hydrolysate.
SOURCE: E. Chargaff and J. Davidson, eds., *The Nucleic Acids*. Academic Press, 1955.

Razão das bases do DNA

Modelo da dupla hélice do DNA

Watson e Crick 1953



equipment, and to Dr. G. E. R. Deacon and the captain and officers of R.R.S. *Discovery II* for their part in making the observations.

¹ Young, F. B., Gerrard, H., and Jevons, W., *Phil. Mag.*, **40**, 149 (1920).

² Longuet-Higgins, M. S., *Mon. Not. Roy. Astro. Soc., Geophys. Supp.*, **5**, 285 (1949).

³ Von Arx, W. S., *Woods Hole Papers in Phys. Oceanog. Meteor.*, **11** (3) (1950).

⁴ Ekman, V. W., *Arkiv. Mat. Astron. Fysik. (Stockholm)*, **2** (11) (1905).

MOLECULAR STRUCTURE OF NUCLEIC ACIDS

A Structure for Deoxyribose Nucleic Acid

WE wish to suggest a structure for the salt of deoxyribose nucleic acid (D.N.A.). This structure has novel features which are of considerable biological interest.

A structure for nucleic acid has already been proposed by Pauling and Corey¹. They kindly made their manuscript available to us in advance of

is a residue on each chain every 10 Å. We have assumed that adjacent residues in the same structure repeats after 10 residues, after 34 Å. The distance from the fibre axis is 10 Å. At the outside, cations have easy access to the structure.

The structure is an open one and is rather high. At lower water content we expect the bases to tilt so that the structure becomes more compact.

The novel feature of the structure is in which the two chains are antiparallel. The purine and pyrimidine bases are perpendicular to the fibre axis. They are together in pairs, a single base on each chain, hydrogen-bonded to a single base on the other chain, so that the two lie side by side. The z-co-ordinates. One of the pairs is a purine, the other a pyrimidine for each pair. The hydrogen bonds are made as follows: purine position 1 to pyrimidine position 1, purine position 6 to pyrimidine position 6.

If it is assumed that the structure is the most compact



This figure is purely diagrammatic. The two ribbons symbolize the two phosphate—sugar chains, and the horizontal rods the pairs of bases holding the chains together. The vertical line marks the fibre axis

April 25, 1953: James Watson e Francis Crick publicaram o clássico paper que descreve pela primeira vez a estrutura de dupla hélice do DNA. Eles notaram que a estrutura sugeria o mecanismo de replicação do material genético.

Premio Nobel em Fisiologia e Medicina em 1962



Francis Harry
Compton Crick



James Dewey
Watson



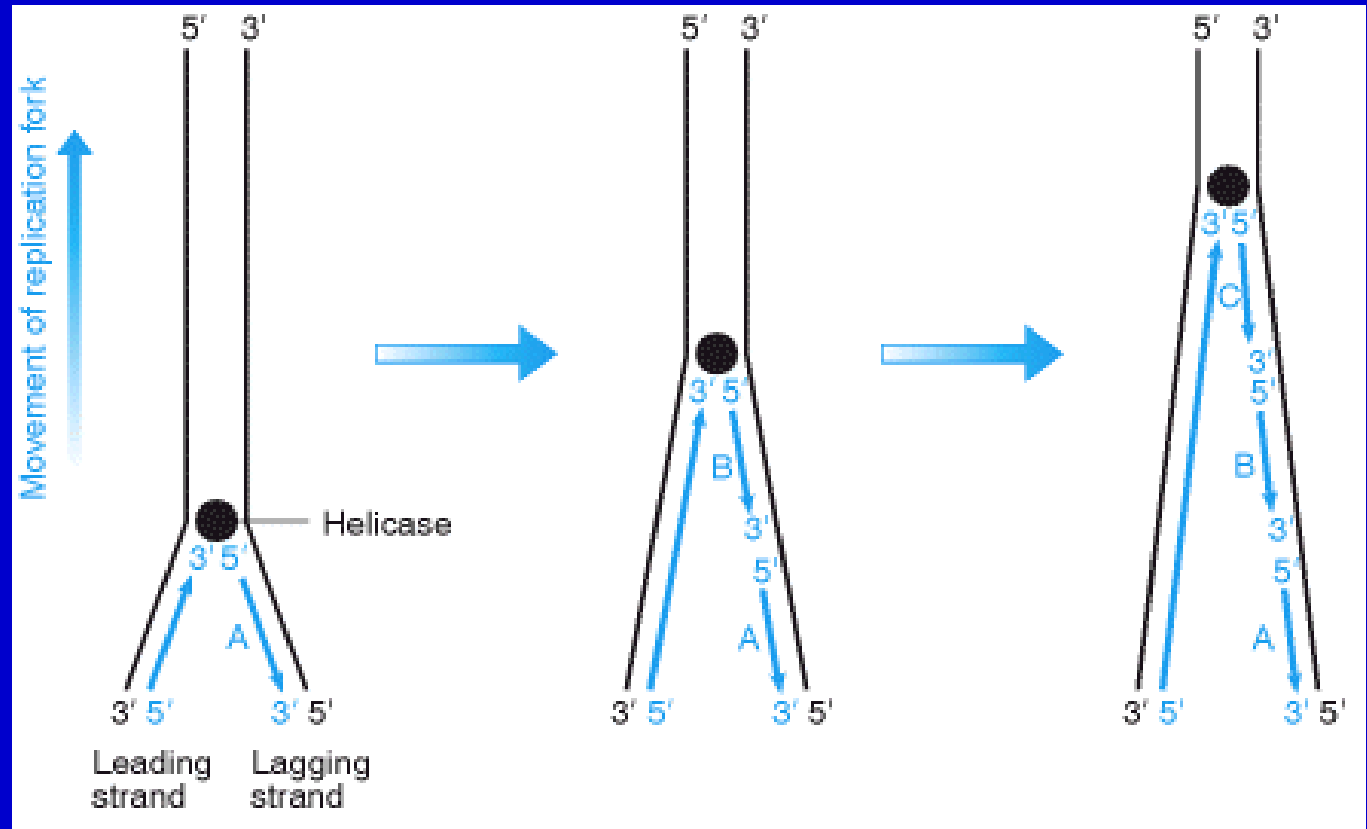
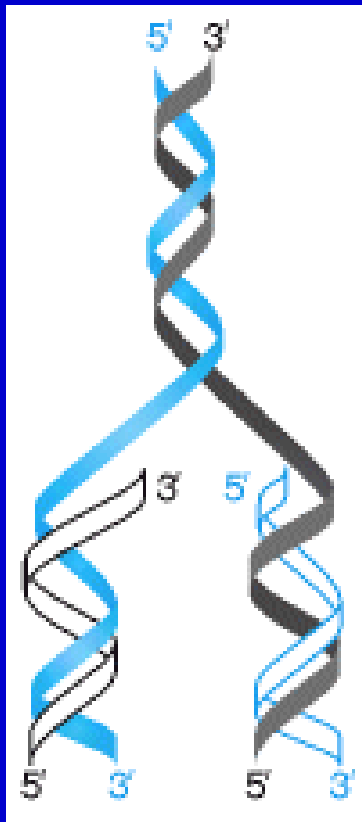
Maurice Hugh
Frederick Wilkins

The Nobel Prize in Physiology or Medicine 1962 was awarded jointly to Francis Harry Compton Crick, James Dewey Watson and Maurice Hugh Frederick Wilkins "for their discoveries concerning the molecular structure of nucleic acids and its significance for information transfer in living material".

Quais são as funções do DNA?

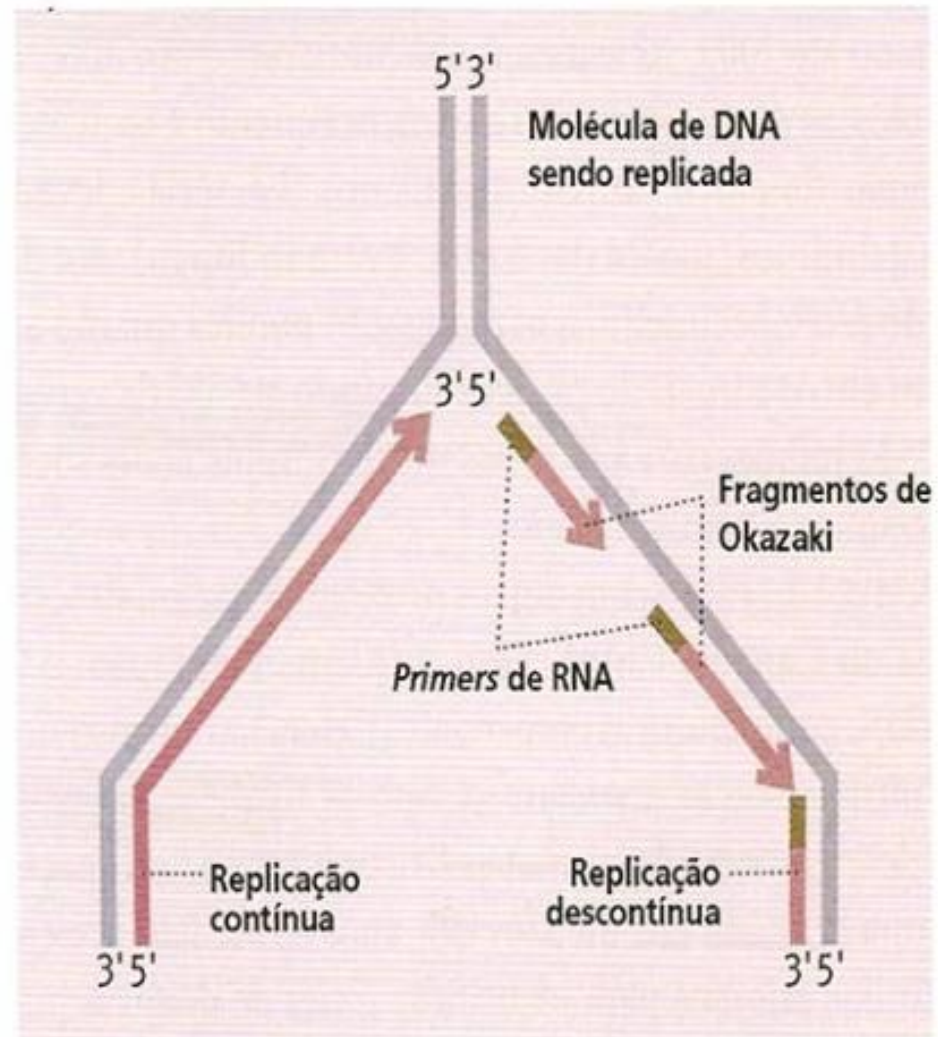
- Replicação: preservação da espécie por que é o material hereditário
- Transcrição: síntese de RNA

