# ORGANIZAÇÃO E FUNCIONALIDADE DO GENOMA HUMANO

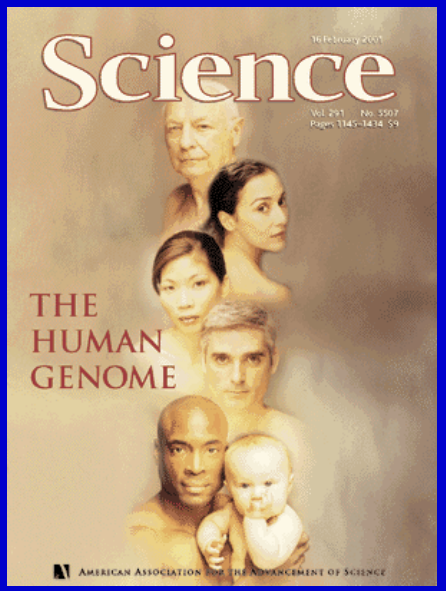

## Departamento de Genética

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[1–3] 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

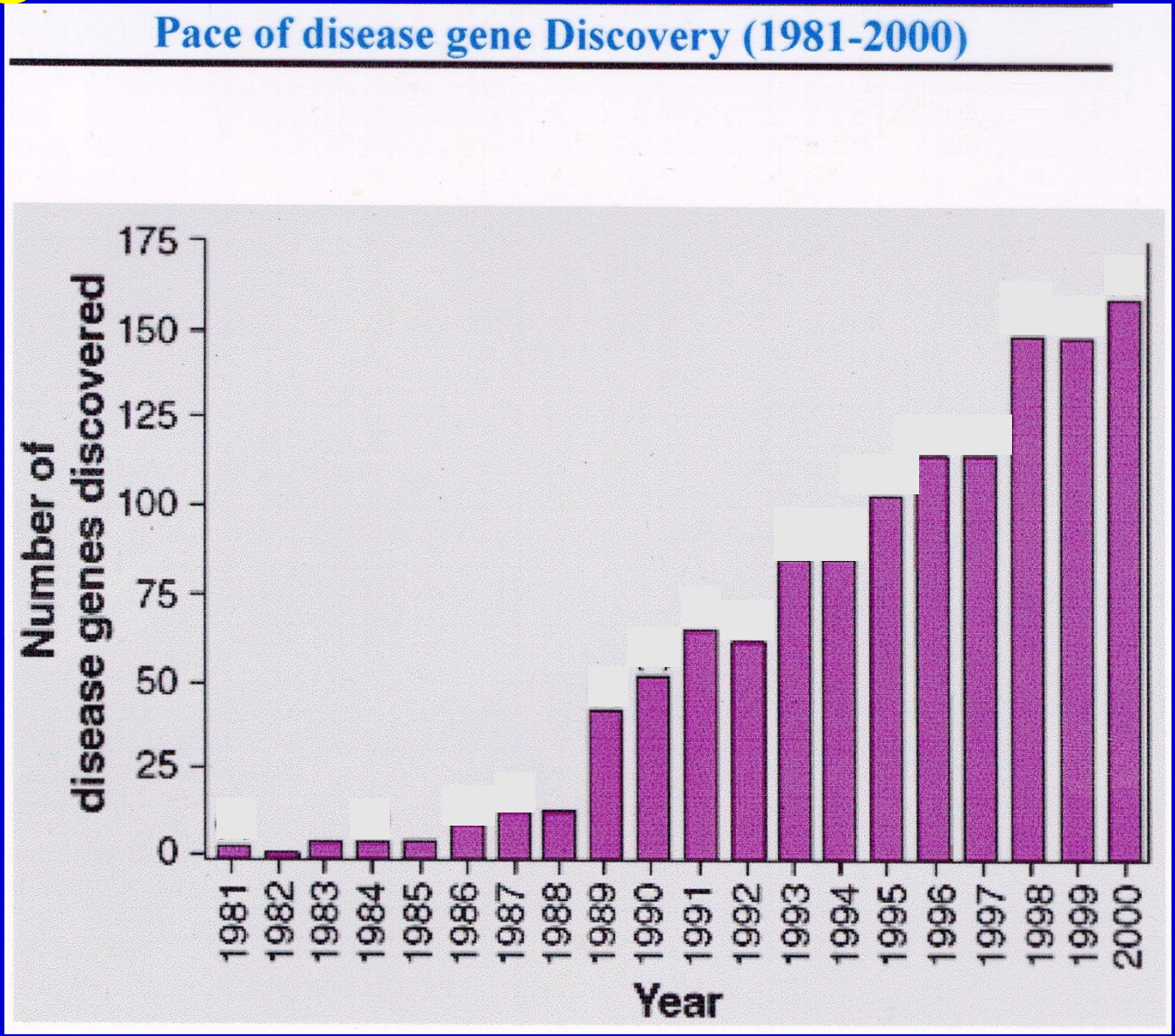
coordinate regulation of the genes in the clusters.

● There appear to be about 30,000–40,000 protein-coding genes in the human genome—only about twice as many as in worm or fly. However, the genes are more complex, with more alternative splicing generating a larger number of protein products.

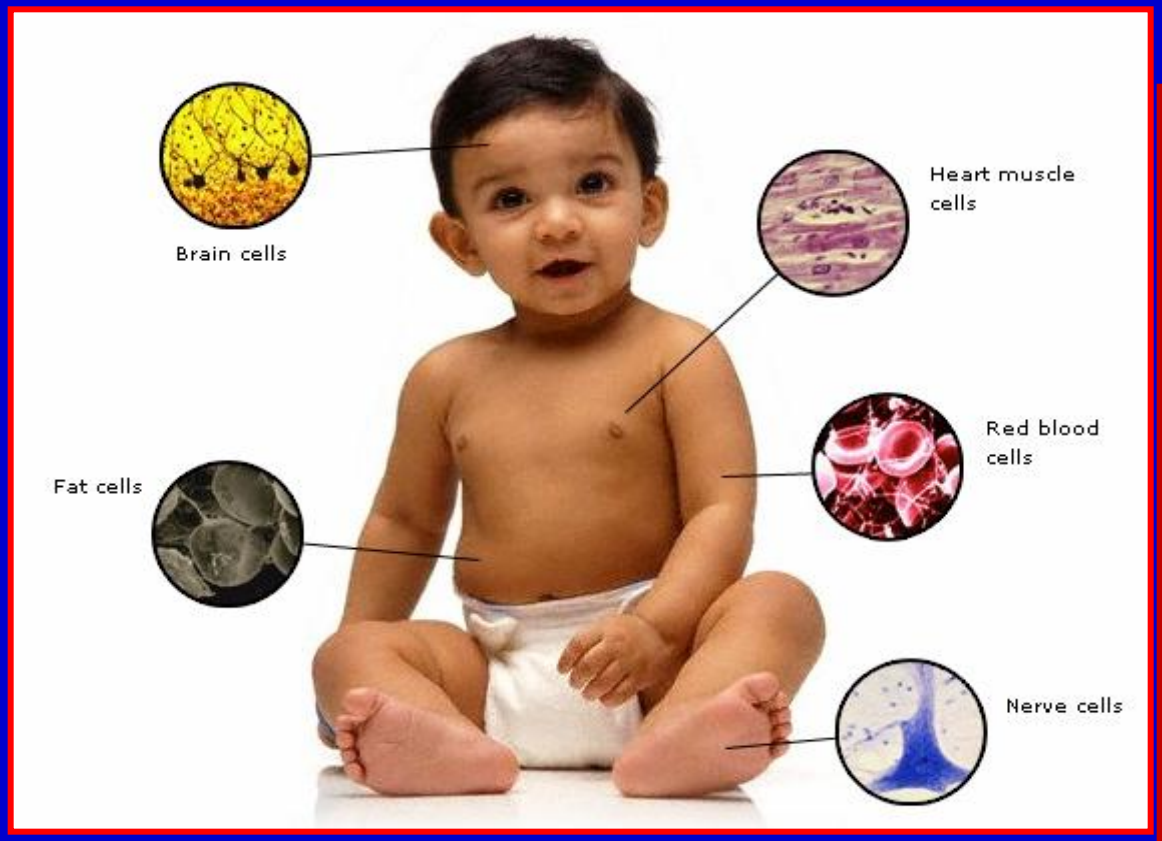
● The full set of proteins (the 'proteome') encoded by the human

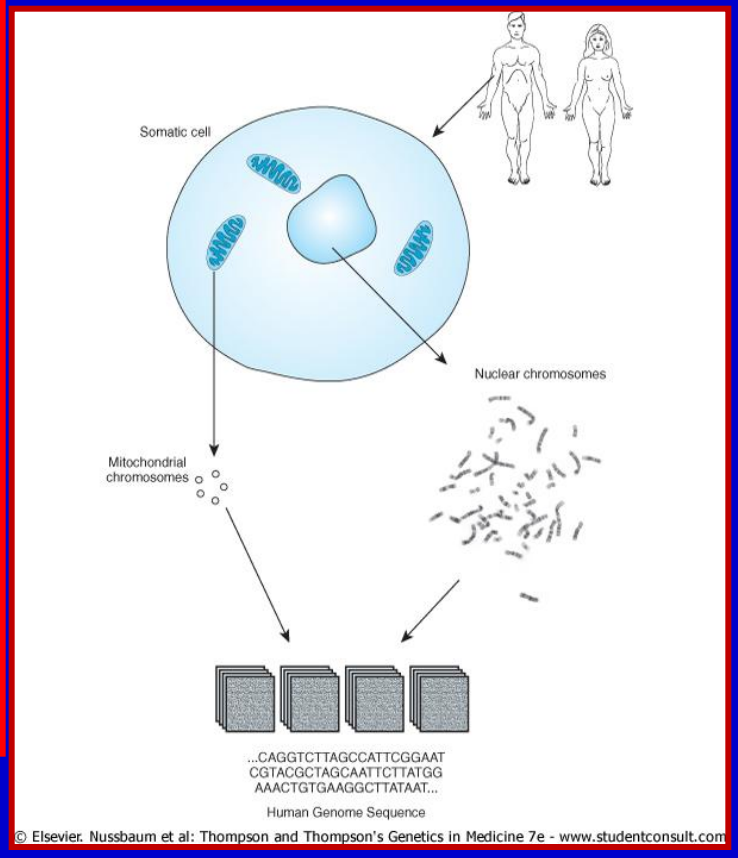
**Genoma: sequencia de DNA de todos os cromossomos de um organismo**