A replicação do DNA é semiconservativa e a síntese das fitas de DNA é semidescontínua

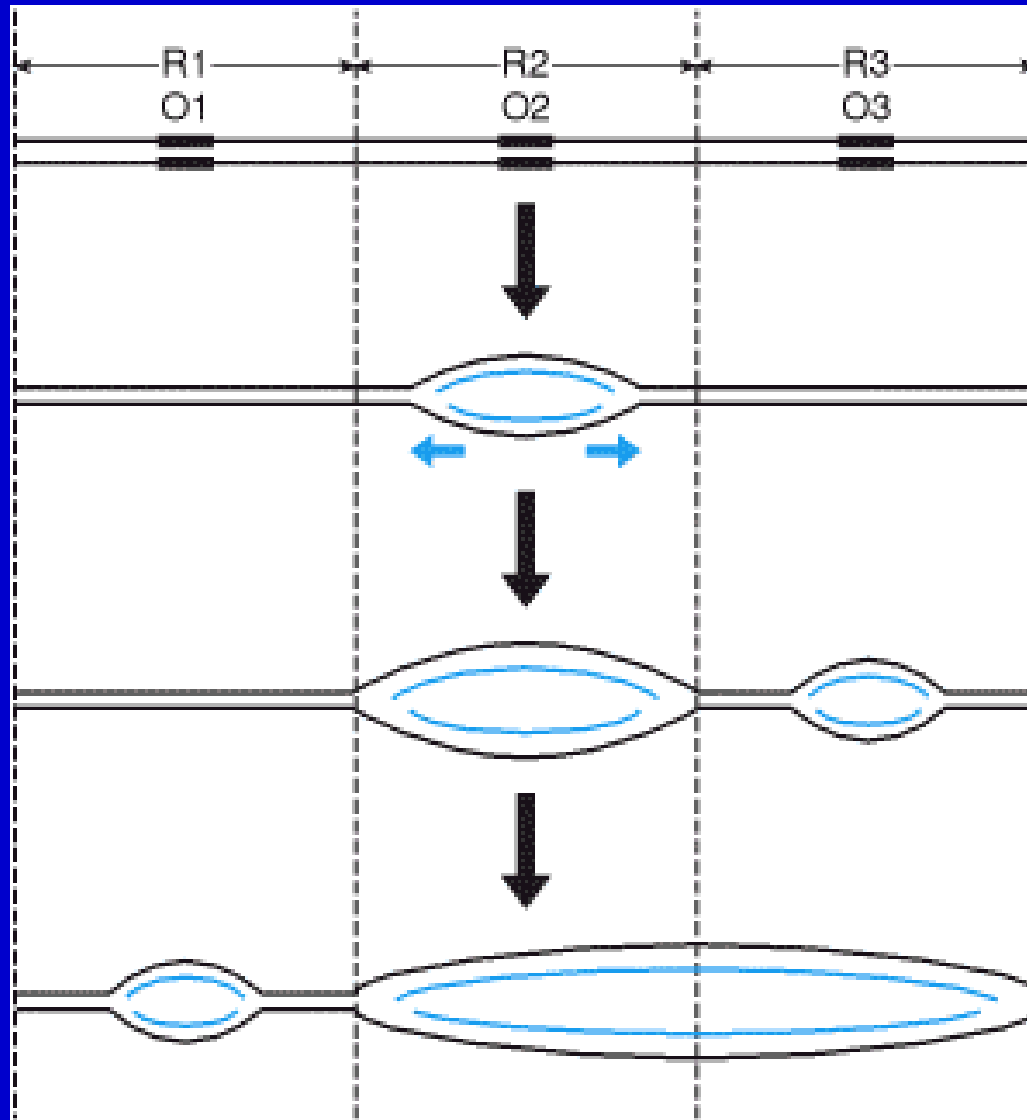


Replicação do DNA e os fragmentos de Okazaki

Uma fita contínua e uma fita descontínua



Os cromossomos dos organismos complexos têm muitas origens de replicação



- Como os genes estão distribuídos ao longo dos cromossomos ?

Média de tamanho de um gene humano: 27 kb

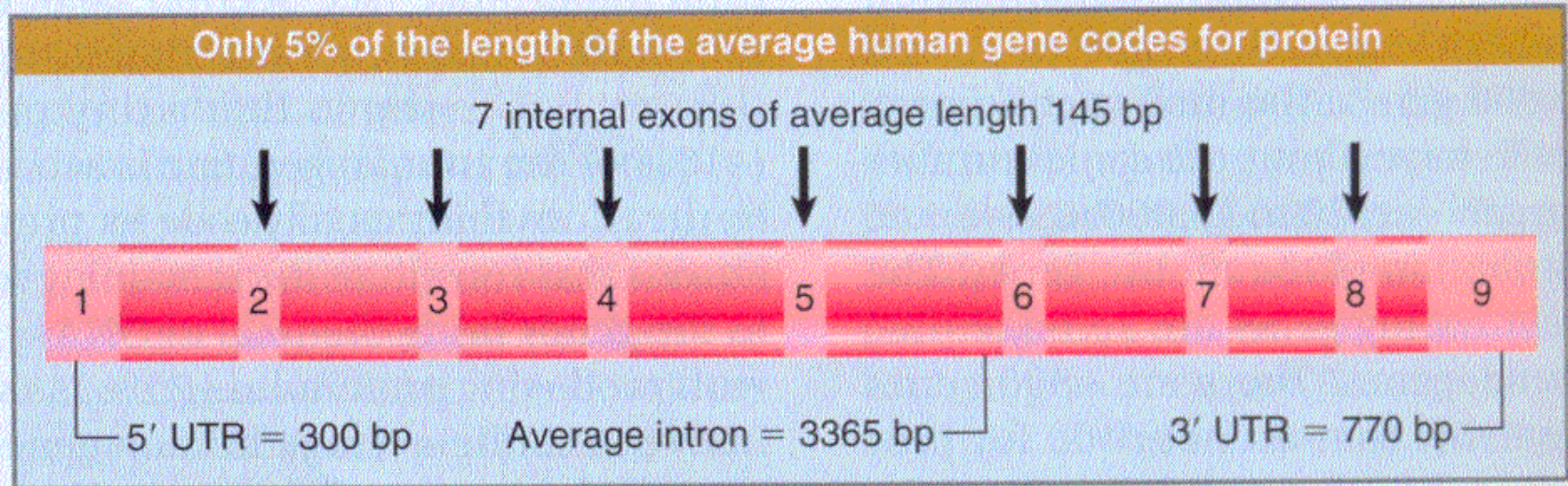
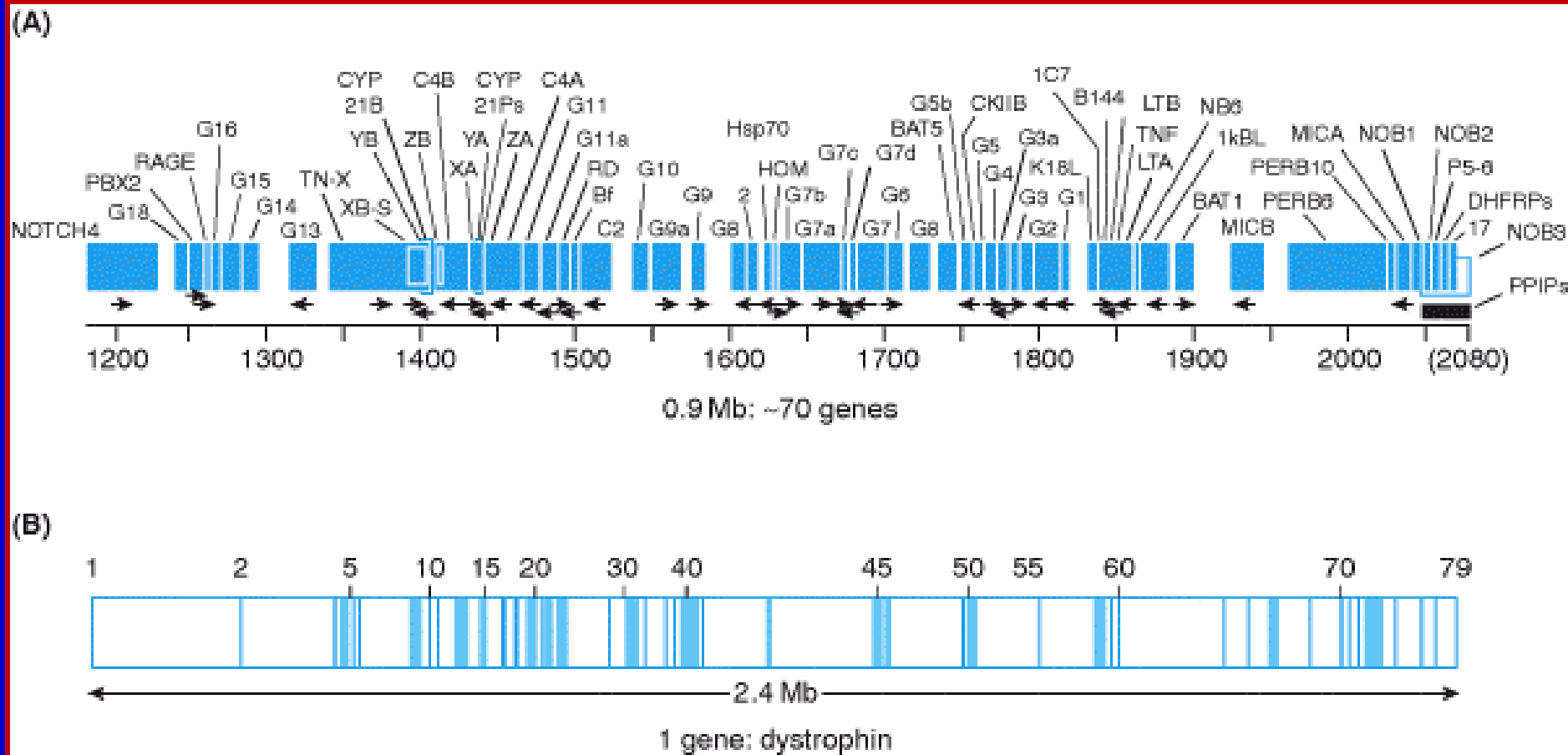