# Evolução das descobertas da origem genética de doenças humanas



# Onde estão os genes?

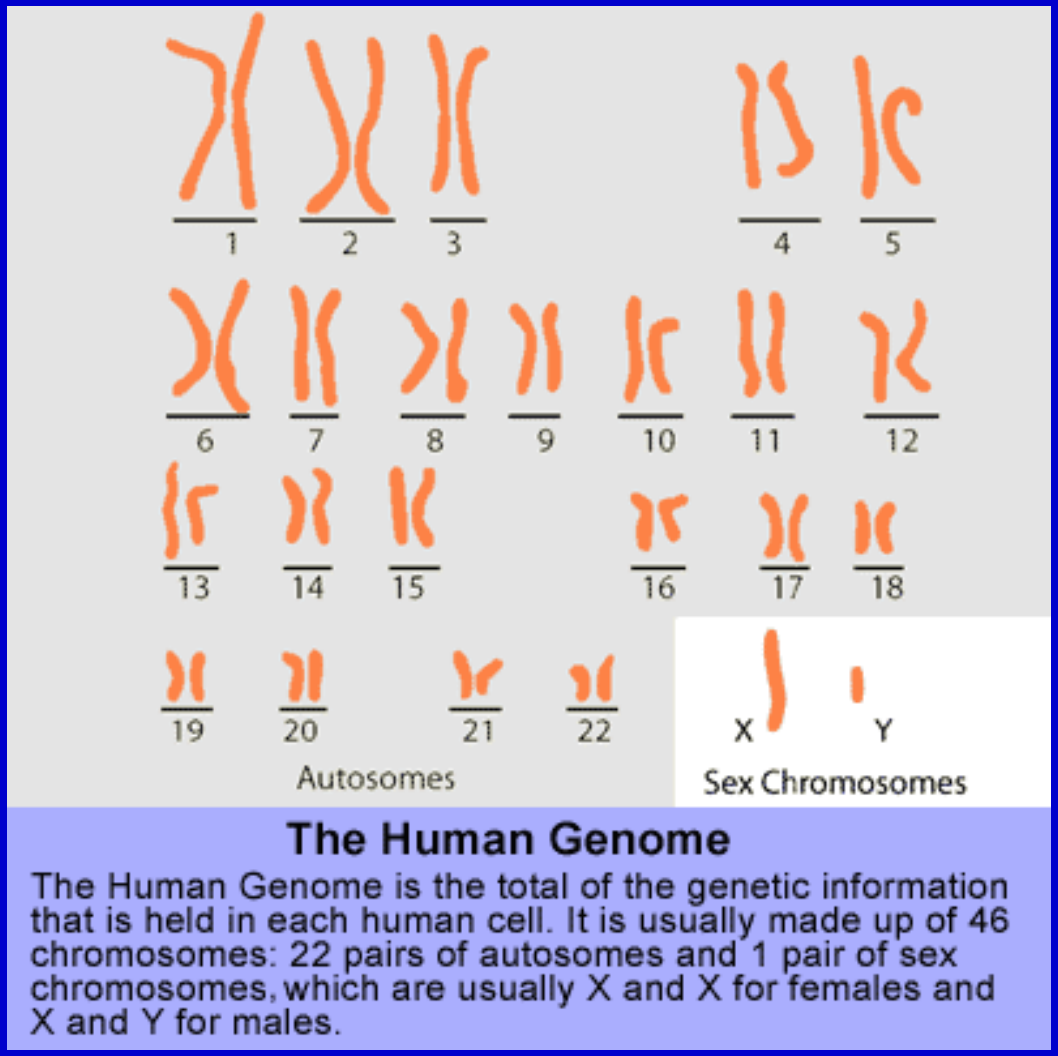


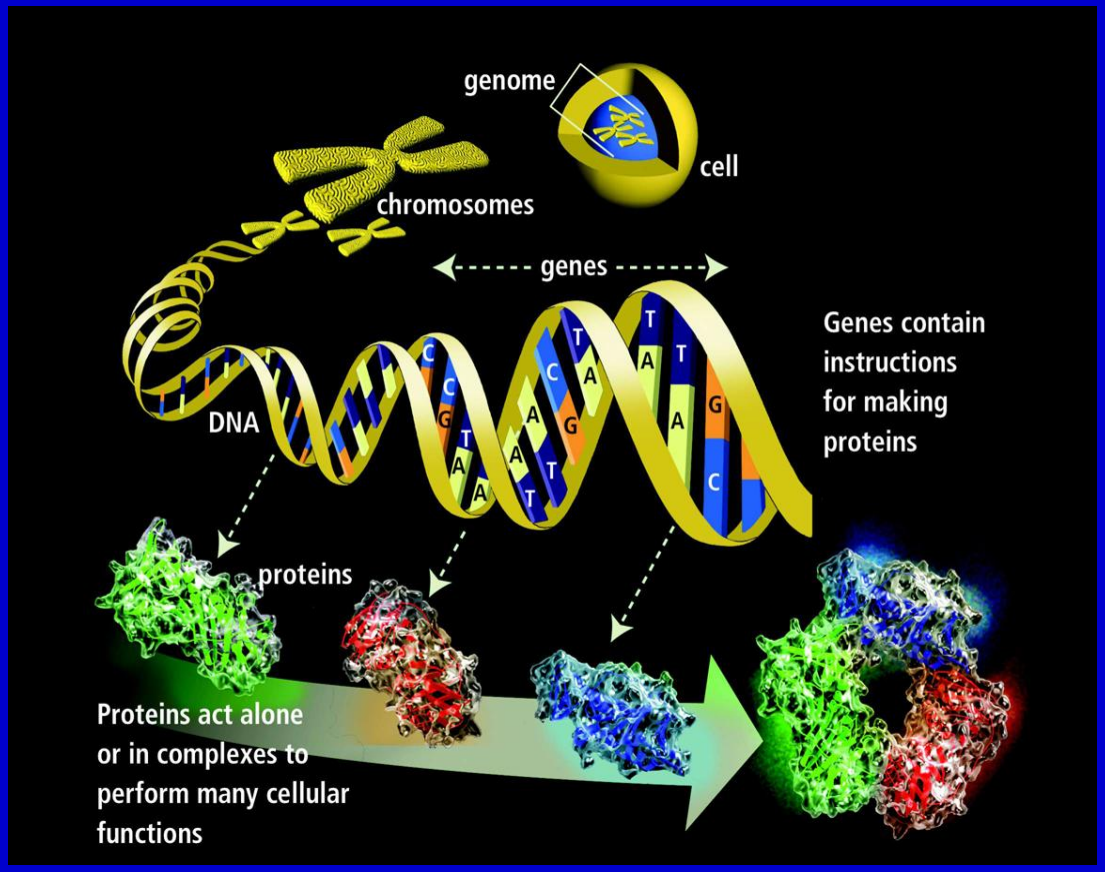

Cromossomos nucleares e mitocondriais

# Cromossomos Humanos

Cariótipo Humano



# DNA a molécula da vida




Cada célula tem:

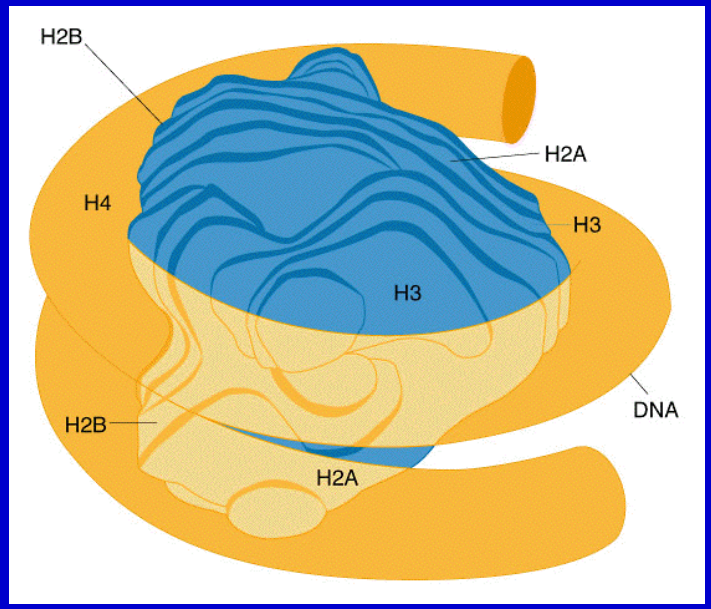
46 cromossomos humanos

2 metros de DNA

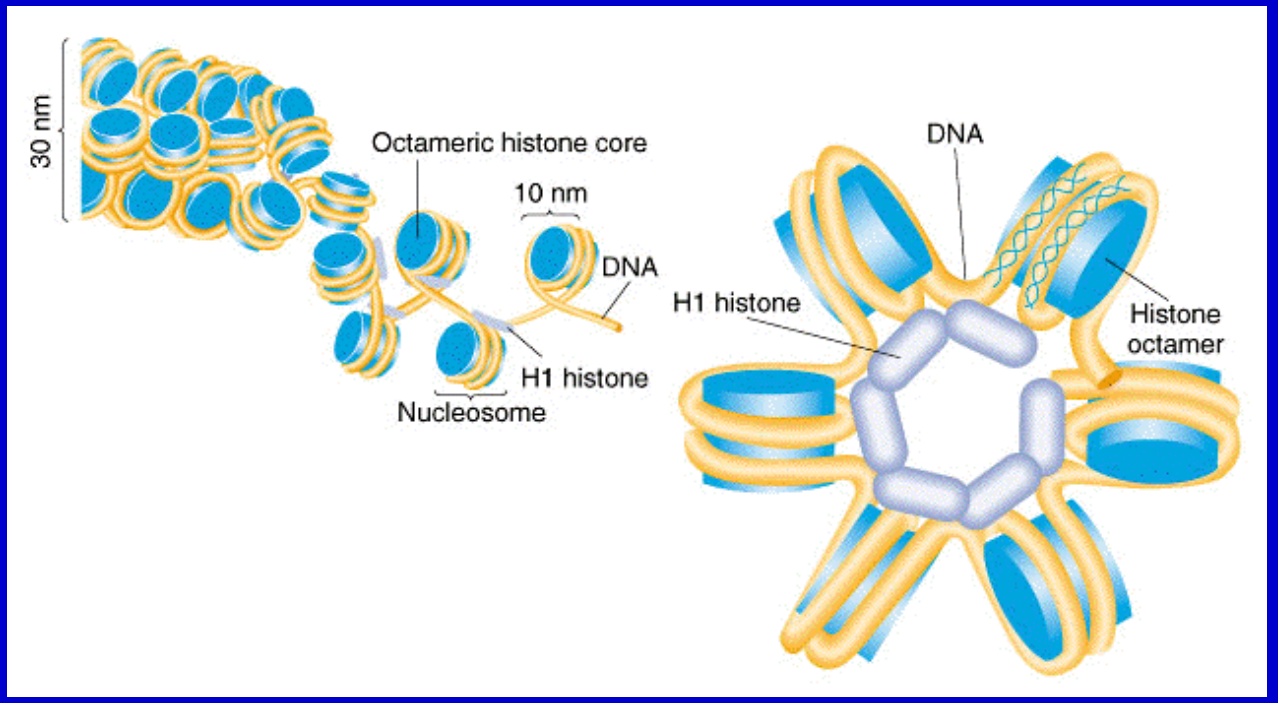
3 bilhões de subunidades de DNA (bases A, C, T,G)