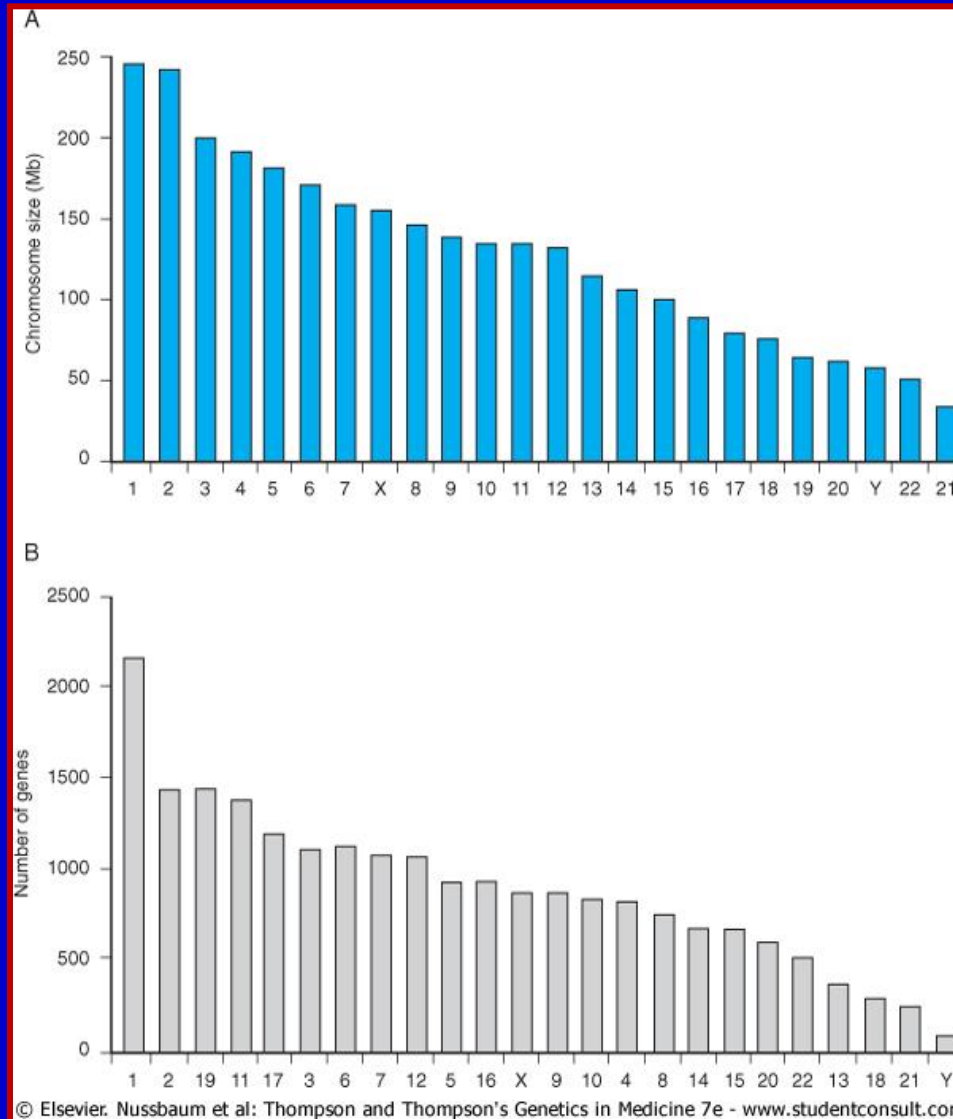
FIGURE 5.12 The average human gene is 27 kb long and has nine exons, usually comprising two longer exons at each end and seven internal exons. The UTRs in the terminal exons are the untranslated (noncoding) regions at each end of the gene. (This is based on the average. Some genes are extremely long, which makes the median length 14 kb with seven exons.)

Contraste entre as densidades gênicas humanas



Não há relação entre o tamanho do cromossomo e o número de genes no genoma humano

Tamanho dos cromossomos



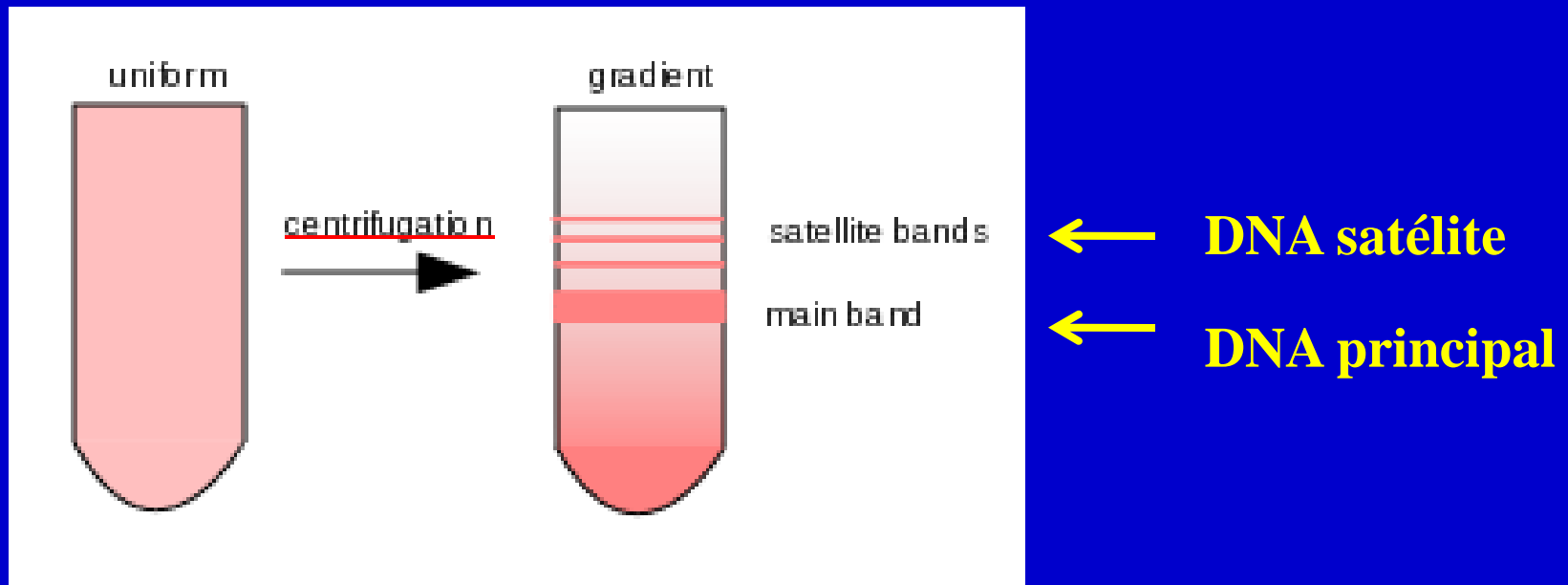
Número de genes por cromossomo



Sequências de DNA repetitivo

- Família Alu: tamanho 300 nucleotídeos.
Compõem 10 % do genoma humano.
- Line: tamanho 6 Kb.
Ocupa 20 % do genoma humano.
- Regiões centroméricas e teloméricas.

50% do genoma humano é constituído de DNA repetitivo (DNA satélite)



Número de minisatélites (10 a 100 pb) difere entre os indivíduos e entre os alelos de um mesmo indivíduo (heterozogoto).

DNA fingerprinting

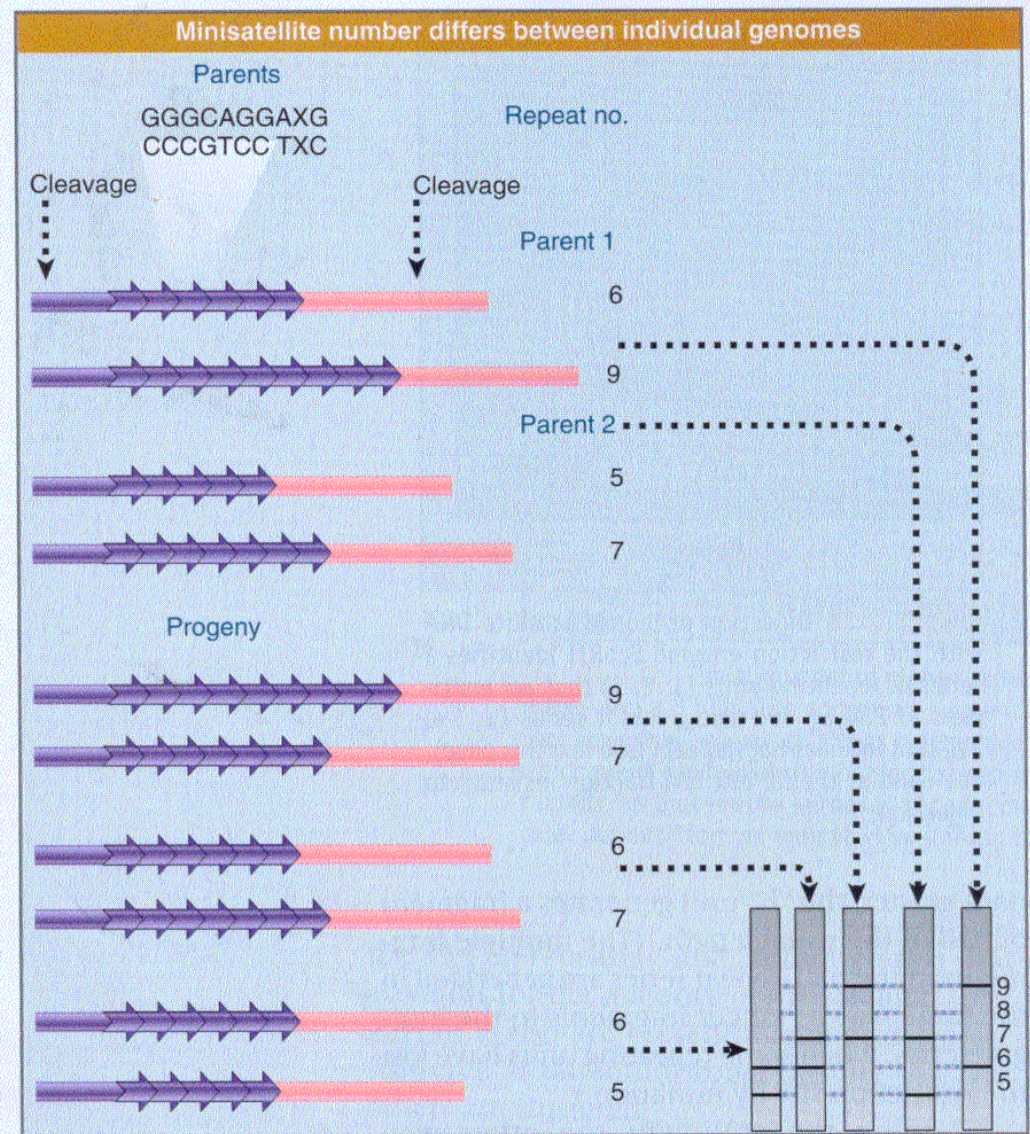


FIGURE 6.29 Alleles may differ in the number of repeats at a minisatellite locus, so that cleavage on either side generates restriction fragments that differ in length. By using a minisatellite with alleles that differ between parents, the pattern of inheritance can be followed.