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

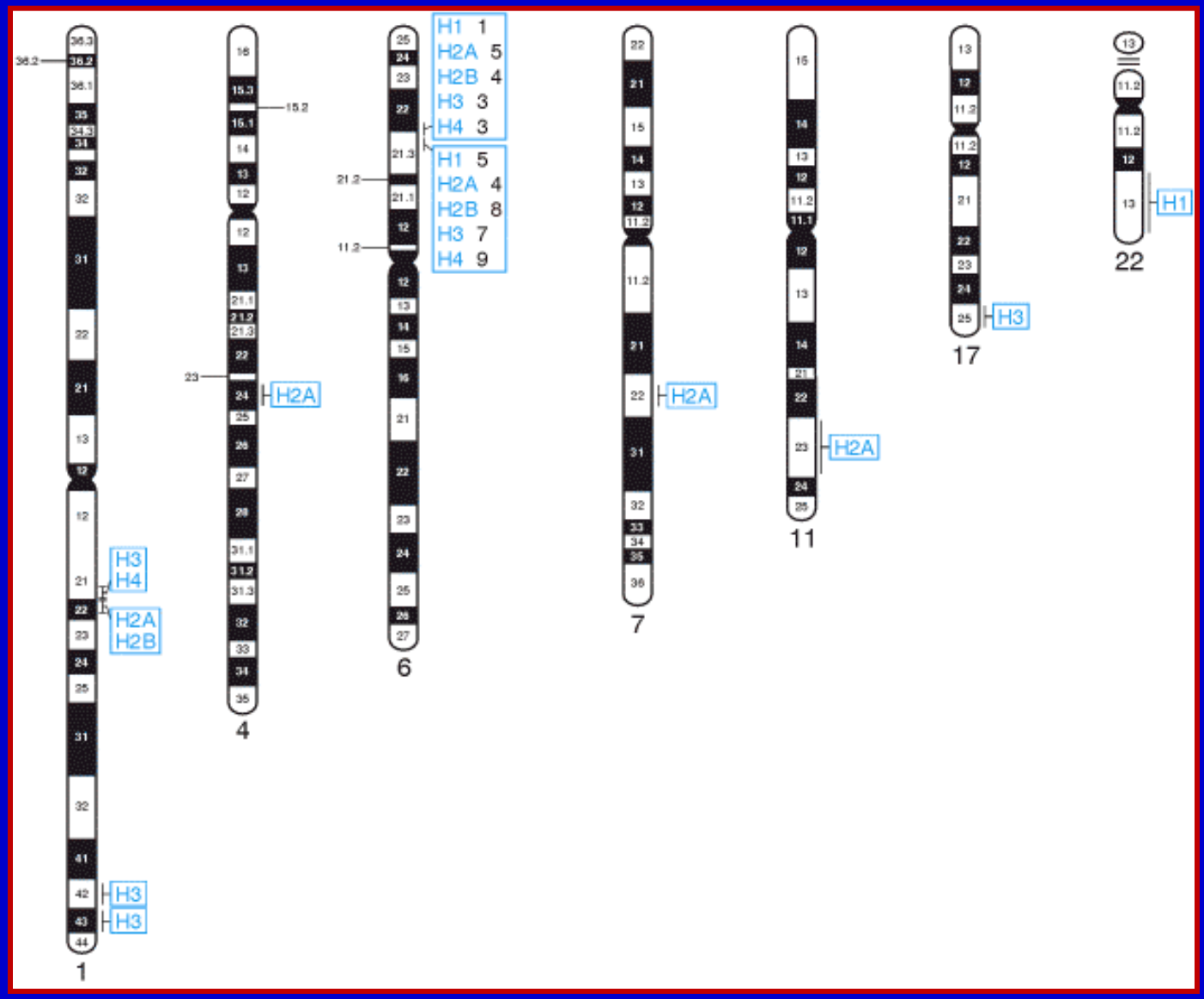
# Níveis de empacotamento do DNA



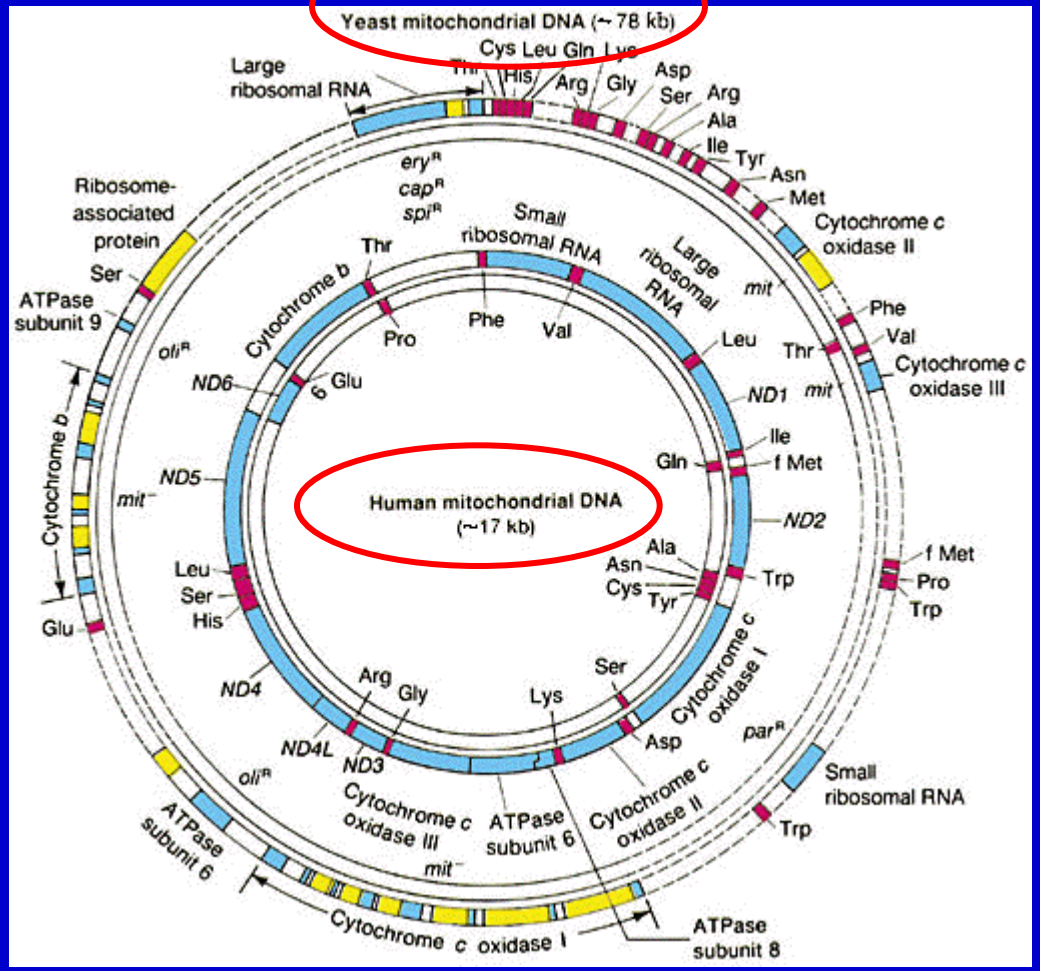

Nucleossomo



# Genes Humanos envolvidos na síntese de histonas



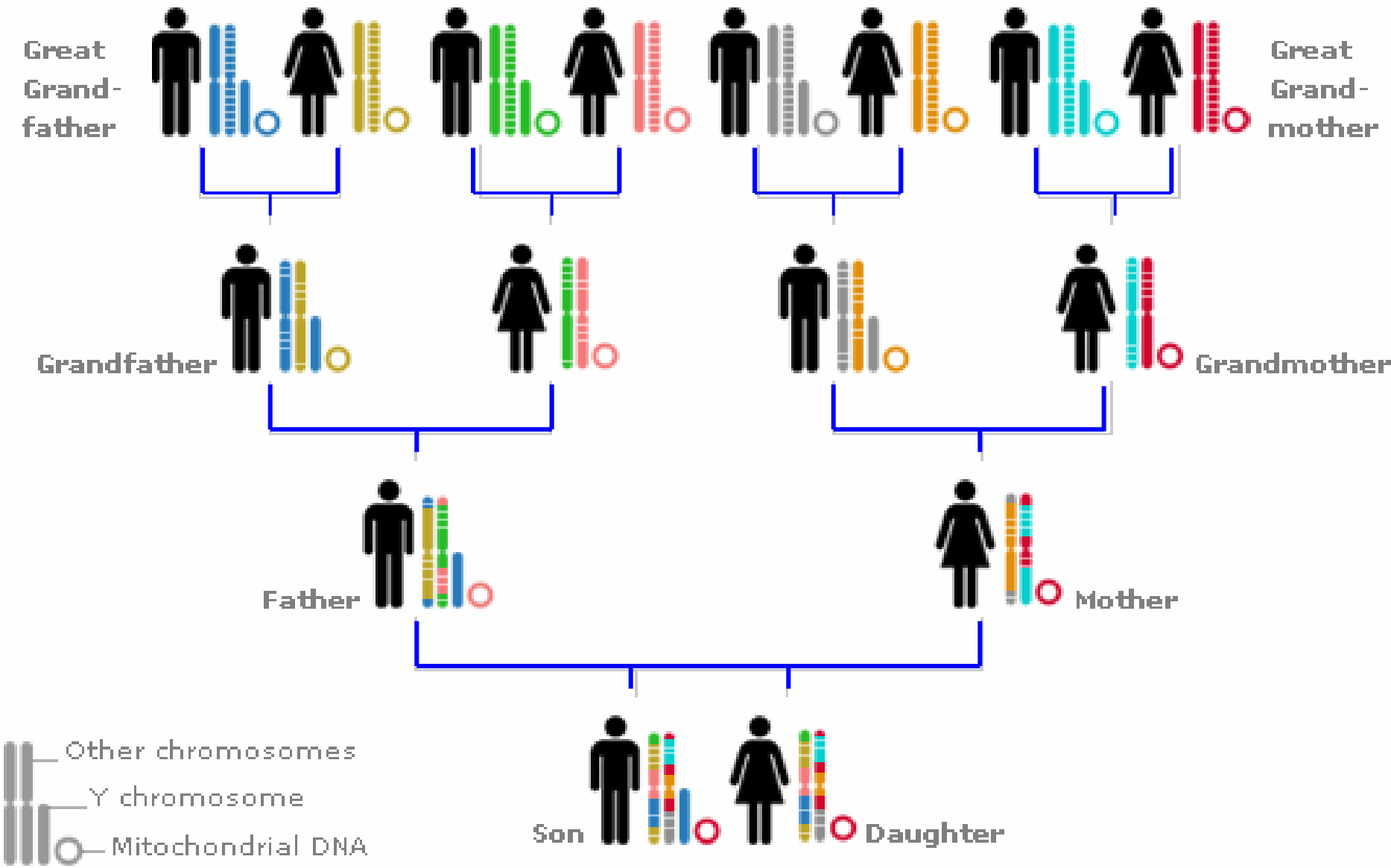
# DNA mitocondrial




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

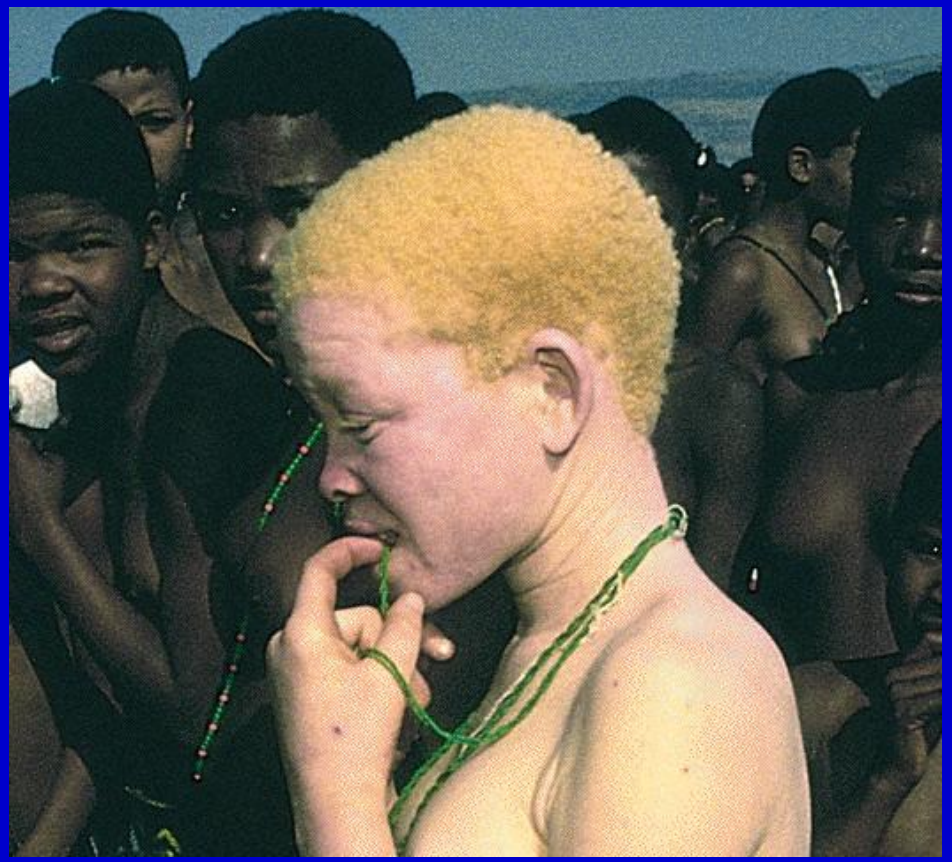


# Principais tipos de doenças genéticas

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

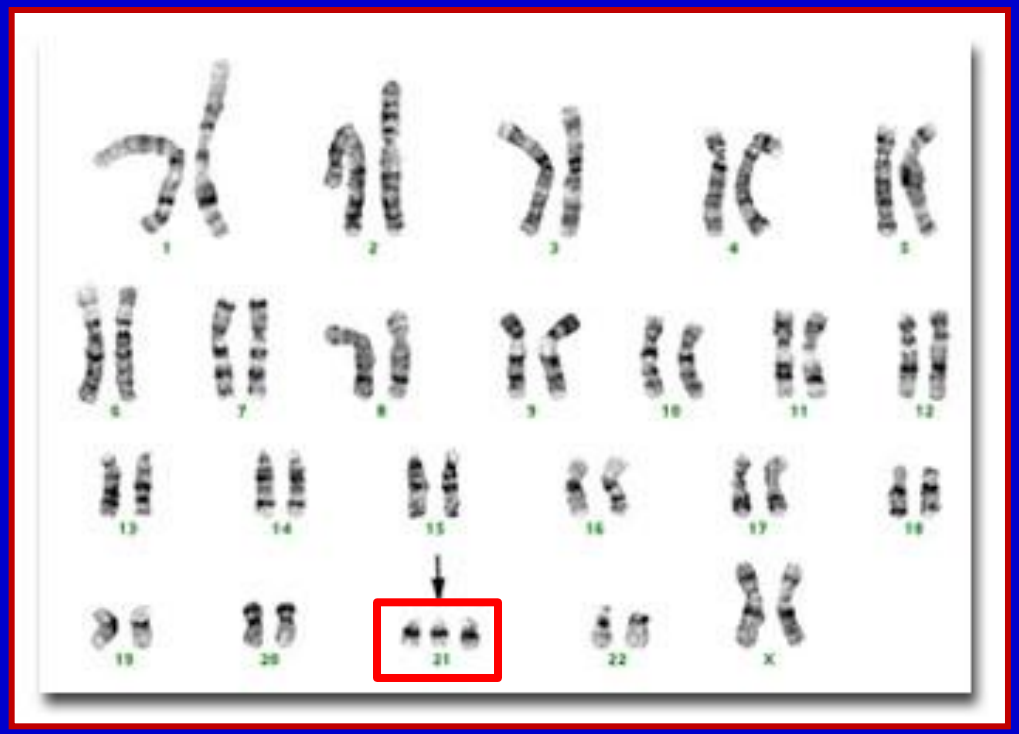
# Exemplos de doenças monogênicas

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



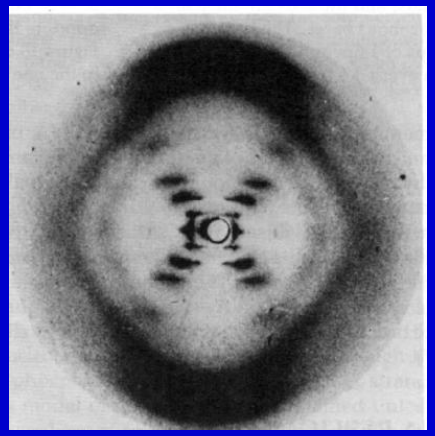
**Acondroplasia:**
substituição de uma glicina por arginina no domínio transmembranar do receptor do fator de crescimento fibroblástico 3

# 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

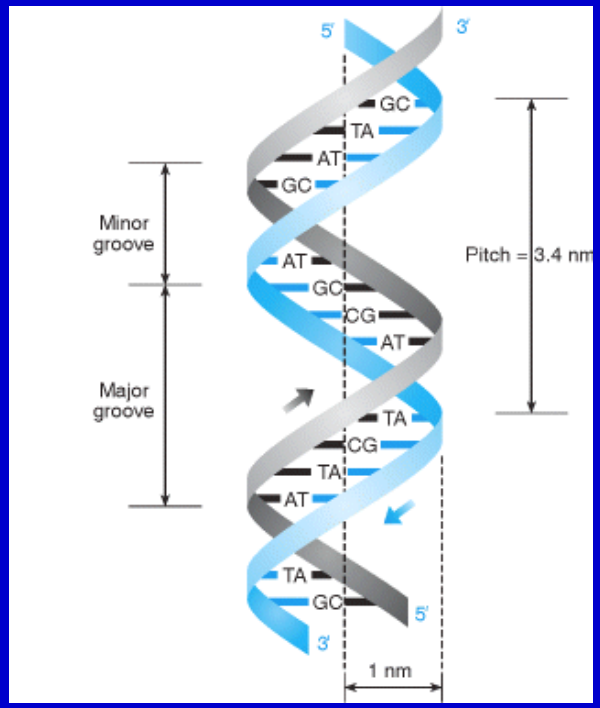
| Organism | Tissue | Adenine | Thymine | Guanine | Cytosine |
|---|---|---|---|---|---|
| *Escherichia coli* (K12) | — | 26.0 | 23.9 | 24.9 | 25.2 |
| *Diplococcus pneumoniae* | — | 29.8 | 31.6 | 20.5 | 18.0 |
| *Mycobacterium tuberculosis* | — | 15.1 | 14.6 | 34.9 | 35.4 |
| Yeast | — | 31.3 | 32.9 | 18.7 | 17.1 |
| *Paracentrotus lividus* (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 |

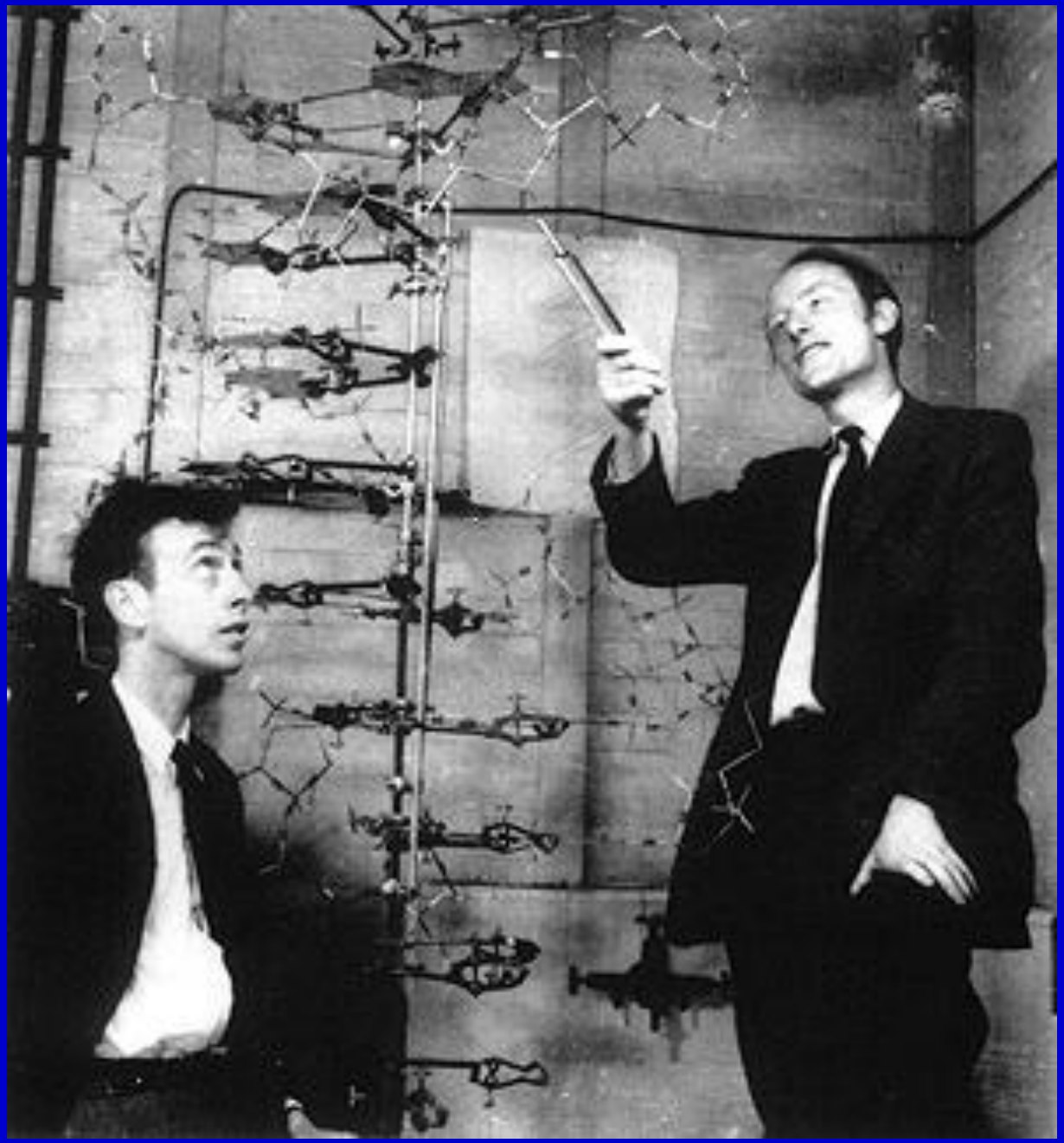
*Defined as moles of nitrogenous constituents per 100 g-atoms phosphate in hydrolysate.
SOURCE: E. Chargaff and J. Davidson, eds., *The Nucleic Acids*. Academic Press, 1955.

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 Corey[1]. They kindly made their manuscript available to us in advance of

is a residue on each chain ev
tion. We have assumed a
adjacent residues in the sa
structure repeats after 10 res
is, after 34 A. The distanc
from the fibre axis is 10 A.
the outside, cations have eas

The structure is an open o
is rather high. At lower w
expect the bases to tilt so
become more compact.