Menos que 2 % do genoma humano codifica proteína.

1% of the human genome codes for protein

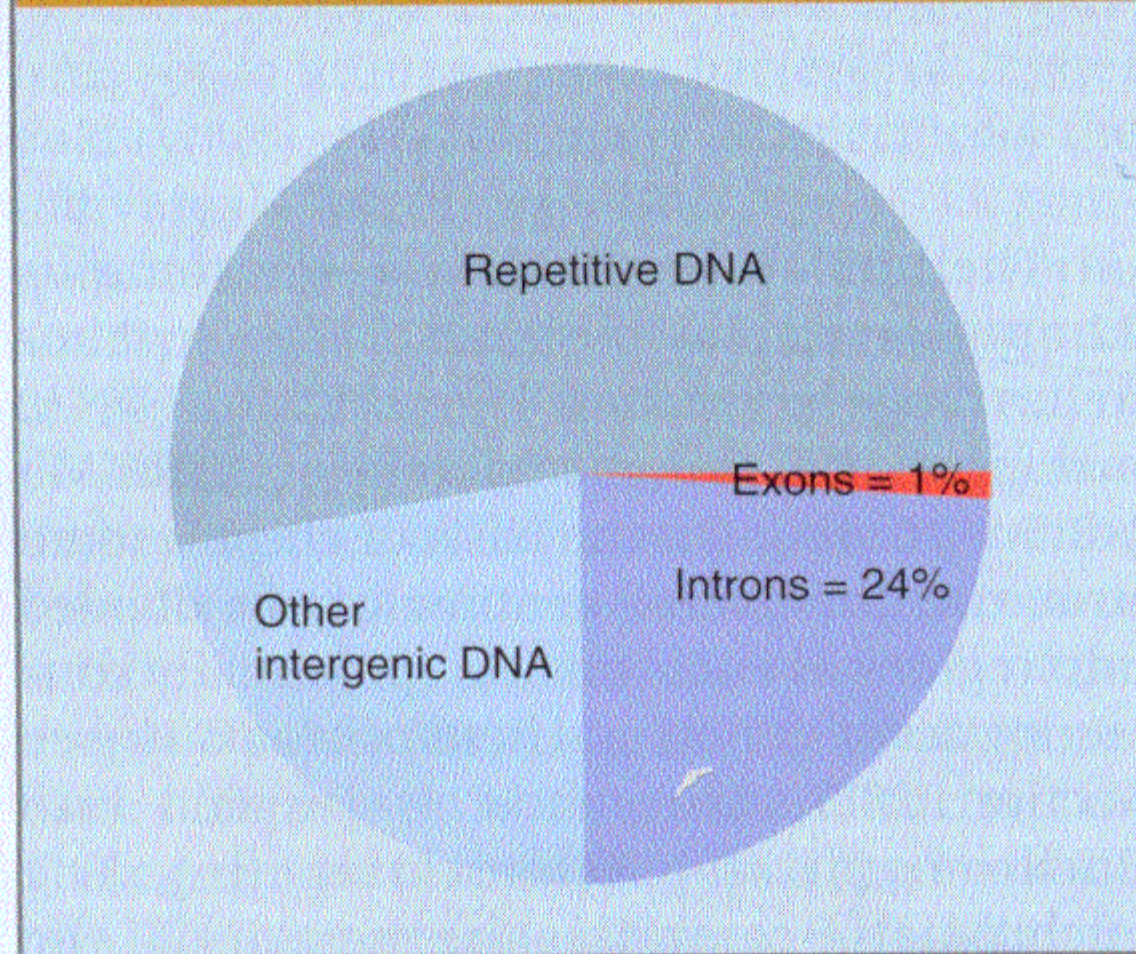
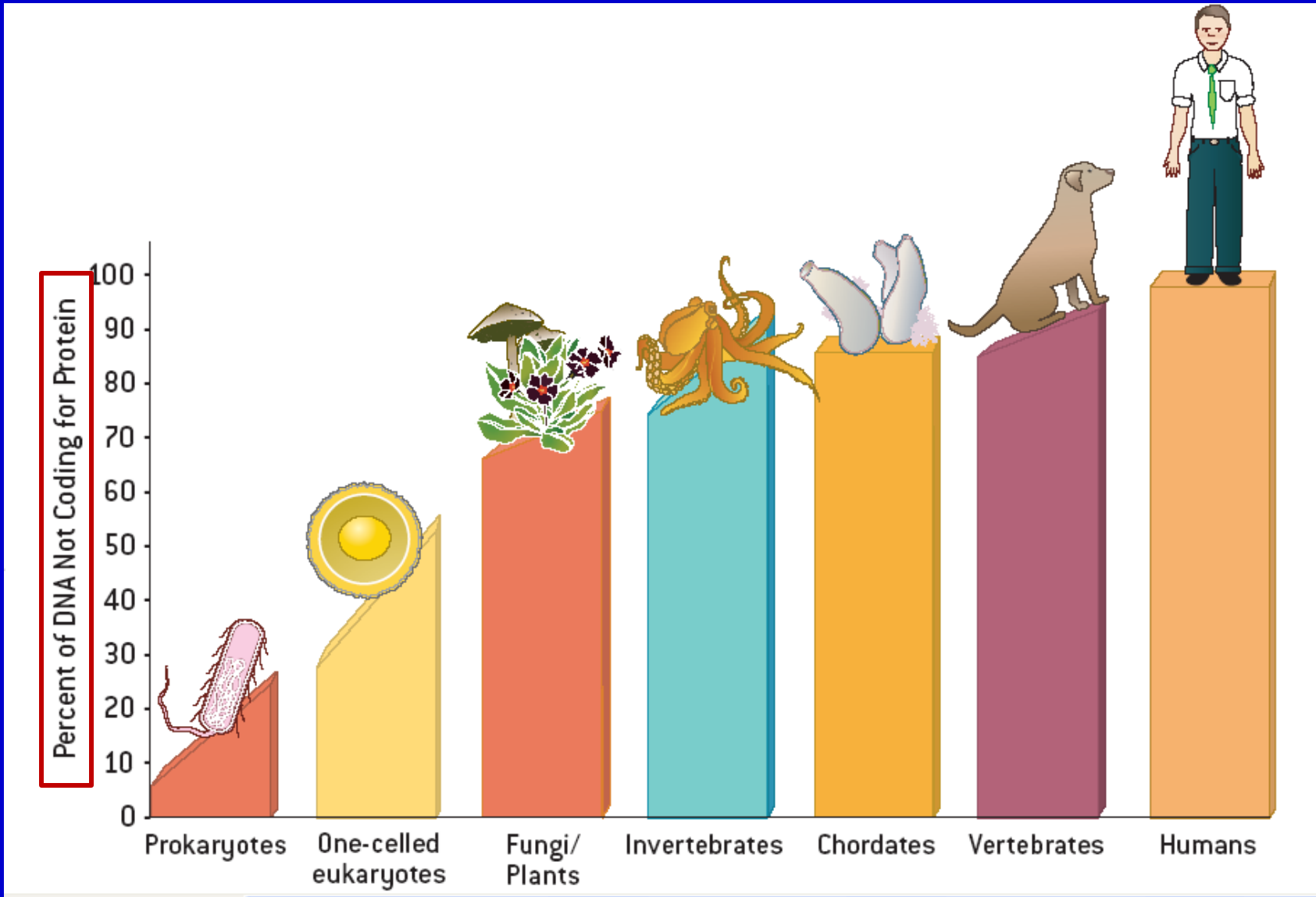


FIGURE 5.11 Genes occupy 25% of the human genome, but protein-coding sequences are only a tiny part of this fraction.

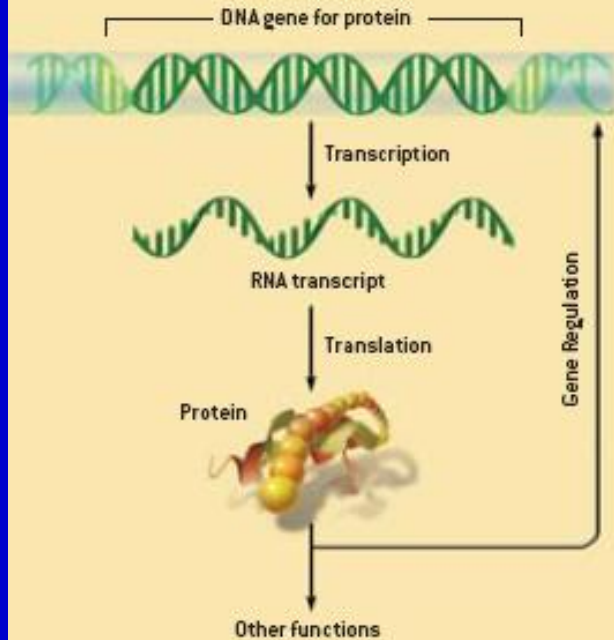
A quantidade de DNA não codificador está relacionada a complexidade dos organismos



Visão tradicional da atividade gênica

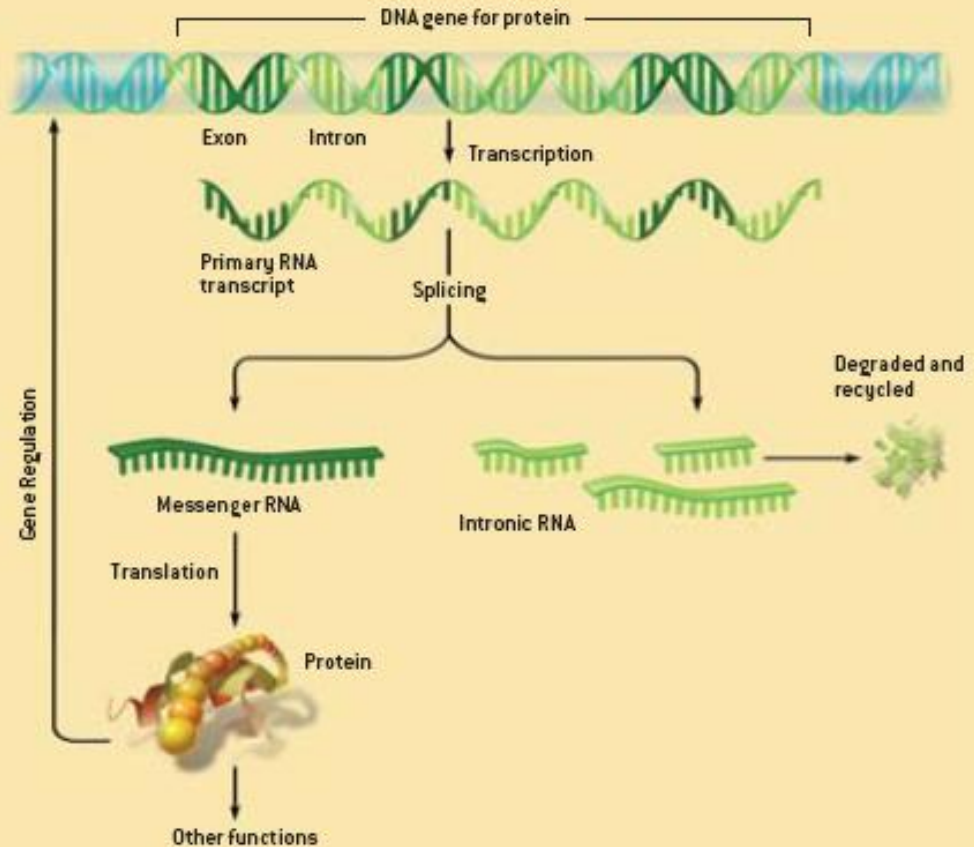
GENE ACTIVITY IN PROKARYOTES

Prokaryotes (bacteria and other simple cells) have DNA that consists almost entirely of protein-coding genes. When those genes are active, they give rise to RNA transcripts that are immediately translated into proteins, which in turn regulate genetic activity and provide other functions.