The novel feature of the
in which the two chains a
purine and pyrimidine bases.
are perpendicular to the fibr
together in pairs, a single ba
hydrogen-bonded to a sing
chain, so that the two lie si
*z*-co-ordinates. One of the pa
the other a pyrimidine for
hydrogen bonds are made as
1 to pyrimidine position 1
pyrimidine position 6.

If it is assumed that the

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

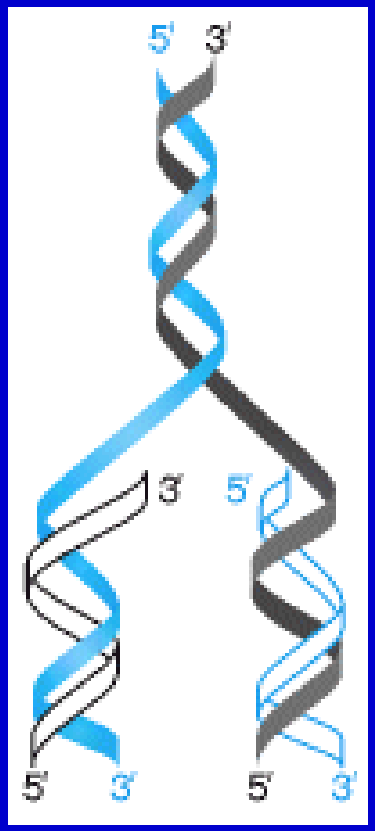
The Nobel Prize in Physiology or Medicine 1962 was awarded jointly to Francis Harry Compton Crick, James Dewey Watson and Maurice Hugh Frederick Wilkins *"for their discoveries concerning the molecular structure of nucleic acids and its significance for information transfer in living material"*.

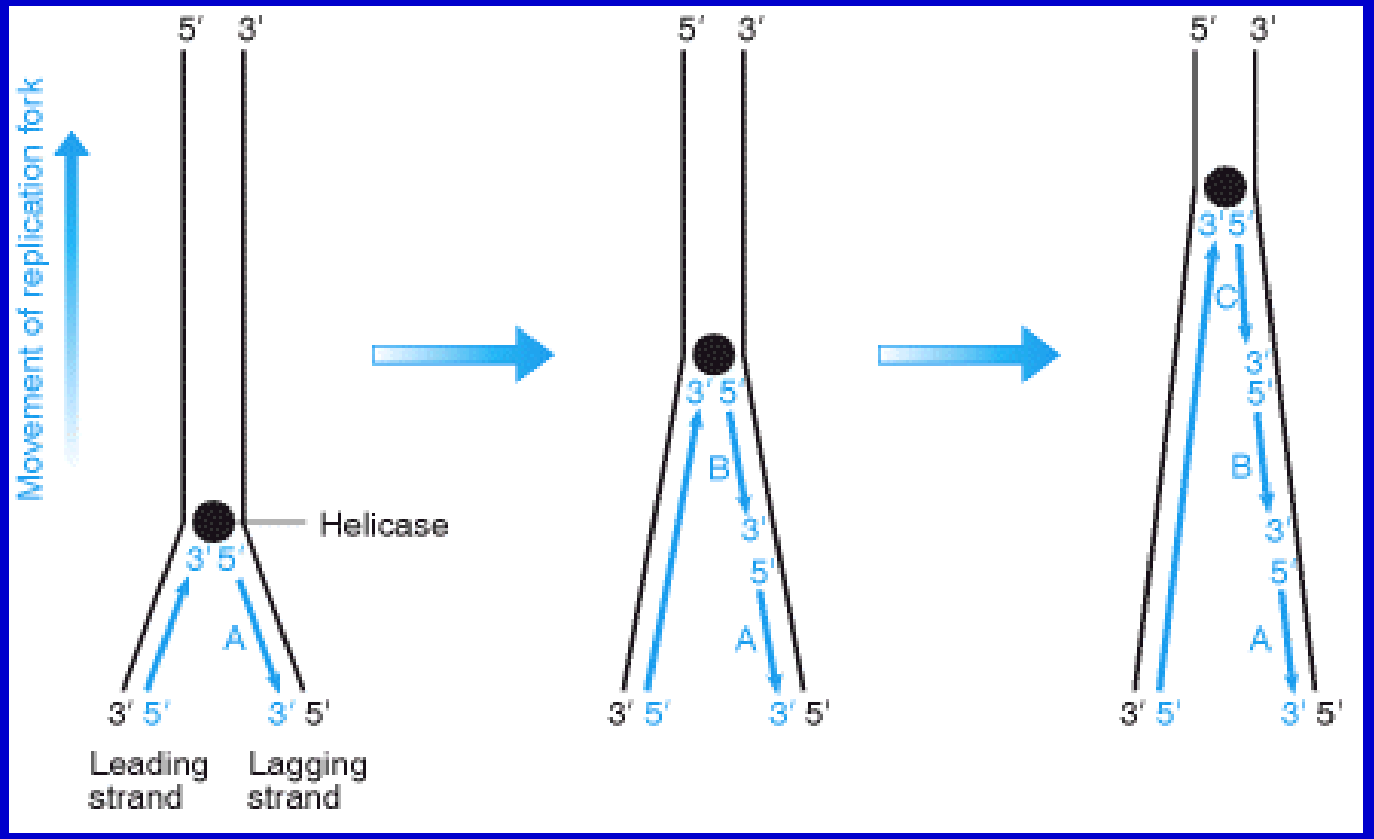
# Quais são as funções do DNA?

- Replicação: preservação da espécie por que é o material hereditário

- Transcrição: síntese de RNA

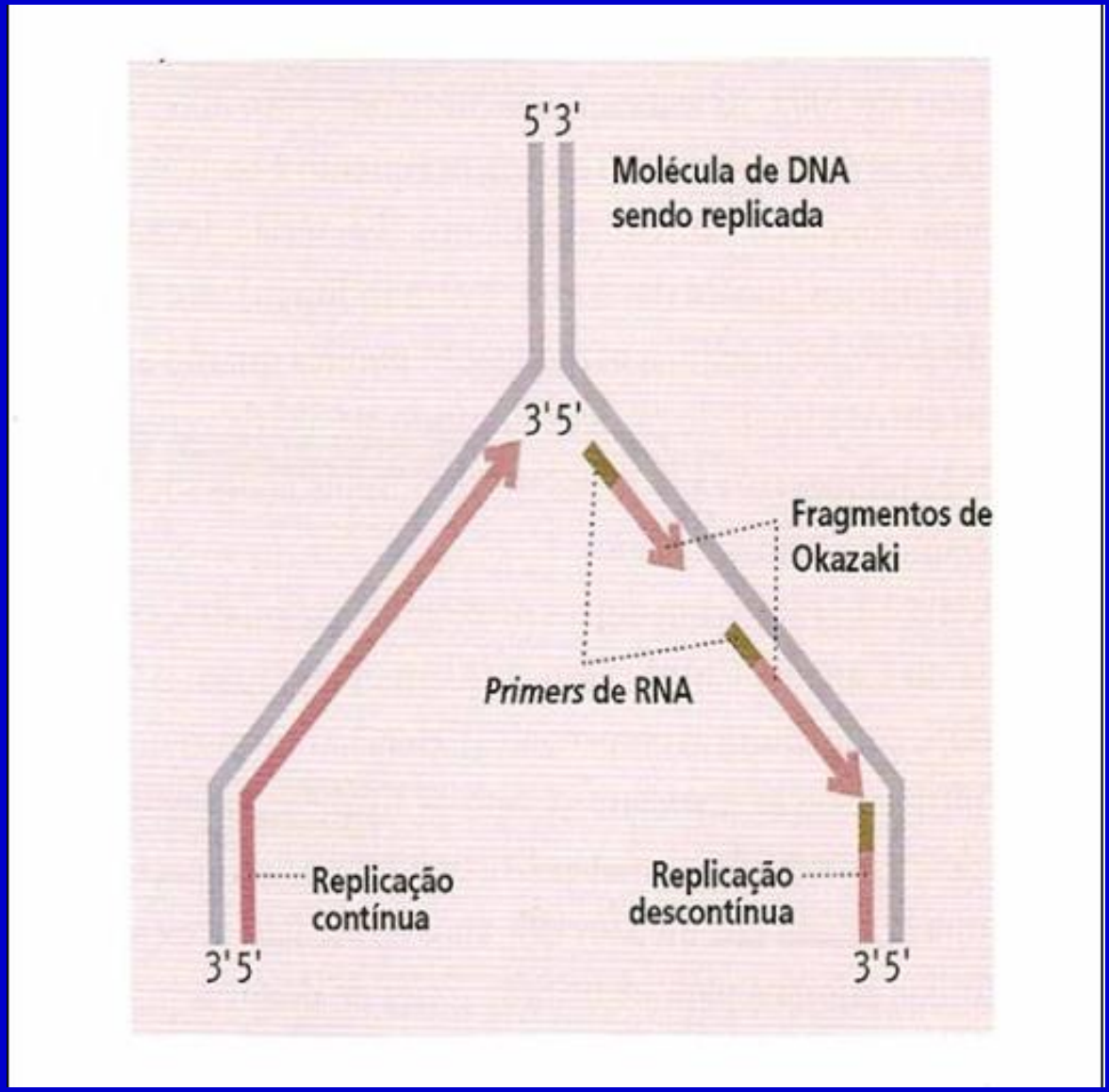
# A replicação do DNA é semiconservativa e a síntese das fitas de DNA é semidescontínua