TRADITIONAL VIEW OF GENE ACTIVITY IN EUKARYOTES

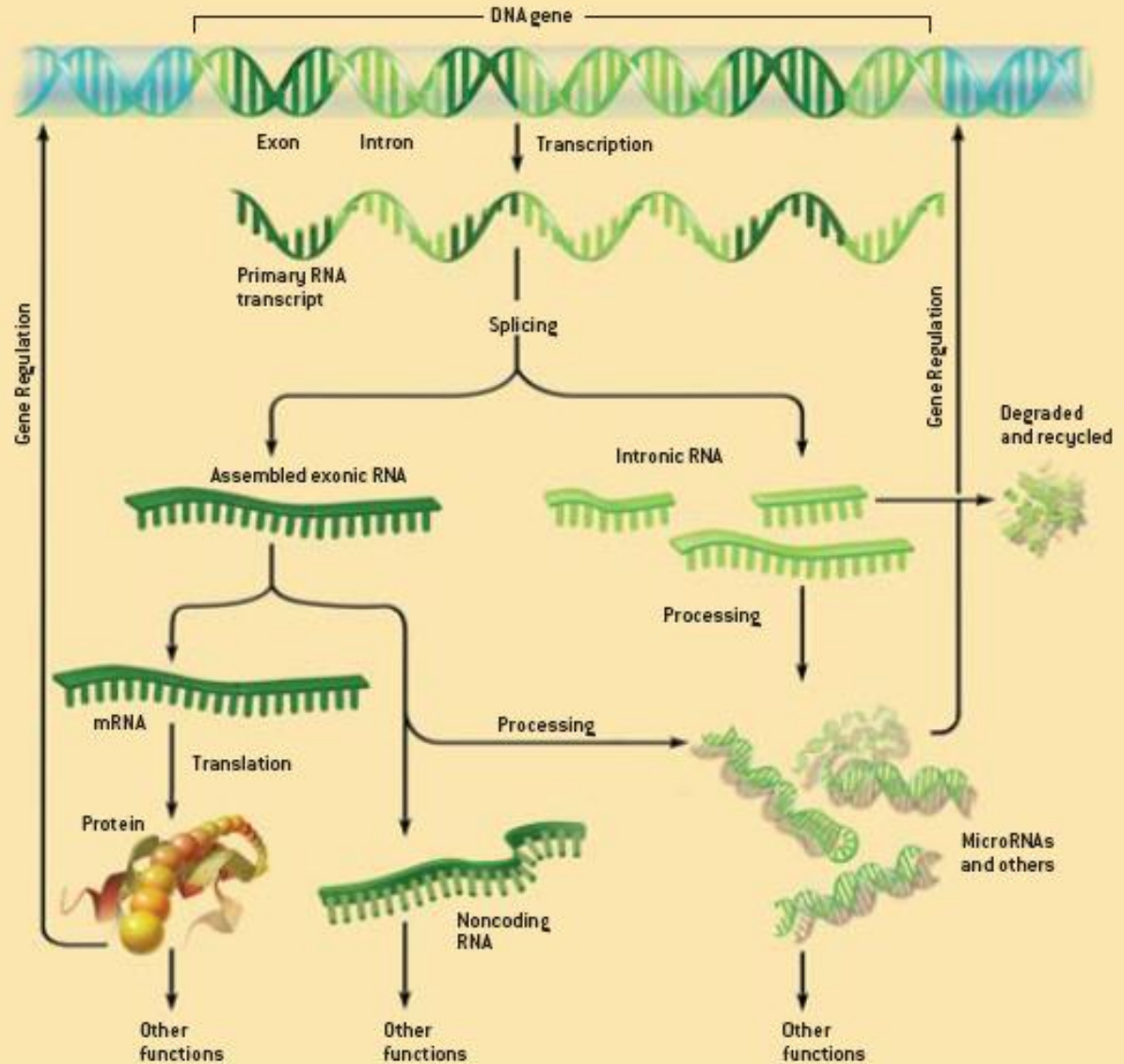
In the DNA of eukaryotes (complex organisms), individual genes comprise "exon" sequences that code for segments of protein separated by noncoding "intron" sequences. When a gene is active, it is entirely transcribed as RNA, but then the intronic RNA is spliced out and the exonic RNA is assembled as messenger RNA. The cell translates the messenger RNA into protein while breaking down and recycling the intronic RNA, which serves no purpose.



Visão atual da atividade gênica

NEW VIEW OF GENE ACTIVITY IN EUKARYOTES

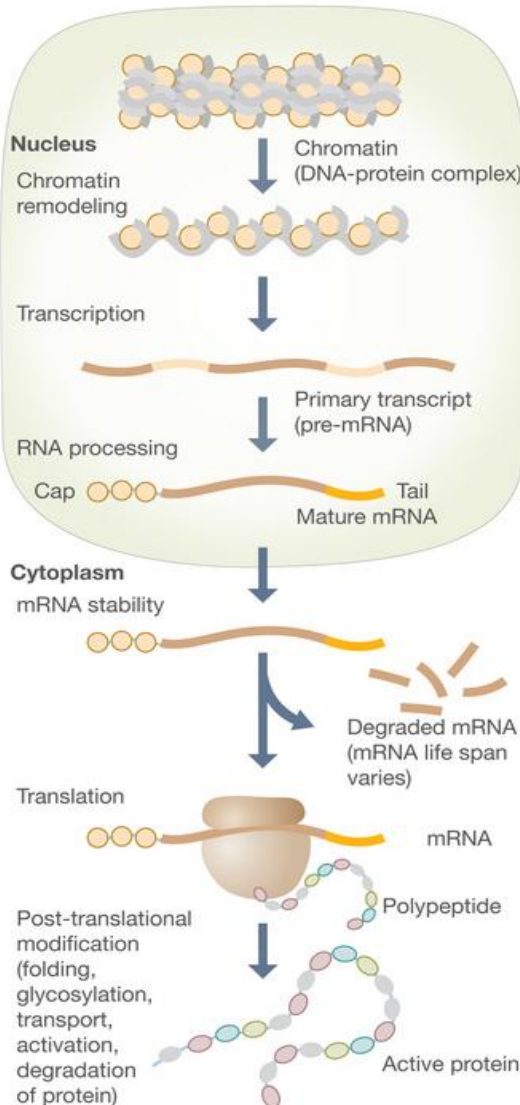
Some of the intronic RNA and even some of the assembled exonic RNA may play a direct regulatory role by interacting with the DNA, other RNA molecules or proteins. By modifying protein production at various levels, these noncoding RNAs may superimpose additional genetic instructions on a cell.



Papel do RNA não codificador (ncRNA) na expressão gênica

- Desenvolvimento embrionário
- Diferenciação celular
- Estabilidade genômica
- Estabilidade do mRNA
- Altera a estrutura da cromatina

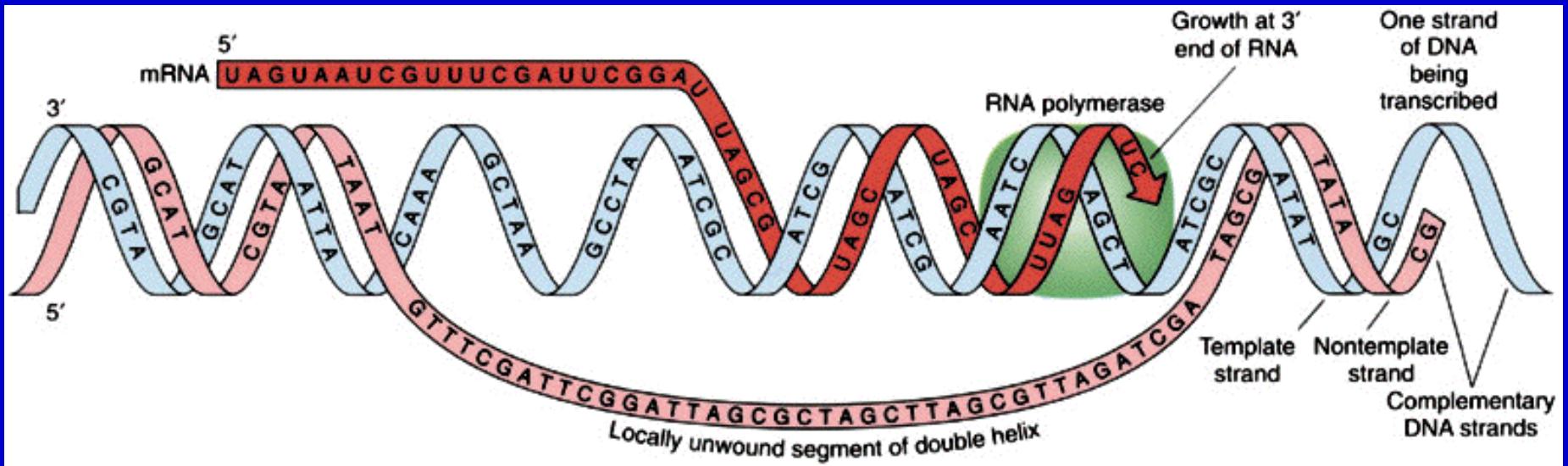
**Regulation of Gene Expression in Eukaryotes
May Take Place at Several Different Stages**



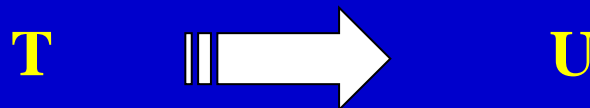
**A regulação da
expressão gênica
em eucariotos
pode
ocorrer em
vários estágios**



A síntese do mRNA é fiel ao DNA

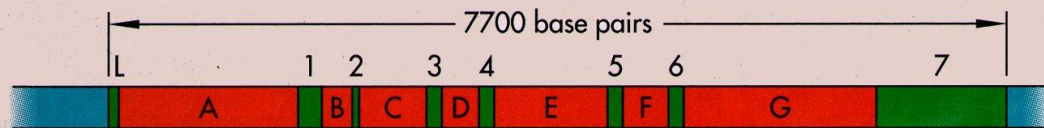
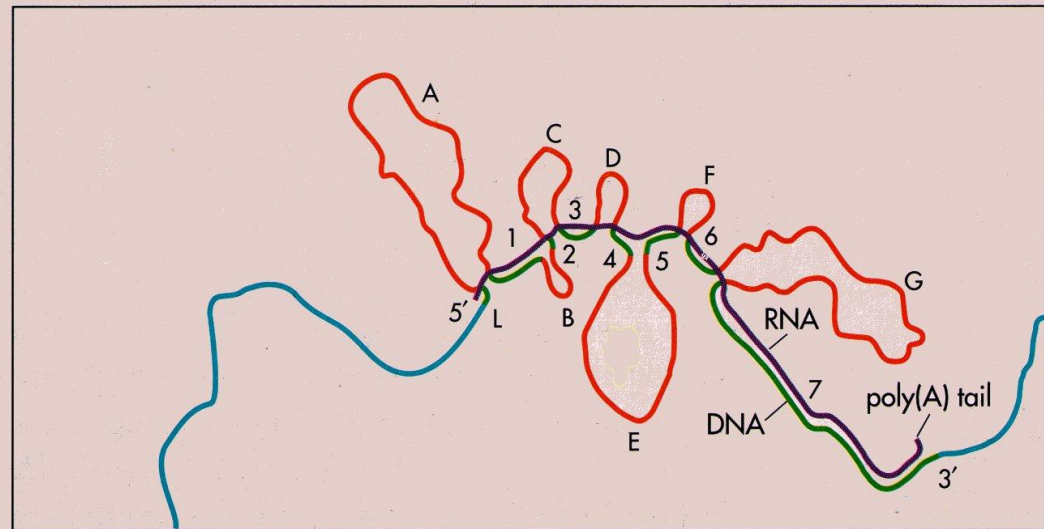
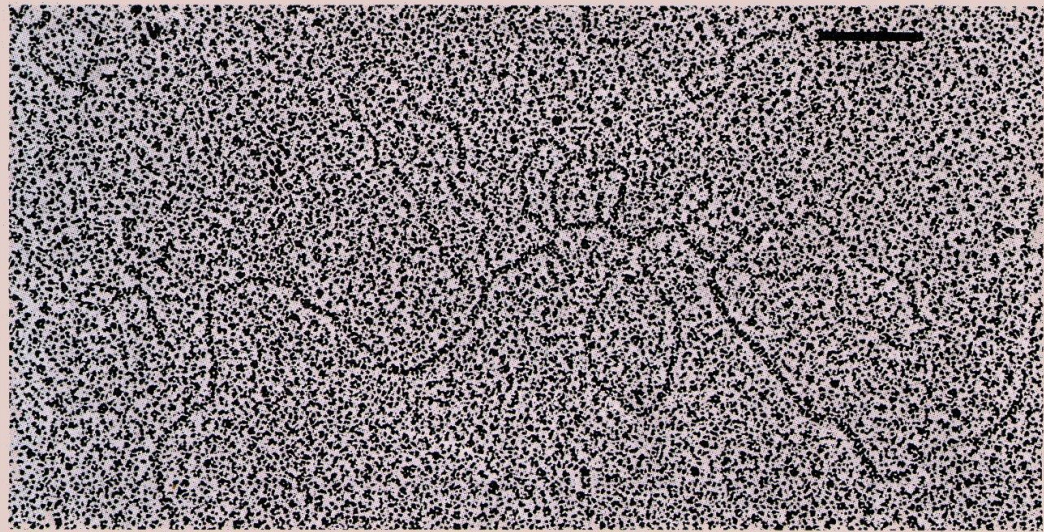


Nontemplate strand 5' - CTGCCATTGTCAGACATGTATACCCCGTACGTCTTCCCGAGCGAAAACGATCTGCGCTGC - 3' } DNA
 Template strand 3' - GACGGTAACAGTCTGTACATATGGGGCATGCAGAAGGGCTCGCTTTTGCTAGACGCGACG - 5' }
 5' - CUGCCAUUGUCAGACAUGUAUACCCCGUACGUCUUCGCCGAGCGAAAACGAUCUGCGCUGC - 3' mRNA

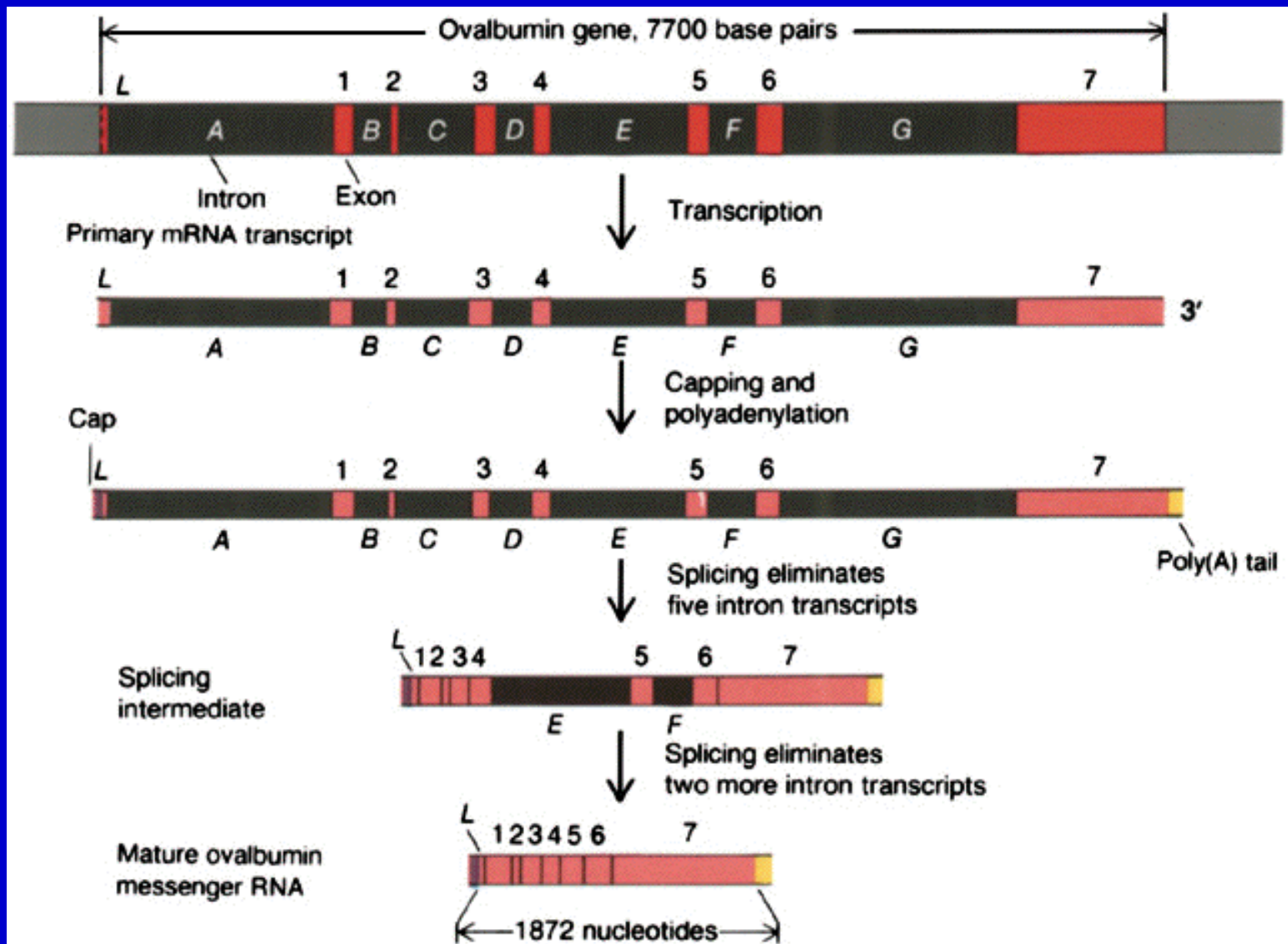


Descoberta do
Processamento
do
RNA
(Intron e
Exon)