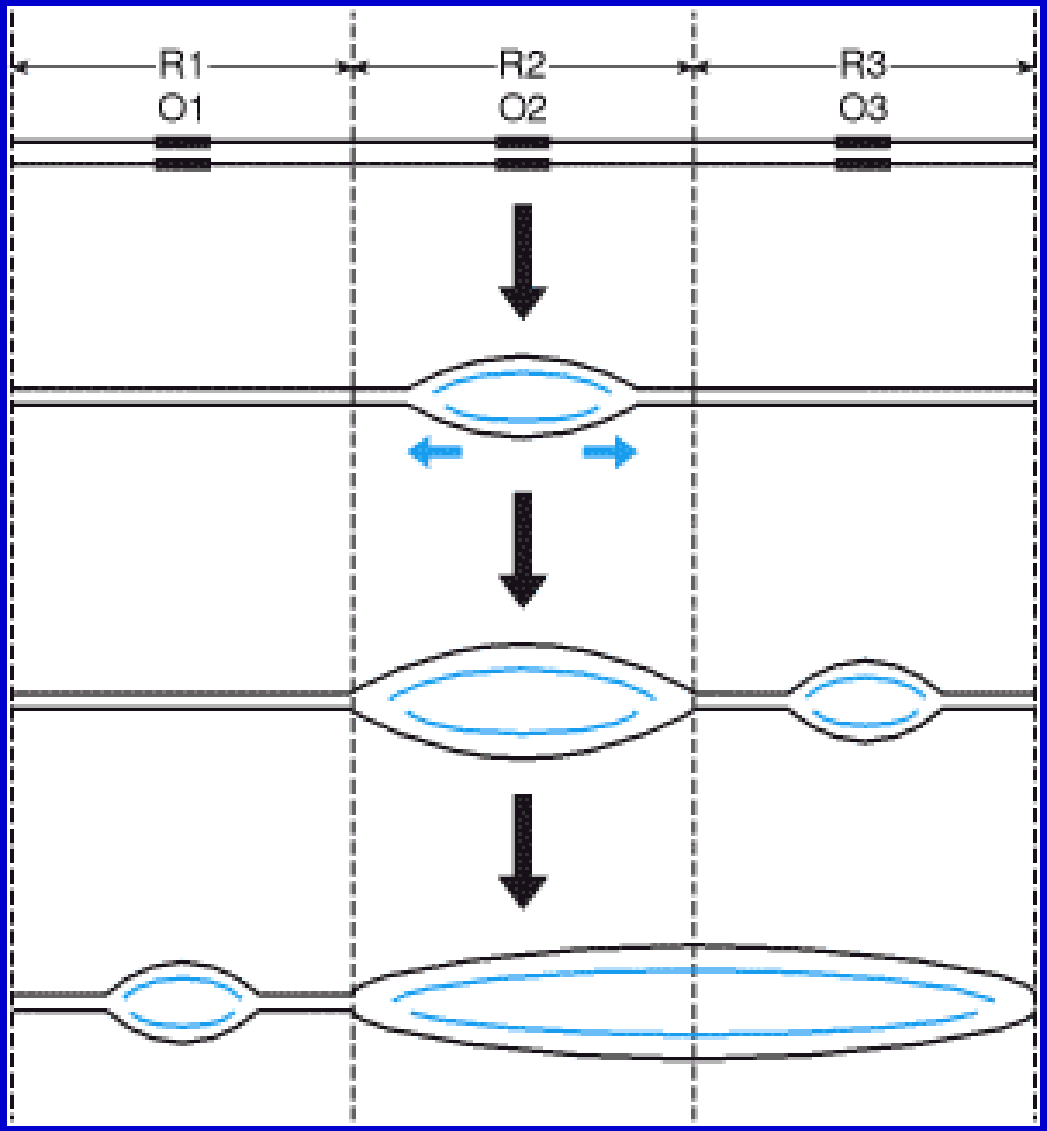



# Replicação do DNA e os fragmentos de Okazaki

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

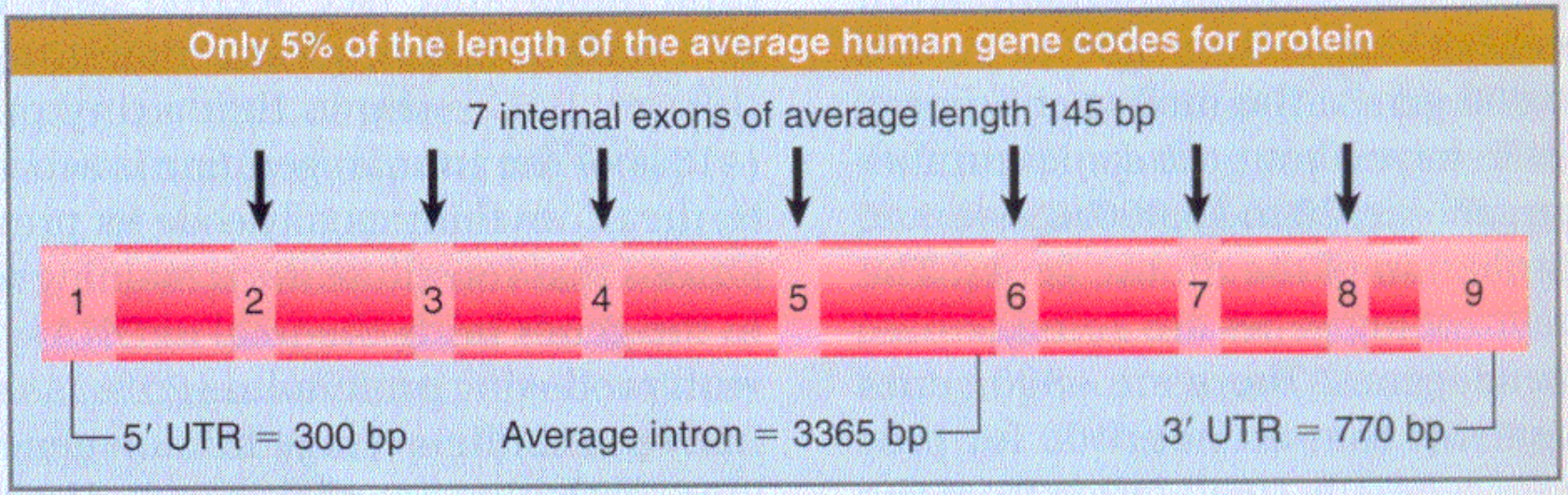
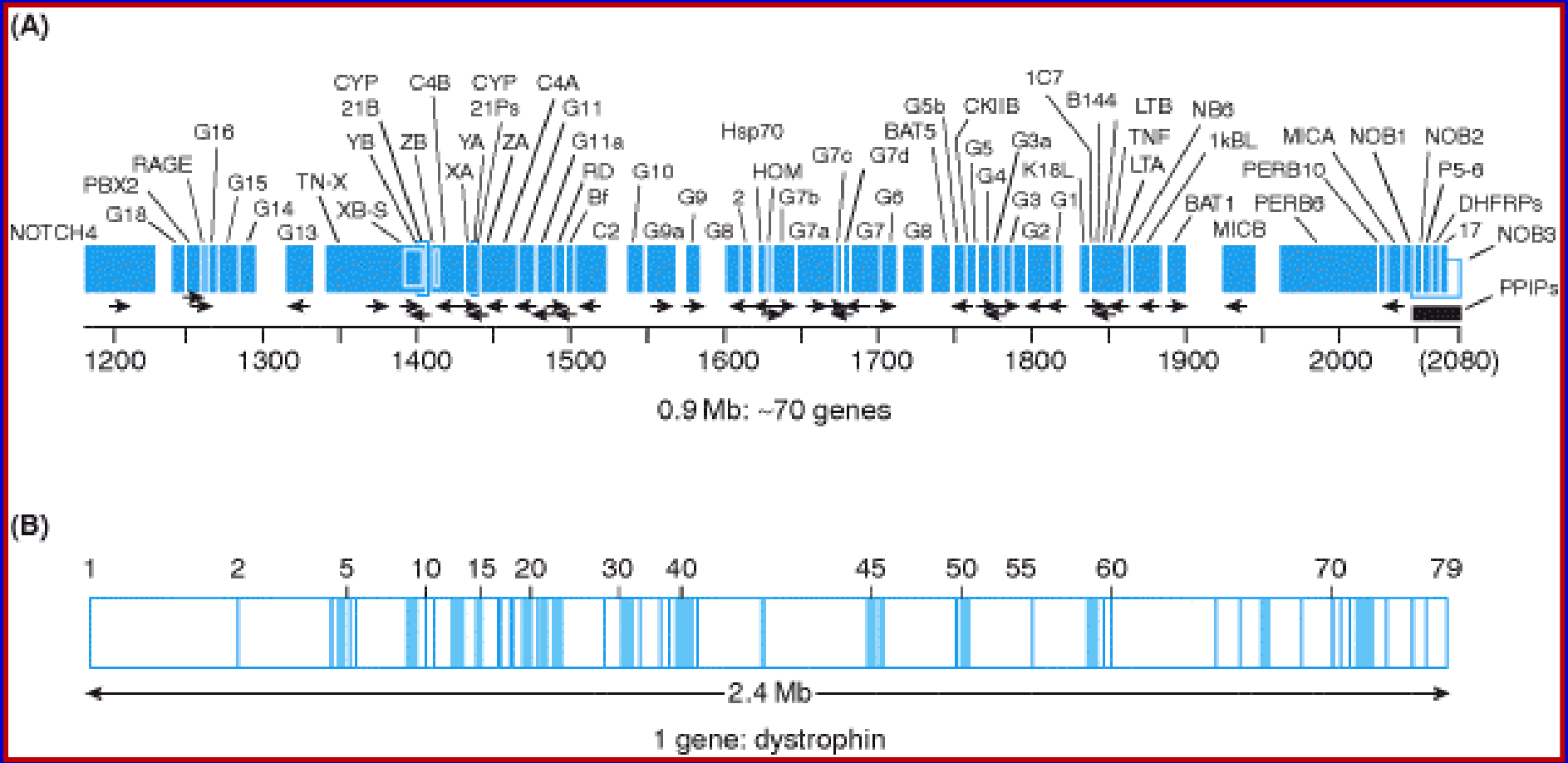



**FIGURE 5.12** The average human gene is 27 kb long and has nine exons, usually comprising two longer exons at each end and seven internal exons. The UTRs in the terminal exons are the untranslated (noncoding) regions at each end of the gene. (This is based on the average. Some genes are extremely long,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

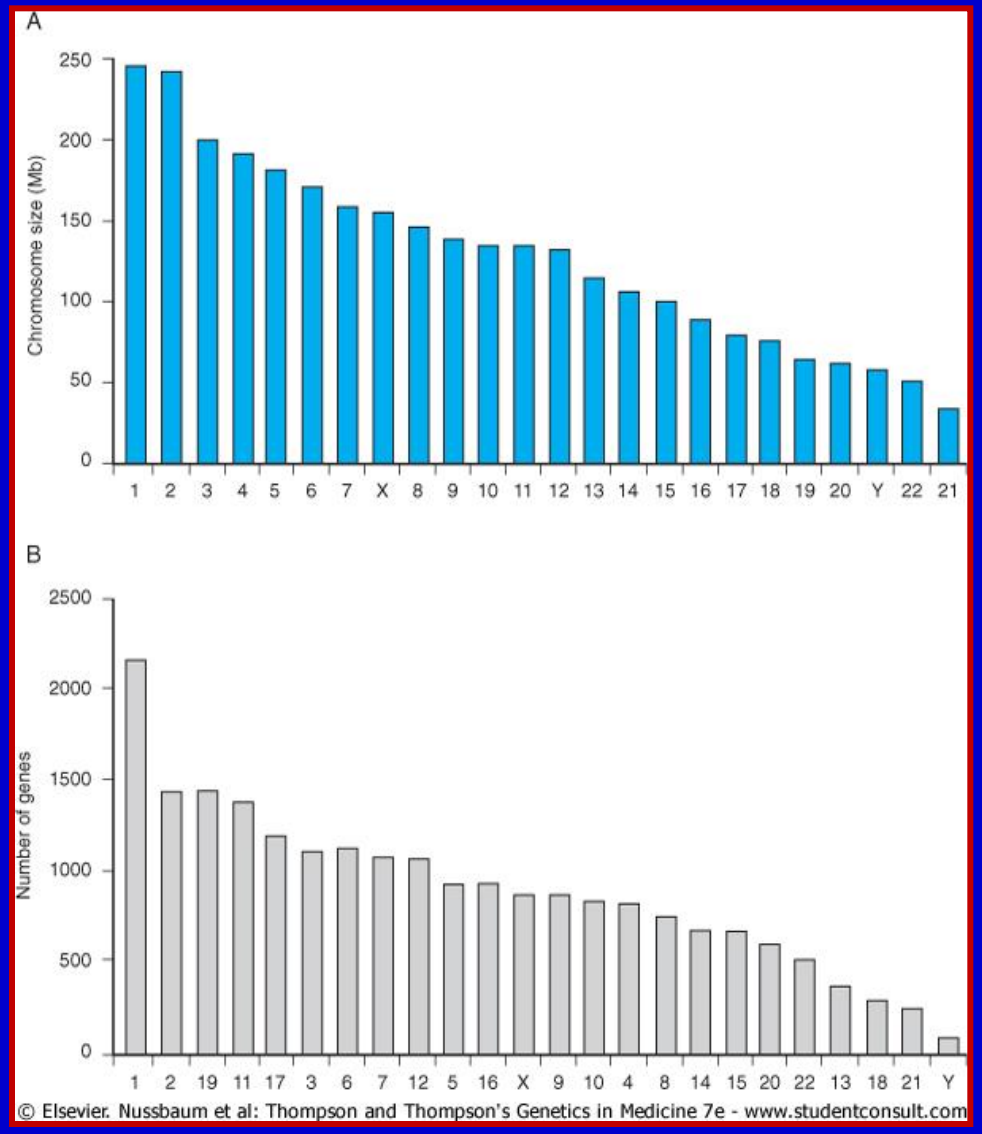
# Contraste entre as densidades gênicas humanas



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

Tamanho dos cromossomos

Número de genes por cromossomo



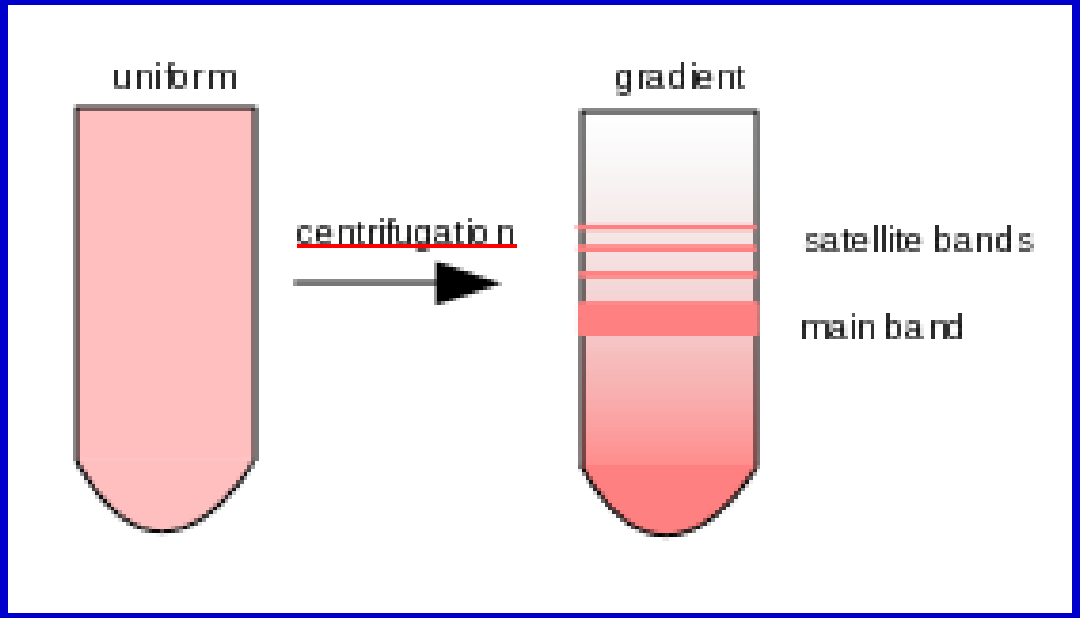
# Sequencias de DNA repetitivo

- 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)




← DNA satélite

← DNA principal

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

DNA fingerprinting

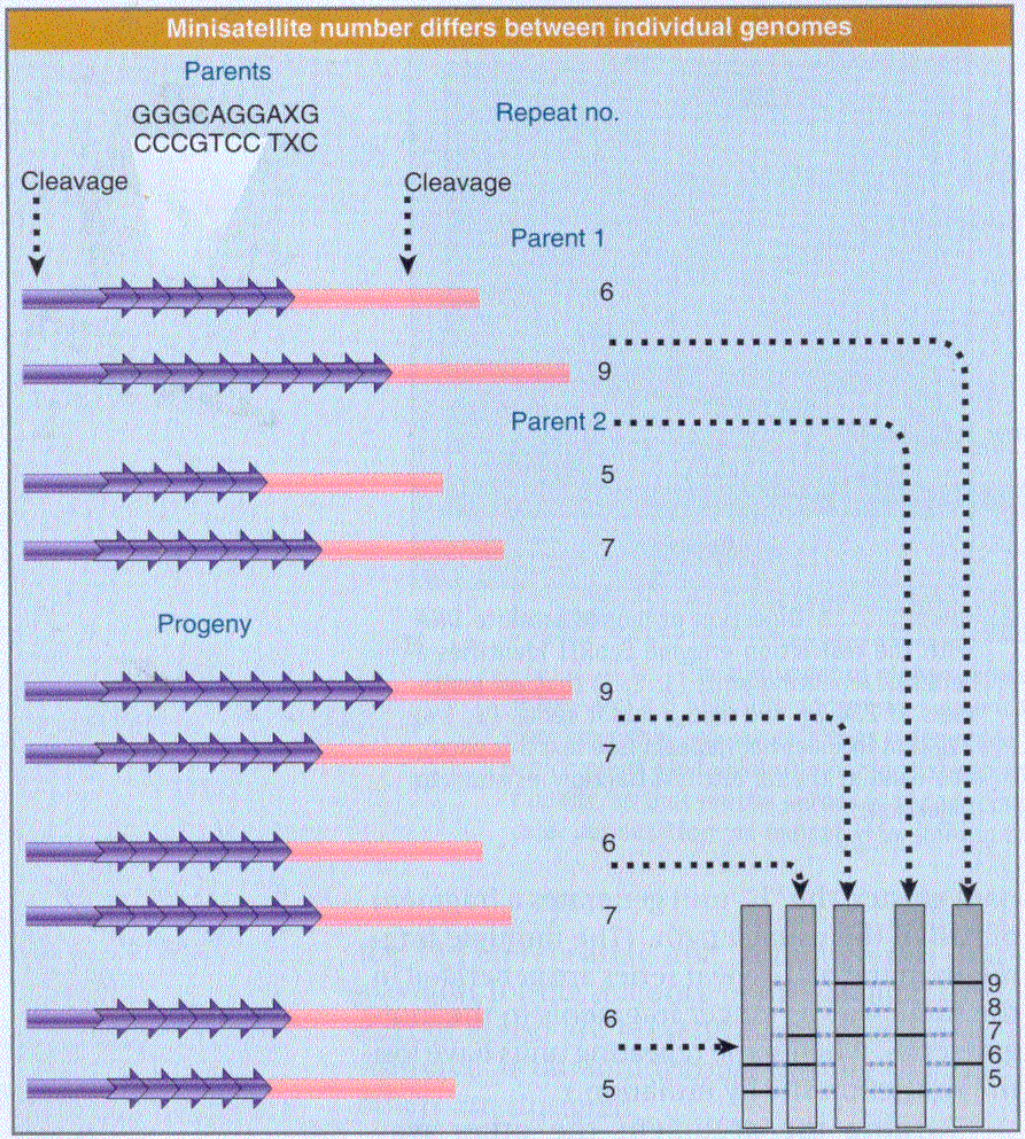



FIGURE 6.29 Alleles may differ in the number of repeats at a minisatellite locus, so that cleavage on either side generates restriction fragments that differ in length. By using a minisatellite with alleles that differ between parents, the pattern of inheritance can be followed.

Menos que 2 % do genoma humano codifica proteína.