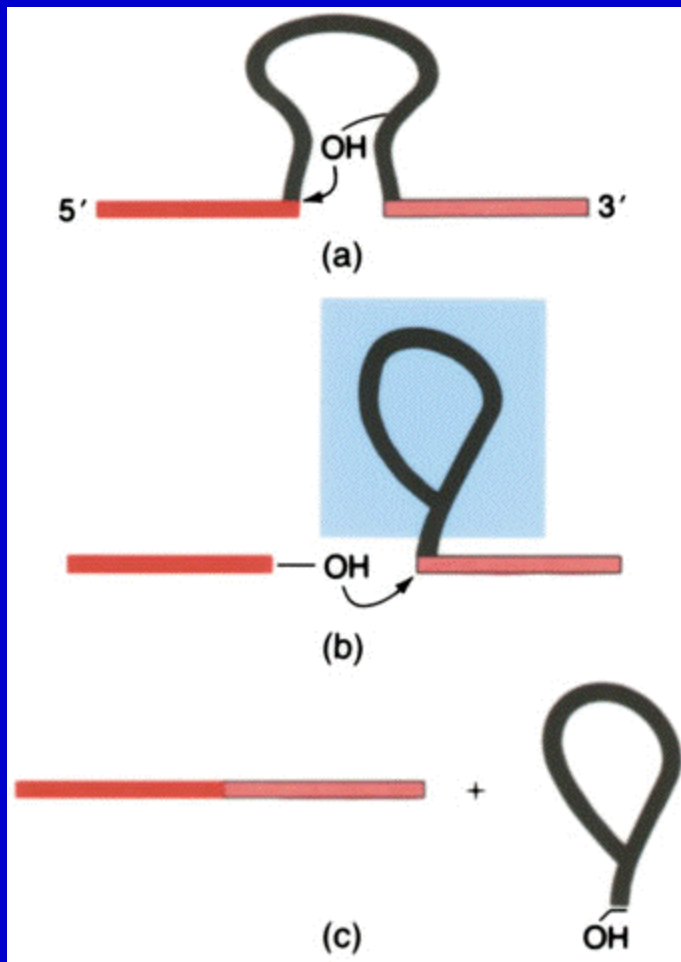
Hibridação
DNA-RNA



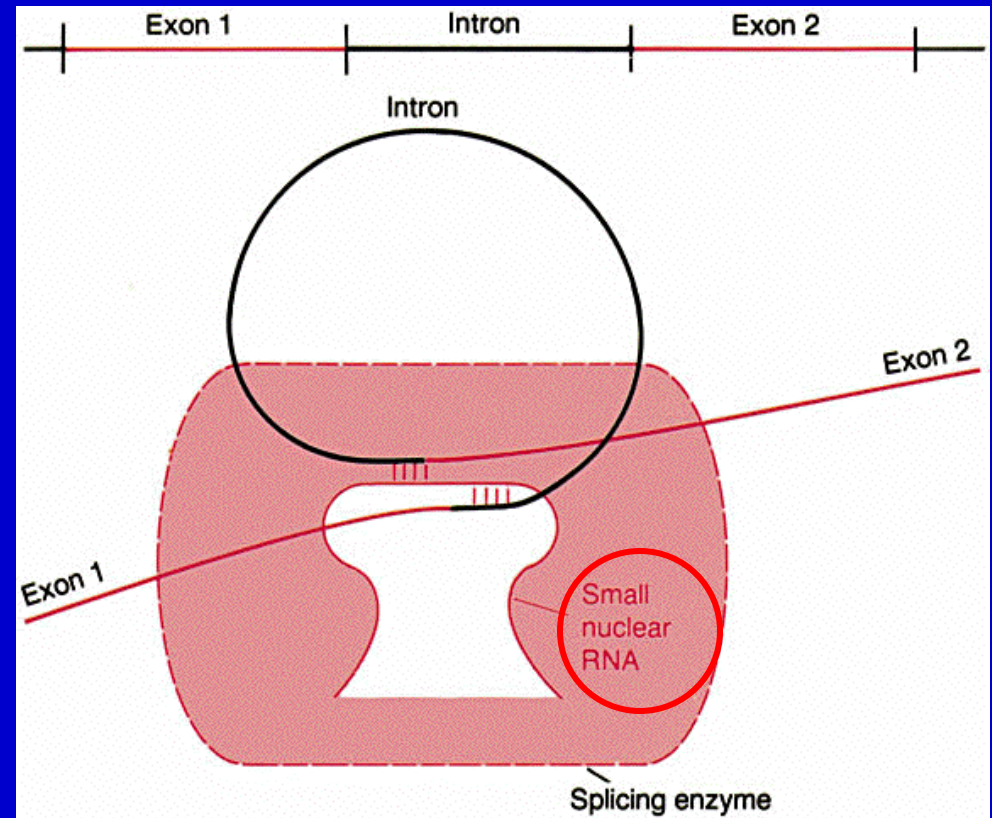
Passos do processamento do RNA



Mecanismos do processamento do RNA

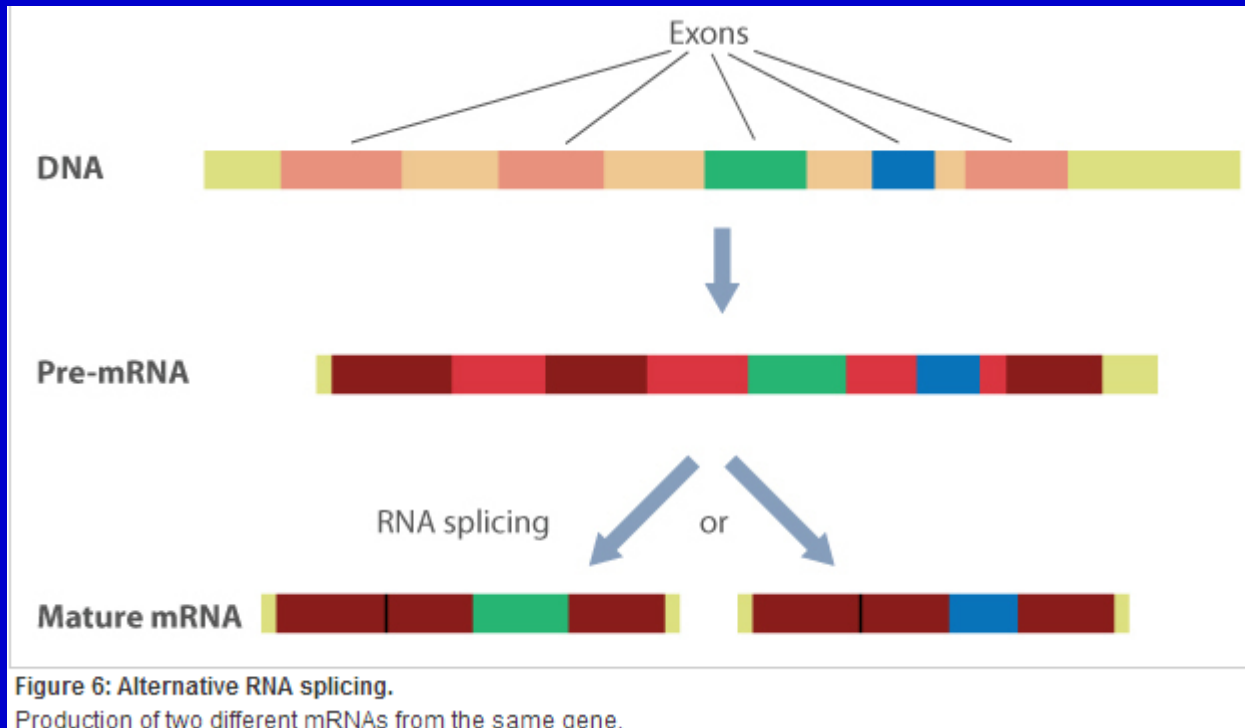


Auto splicing



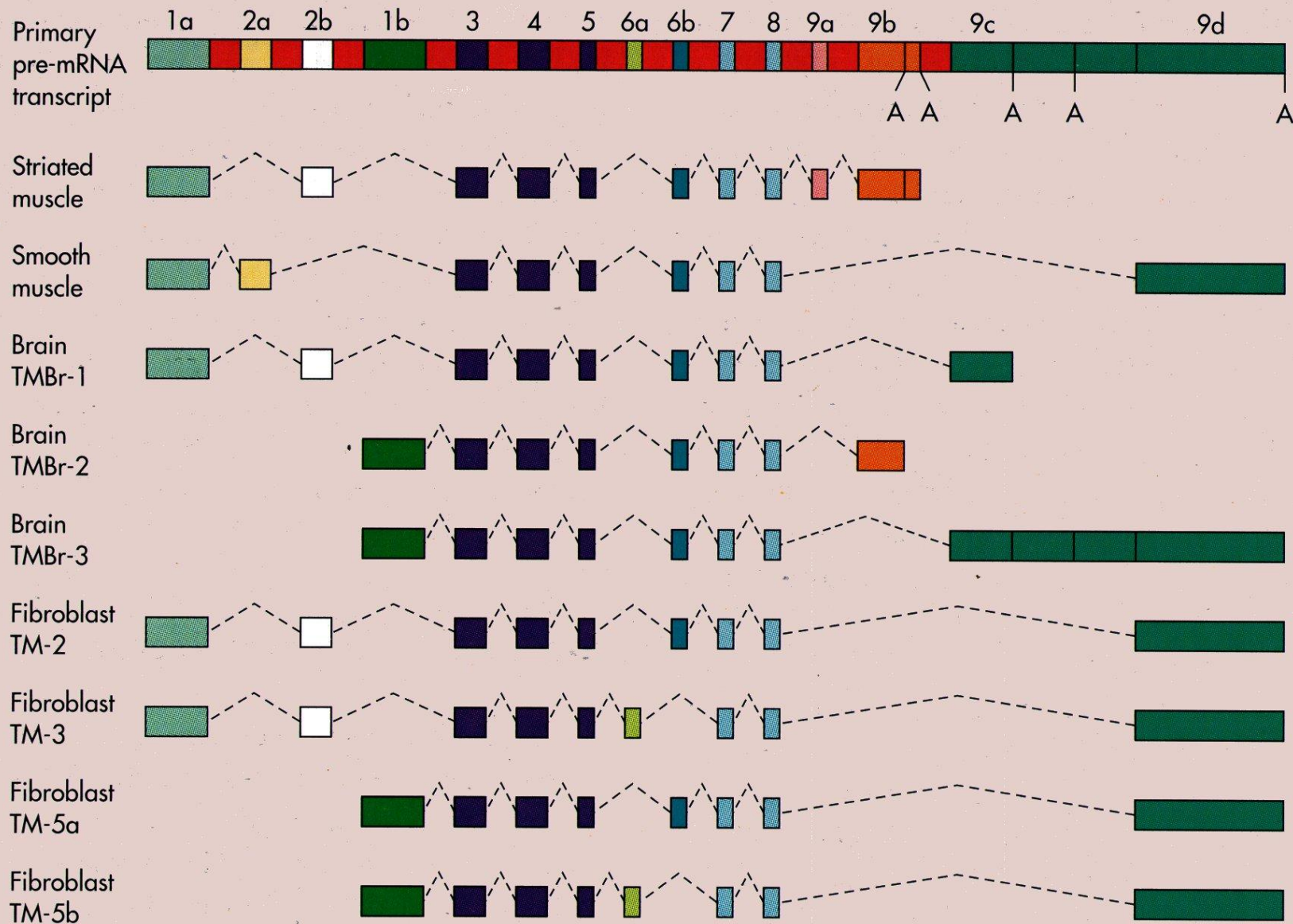
Processamento por spliceossomo

Processamento alternativo do RNA

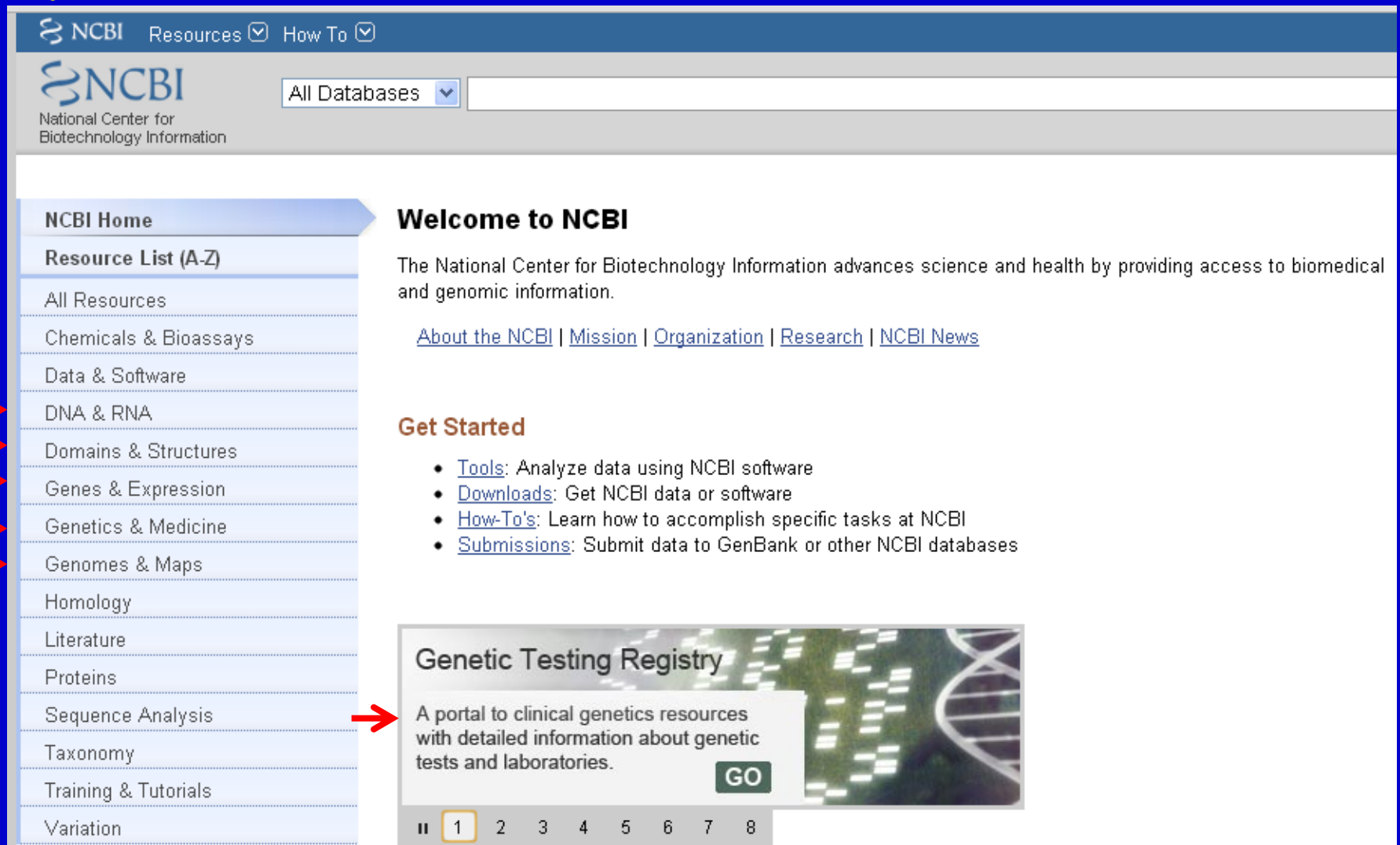


Produz 2 diferentes RNAs a partir do mesmo gene

Processamento diferencial do gene da tropomiosina



Sequências de DNA são submetidas ao NCBI



The image shows a screenshot of the NCBI website. The top navigation bar includes the NCBI logo, 'Resources' with a dropdown arrow, and 'How To' with a dropdown arrow. Below this is a search bar with a dropdown menu set to 'All Databases'. The main content area is divided into a left sidebar and a main body. The sidebar contains a 'Resource List (A-Z)' with various categories. The main body features a 'Welcome to NCBI' section with a brief description and links to 'About the NCBI', 'Mission', 'Organization', 'Research', and 'NCBI News'. Below this is a 'Get Started' section with links to 'Tools', 'Downloads', 'How-To's', and 'Submissions'. At the bottom, there is a 'Genetic Testing Registry' advertisement with a 'GO' button and a pagination bar showing '1' selected.

NCBI Resources How To

NCBI
National Center for
Biotechnology Information

All Databases

NCBI Home

Resource List (A-Z)

All Resources

Chemicals & Bioassays

Data & Software

DNA & RNA

Domains & Structures

Genes & Expression

Genetics & Medicine

Genomes & Maps

Homology

Literature

Proteins

Sequence Analysis

Taxonomy

Training & Tutorials

Variation

Welcome to NCBI

The National Center for Biotechnology Information advances science and health by providing access to biomedical and genomic information.

[About the NCBI](#) | [Mission](#) | [Organization](#) | [Research](#) | [NCBI News](#)

Get Started

- [Tools](#): Analyze data using NCBI software
- [Downloads](#): Get NCBI data or software
- [How-To's](#): Learn how to accomplish specific tasks at NCBI
- [Submissions](#): Submit data to GenBank or other NCBI databases

Genetic Testing Registry

A portal to clinical genetics resources with detailed information about genetic tests and laboratories.

GO

1 2 3 4 5 6 7 8

National Center for Biotechnology Information

<http://www.ncbi.nlm.nih.gov/>

NCBI: recurso para pesquisar doenças genética


NCBI Genomic Biology Homo sapiens

Search for

Browse your Genome

Click on the Chromosome to show

Genes




1 2 3 4 5 6 7 8
9 10 11 12 13 14 15 16
17 18 19 20 21 22 X Y

Find A Gene


Search for

from

The NCBI Handbook



An online guide to the use of NCBI resources. Titles of selected chapters that refer to human genome resources are shown below.



Human Genome Resources

A challenge facing researchers today is that of piecing together and analyzing the plethora of data currently being generated through the Human Genome Project and scores of smaller projects. NCBI's Web site serves as an integrated, one-stop, genomic information infrastructure for biomedical researchers from around the world so that they may use these data in their research efforts. [More...](#)

Genes and Human Health

- ▶ **Gene Database**
A new database of genes and associated information is now available for searching in Entrez.
- ▶ **RefSeq**
Reference sequences of chromosomes, genomic contigs, mRNAs, and proteins for human and major model organisms.
- ▶ **OMIM**
A guide to human genes and inherited disorders maintained by Johns Hopkins University and collaborators.
- ▶ **dbSNP**
A database of single nucleotide polymorphisms (SNPs) and other nucleotide variations.

Reagents

- ▶ **Full-Length cDNA Clones**
The NIH Mammalian Gene Collection (MGC) provides sequence-verified cDNA clone reagents for most
- ▶ **Molecular Probes**
A new NCBI database provides sequences of molecular probes, their biomedical applications, and



OMIM

Online Mendelian Inheritance in Man



Johns Hopkins University

All Databases PubMed Nucleotide Protein Genome Structure PMC OMIM

Search for

- Entrez
- OMIM
- Search OMIM
- Search Gene Map
- Help
- FAQ
- Statistics
- Update List

- Allied Resources
- The Jackson Laboratory
- Human Gene Nomenclature

- Human Genome Resources
- Entrez Gene
- Genes and Disease
- GeneReviews

Limits Preview/Index History Clipboard Details

- Enter one or more search terms.
- Use **Limits** to restrict your search by search field, chromosome, and other criteria.
- Use **Index** to browse terms found in OMIM records.
- Use **History** to retrieve records from previous searches, or to combine searches.

NCBI is implementing changes to help you find current content in OMIM based on resources omim.org. Please be aware that you will leave NCBI to view OMIM records. Access to full records (via PubMed) will no longer be supported.

OMIM[®] - Online Mendelian Inheritance in Man[®]

Welcome to OMIM[®], Online Mendelian Inheritance in Man[®]. OMIM is a comprehensive, authoritative catalog of human genes and genetic phenotypes. The full-text, referenced overviews in OMIM contain information on Mendelian disorders and over 12,000 genes. OMIM focuses on the relationship between phenotype and genotype. Many entries contain copious links to other genetics resources.

This database was initiated in the early 1960s by Dr. Victor A. McKusick as a catalog of mendelian disorders. Mendelian Inheritance in Man (MIM). Twelve book editions of MIM were published between 1966 and 1994. OMIM, was created in 1985 by a collaboration between the National Library of Medicine and the Johns Hopkins University at Johns Hopkins. It was made generally available on the internet starting in 1987. In 1995, OMIM

BLAST Assembled RefSeq Genomes

Choose a species genome to search, or [list all genomic BLAST databases](#).

- [Human](#)
- [Oryza sativa](#)
- [Gallus gallus](#)
- [Mouse](#)
- [Bos taurus](#)
- [Pan troglodytes](#)
- [Rat](#)
- [Danio rerio](#)
- [Microbes](#)
- [Arabidopsis thaliana](#)
- [Drosophila melanogaster](#)
- [Apis mellifera](#)

Basic BLAST

Choose a BLAST program to run.

<u>nucleotide blast</u>	Search a nucleotide database using a nucleotide query <i>Algorithms: blastn, megablast, discontinuous megablast</i>
<u>protein blast</u>	Search protein database using a protein query <i>Algorithms: blastp, psi-blast, phi-blast, delta-blast</i>
<u>blastx</u>	Search protein database using a translated nucleotide query
<u>tblastn</u>	Search translated nucleotide database using a protein query
<u>tblastx</u>	Search translated nucleotide database using a translated nucleotide query

Specialized BLAST

Choose a type of specialized search (or database name in parentheses)

ENCODE: Encyclopedia of DNA Elements

05

06

Welcome to the

nature

ENCODE explorer

×

What is ENCODE?

Threads: a new approach

Guide to the ENCODE explorer



ENCODE, the Encyclopedia of DNA Elements, is a project funded by the National Human Genome Research Institute to identify all regions of transcription, transcription factor association, chromatin structure and histone modification in the human genome sequence. Thanks to the identification of these functional elements, 80% of the components of the human genome now have at least one biochemical function associated with them. This expansive resource of functional annotations is already providing new insights into the organization and regulation of our genes and genome.

<http://www.nature.com/encode/#/threads>

Bibliografía

Thompson & Thompson
Genética Médica

Exercícios

1. Porque a síntese de DNA é contínua em um filamento e descontínua no filamento oposto?
2. Que tipo de herança ocorre para o DNA mitocondrial e para o cromossomo Y humanos?
3. A quantidade de sequência de DNA não codificadoras de proteínas aumenta proporcionalmente com a complexidade dos organismos. Comente e explique.
4. O que você entende por processamento do RNA tecido-específico? De exemplo para a espécie humana.
5. Compare a visão clássica de "1 gene - 1 enzima" com o atual conhecimento dos processos de transcrição e tradução.