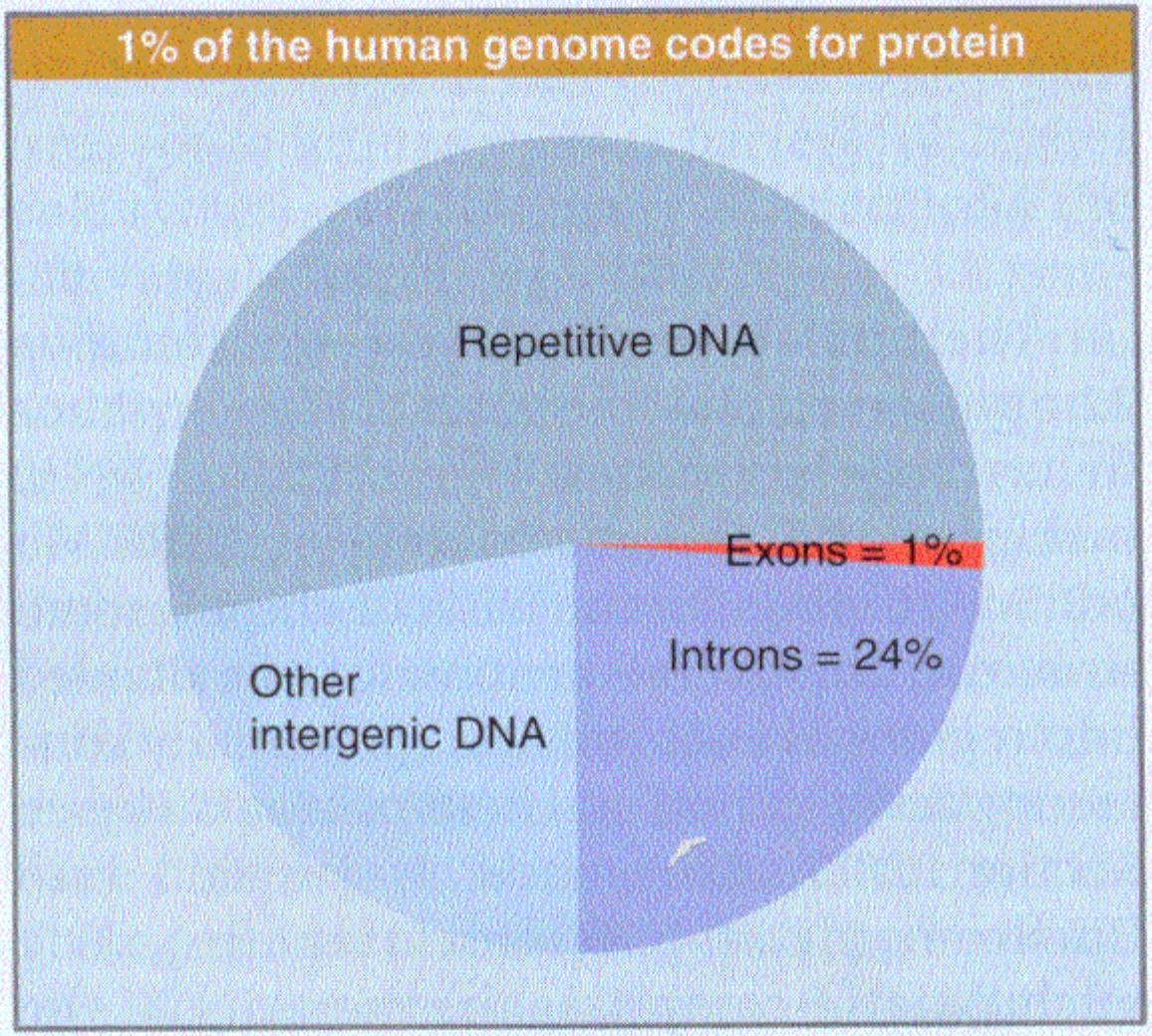
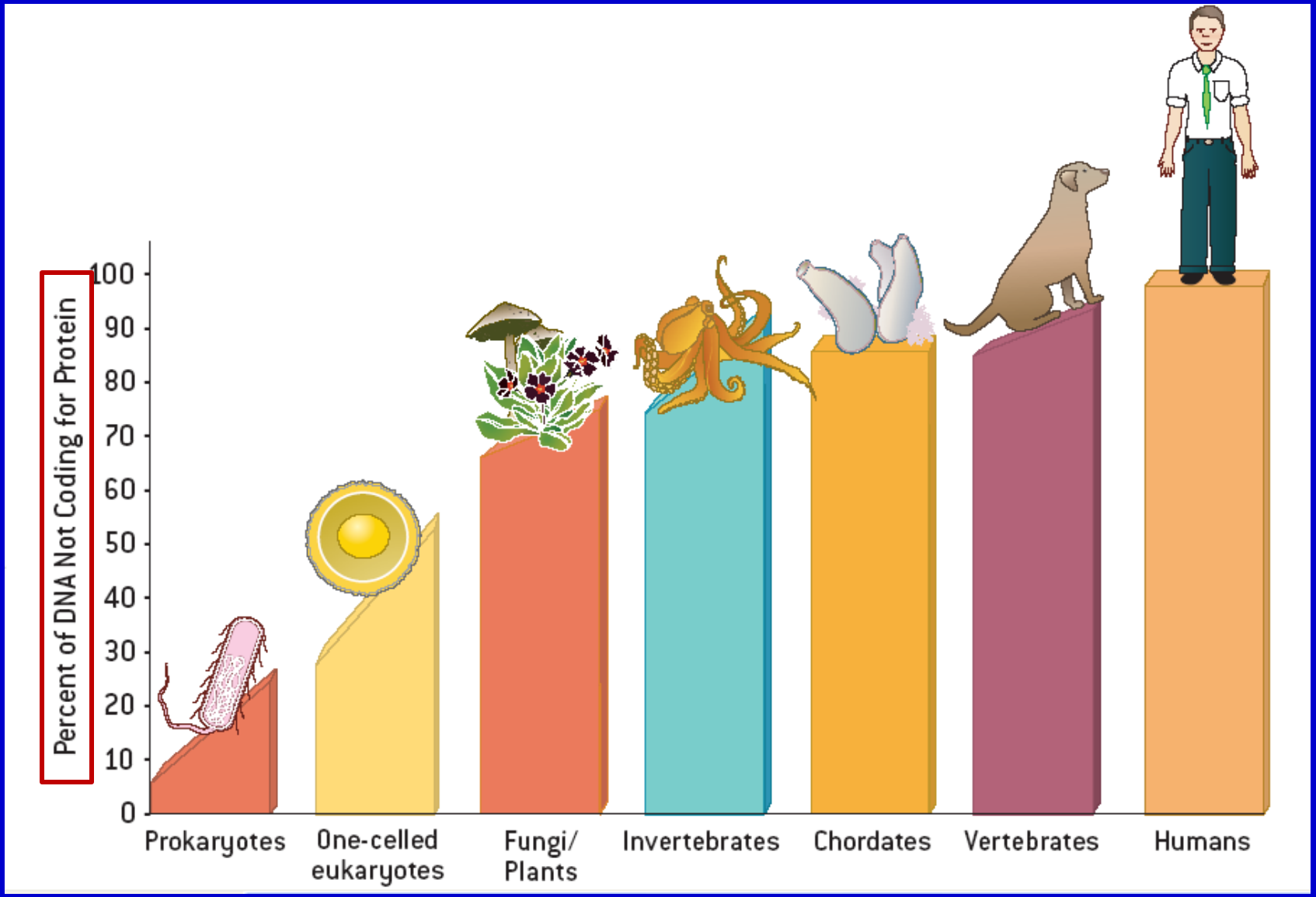



**FIGURE 5.11** Genes occupy 25% of the human genome, but protein-coding sequences are only a tiny part of this fraction.

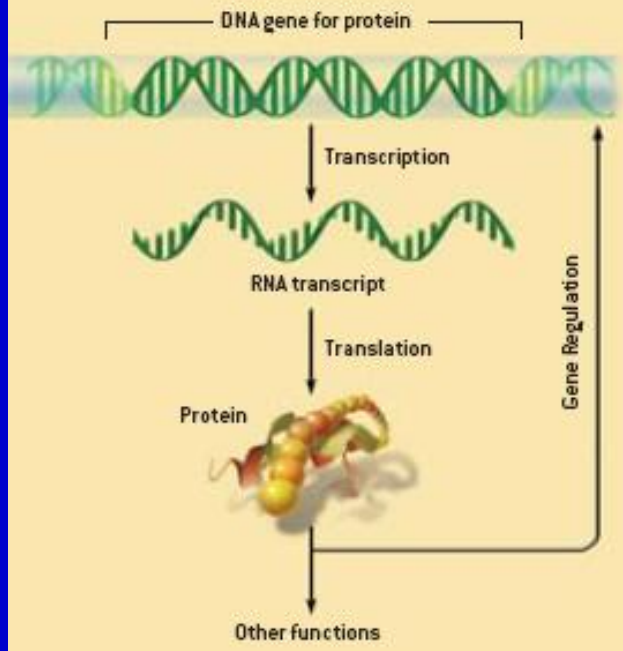
# A quantidade de DNA não codificador está relacionada a complexidade dos organismos



# Visão tradicional da atividade gênica

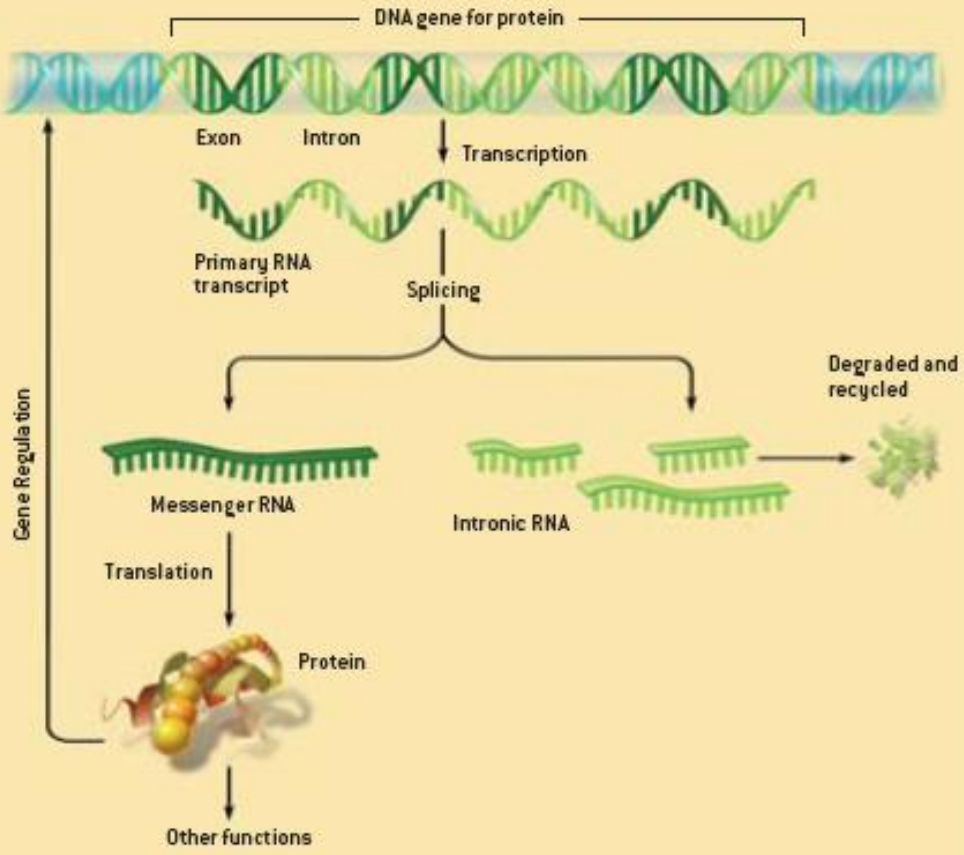
## GENE ACTIVITY IN PROKARYOTES

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.

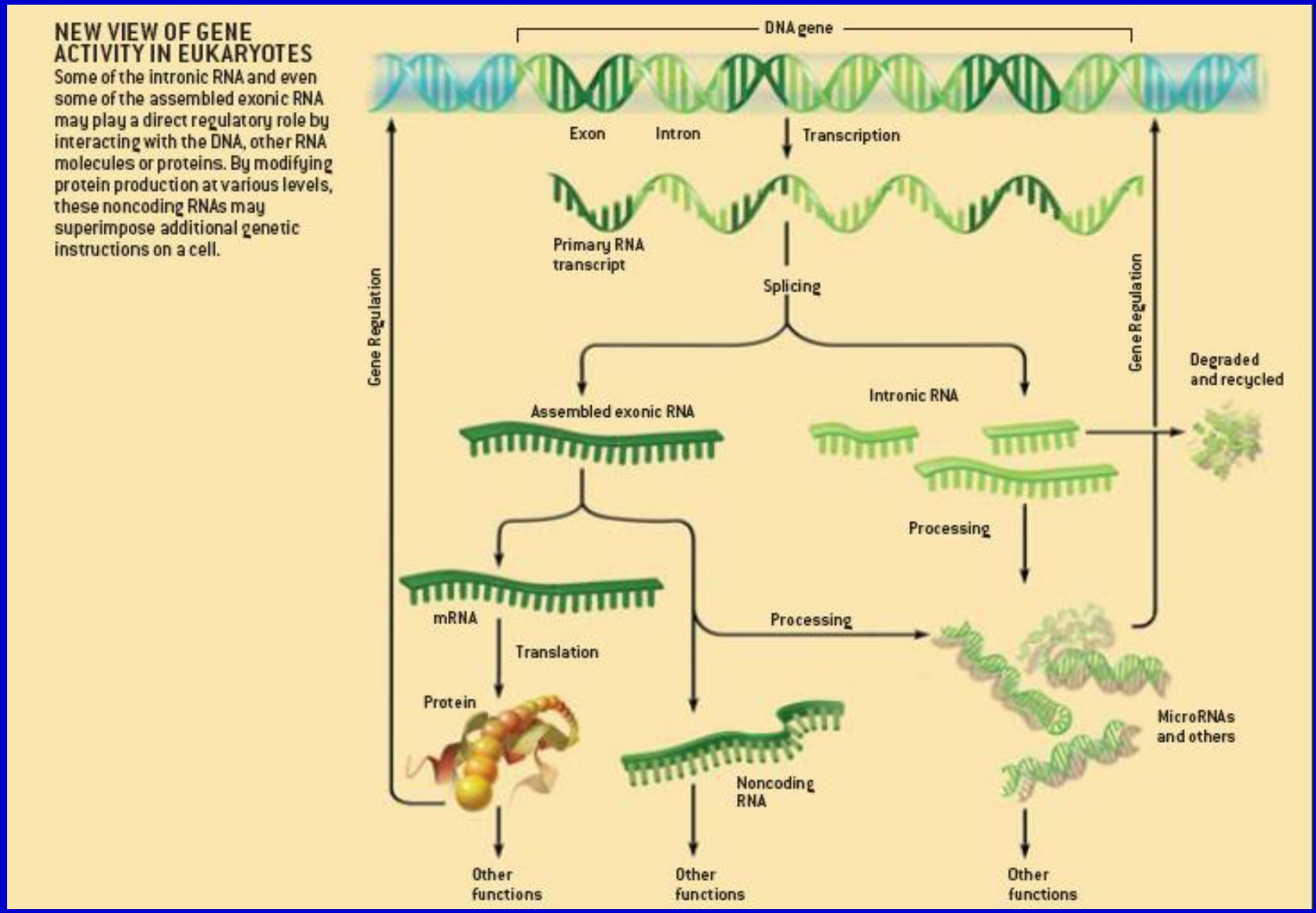



## TRADITIONAL VIEW OF GENE ACTIVITY IN EUKARYOTES

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.

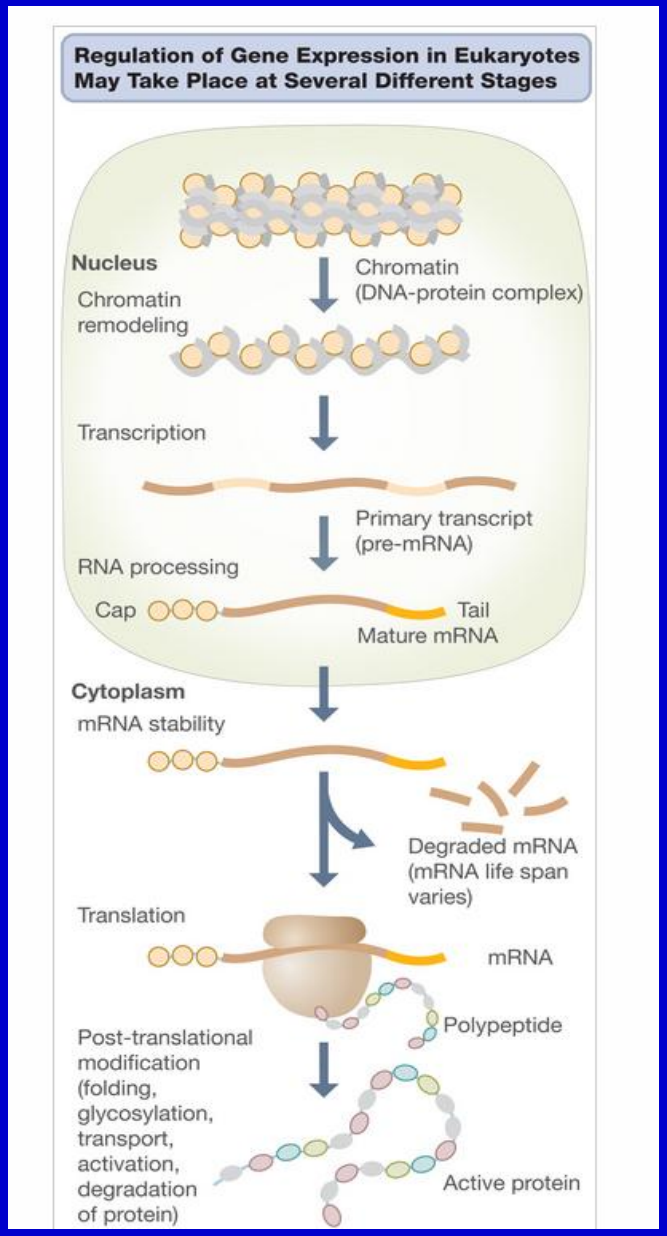


# Visão atual da atividade gênica



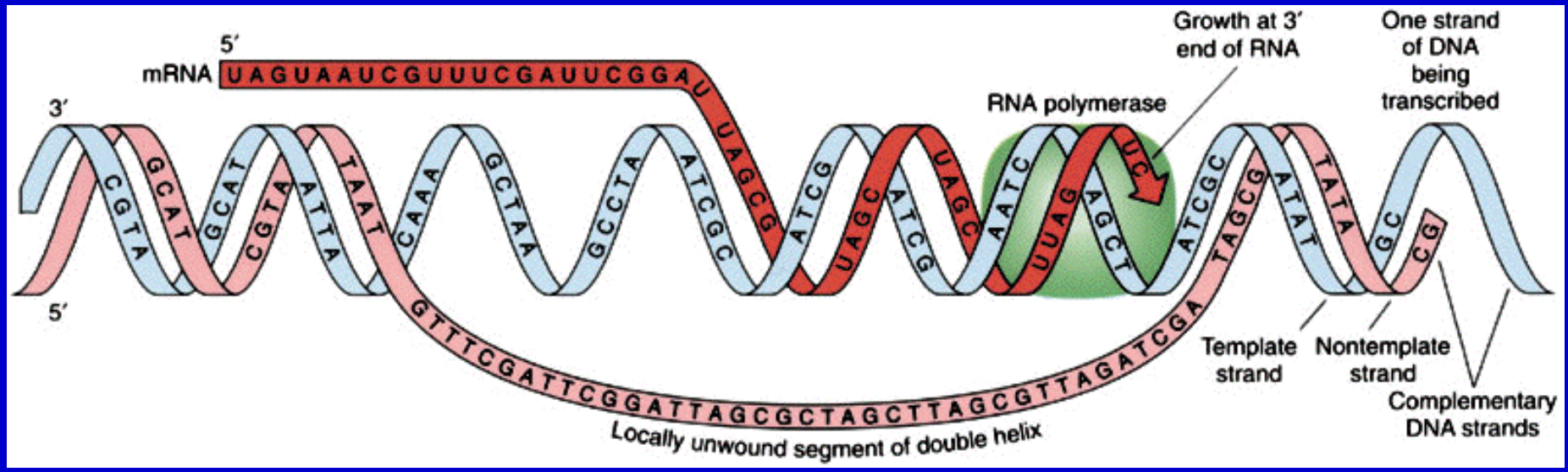
# Papel do RNA não codificador (ncRNA) na expressão gênica

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




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

# A síntese do mRNA é fiel ao DNA

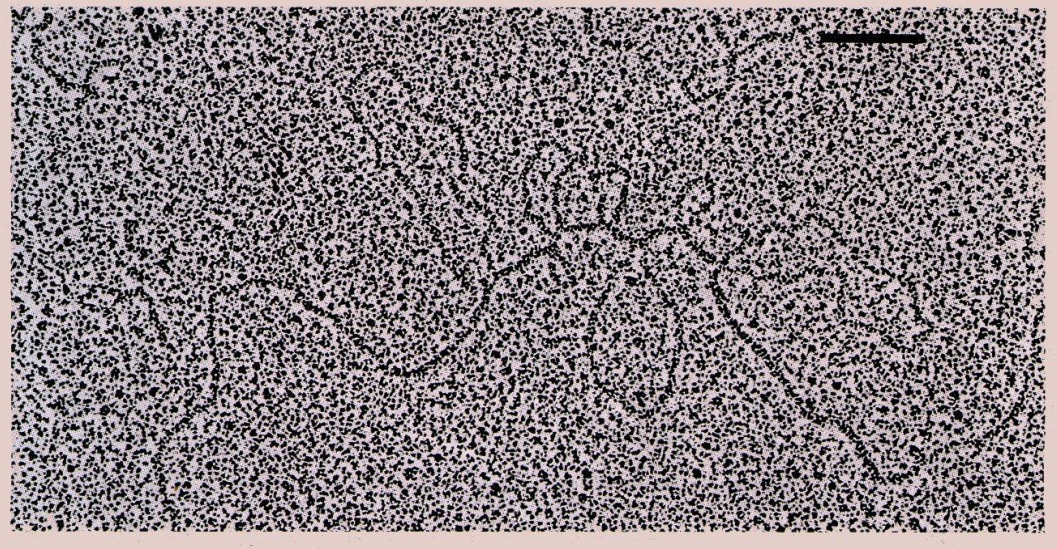


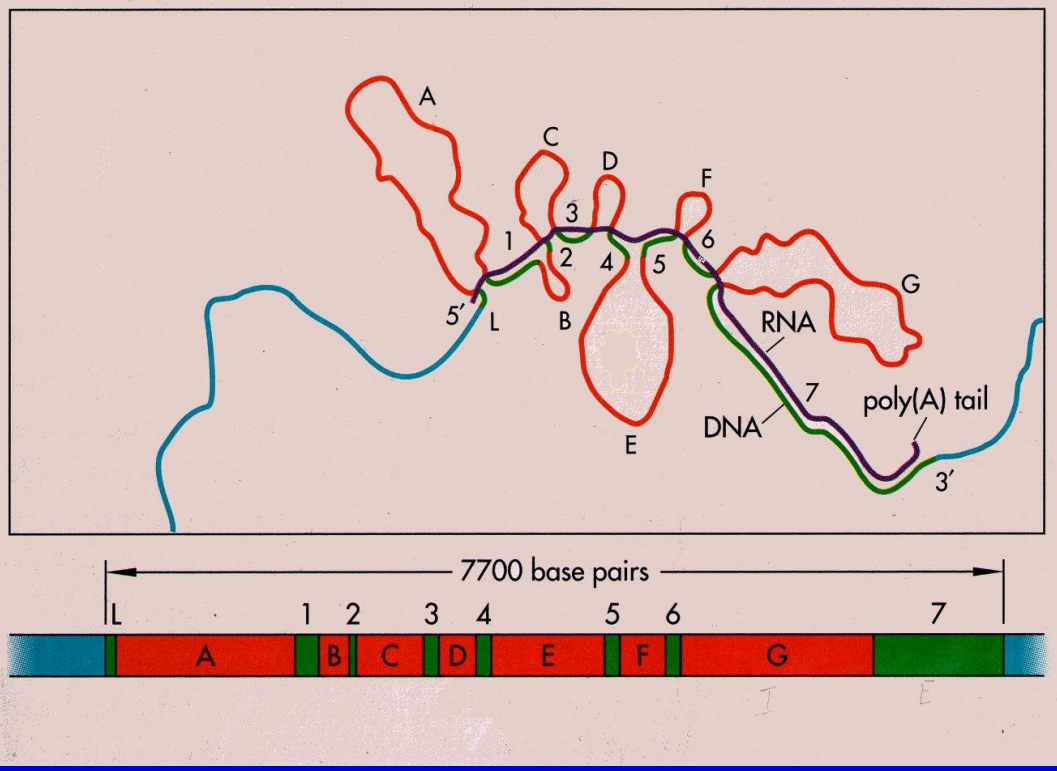

| | | |
|---|---|---|
| Nontemplate strand 5′ – | CTGCCATTGTCAGACATGTATACCCCGTACGTCTTCCCGAGCGAAAACGATCTGCGCTGC | – 3′ } DNA |
| Template strand 3′ – | GACGGTAACAGTCTGTACATATGGGGCATGCAGAAGGGCTCGCTTTTGCTAGACGCGACG | – 5′ } |
| 5′ – | CUGCCAUUGUCAGACAUGUAUACCCCGUACGUCUUCCCGAGCGAAAACGAUCUGCGCUGC | – 3′ mRNA |

T ⇒ U

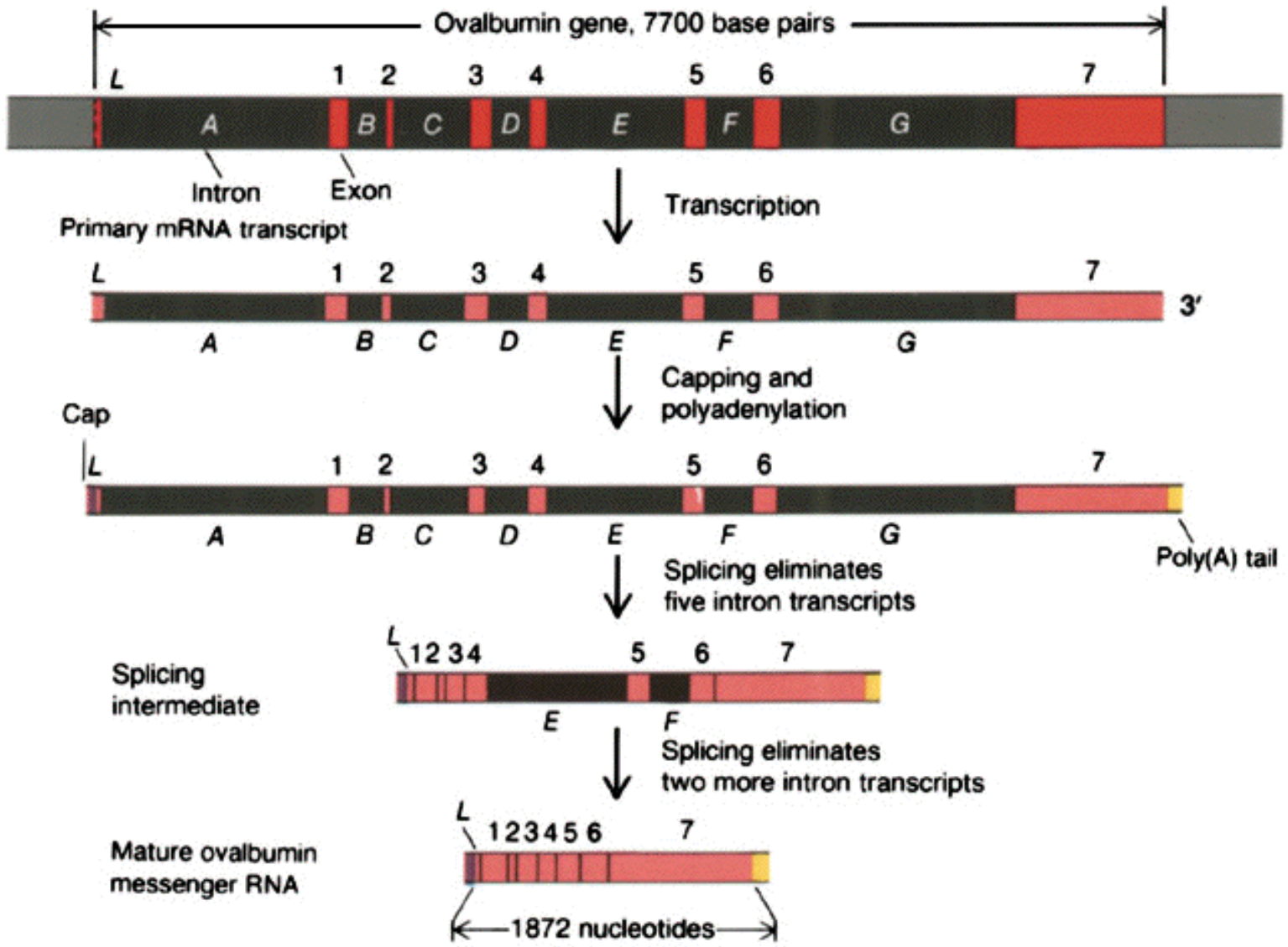
Descoberta do Processamento do RNA (Intron e Exon)

Hibridação DNA-RNA

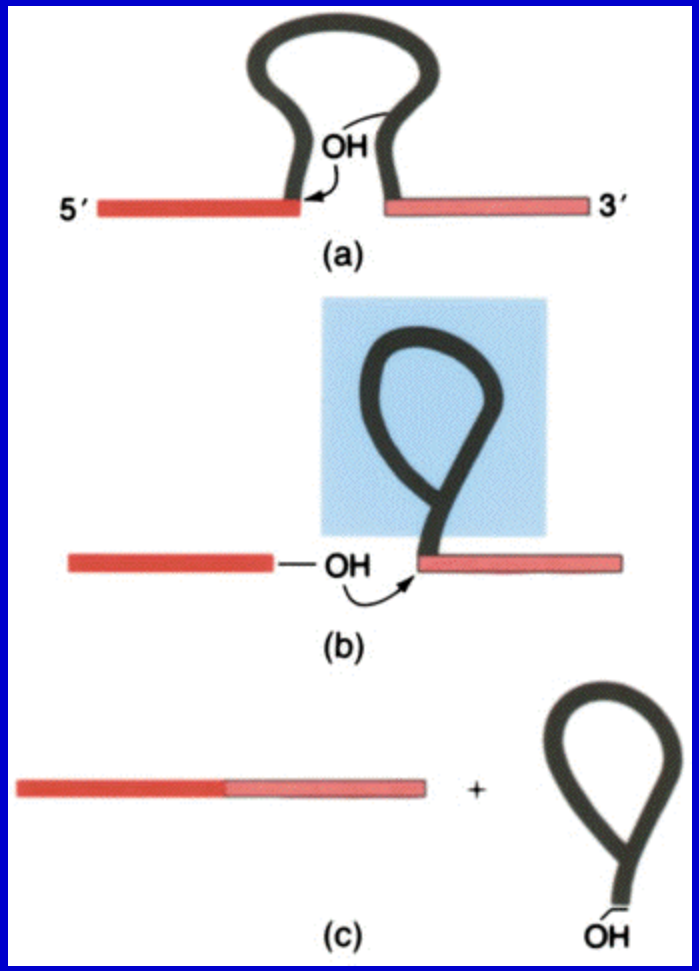




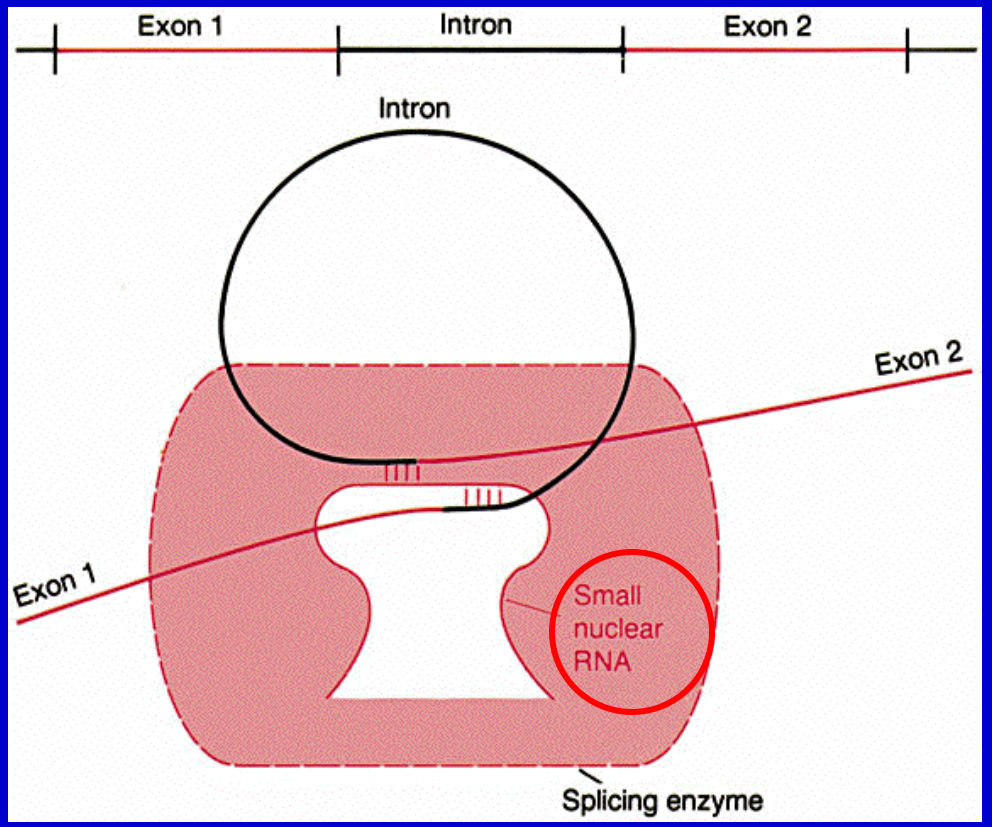
# Passos do processamento do RNA



# Mecanismos do processamento do RNA




**Auto *splicing***




**Processamento por *spliceossomo***

# Processamento alternativo do RNA

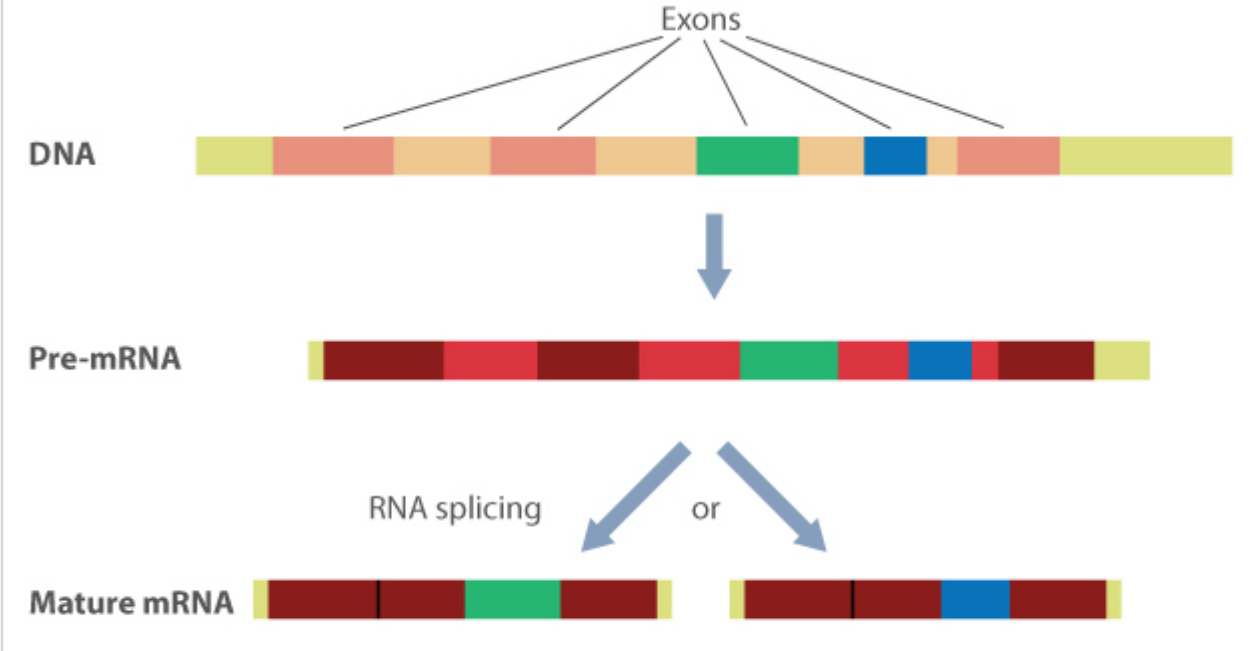
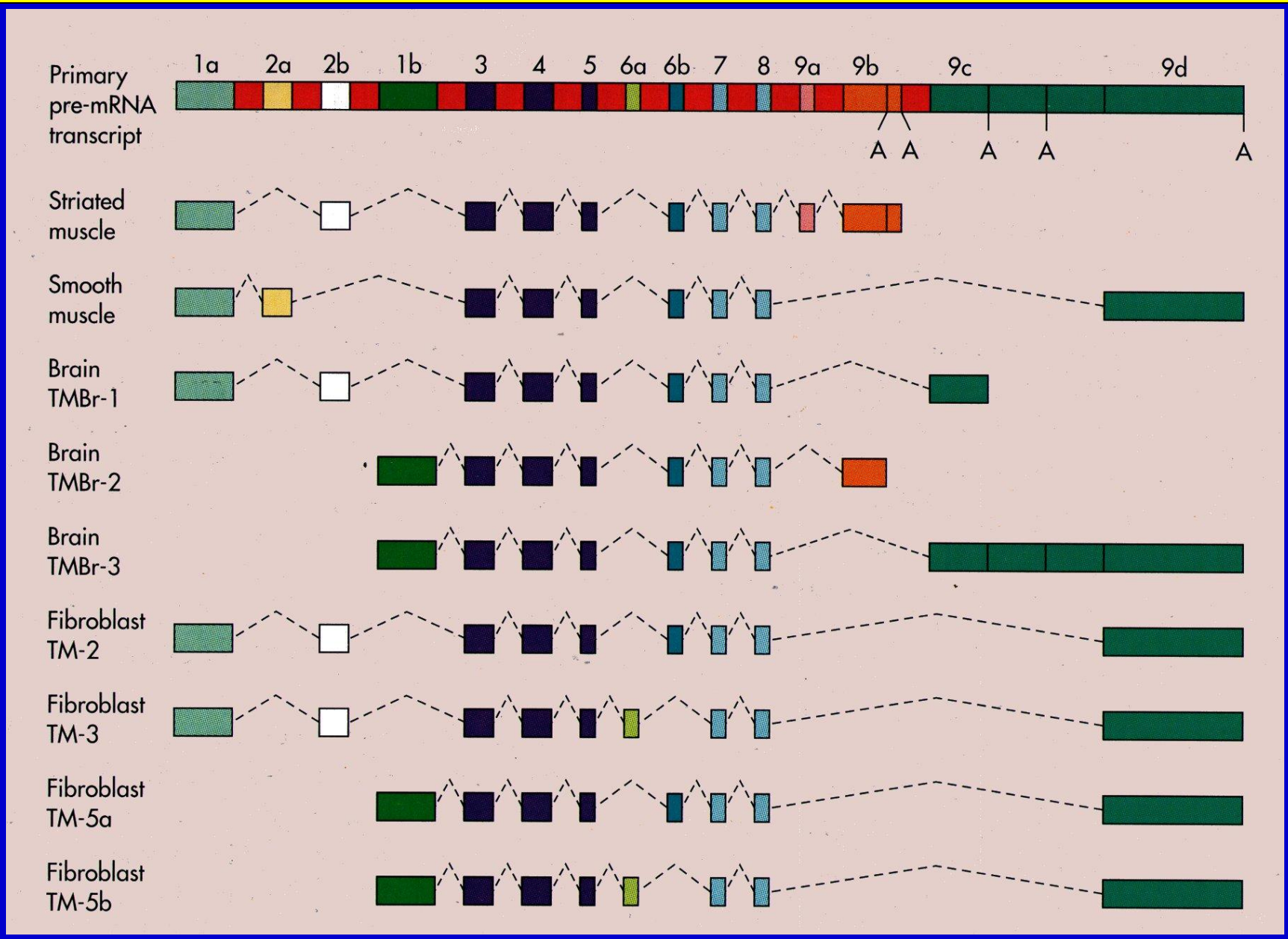


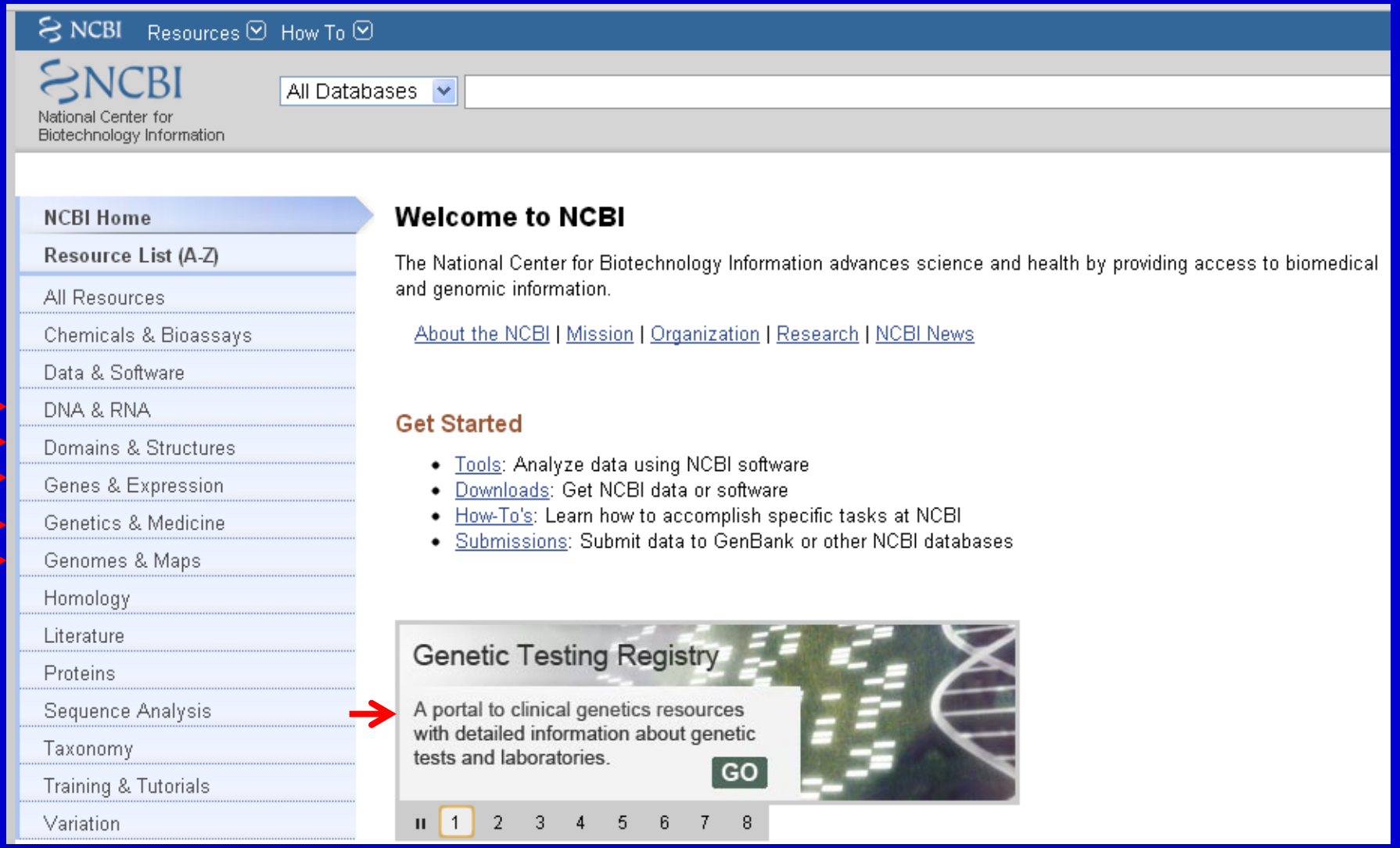

**Figure 6: Alternative RNA splicing.**
Production of two different mRNAs from the same gene.

Produz 2 diferentes RNAs a partir do mesmo gene

# Processamento diferencial do gene da tropomiosina



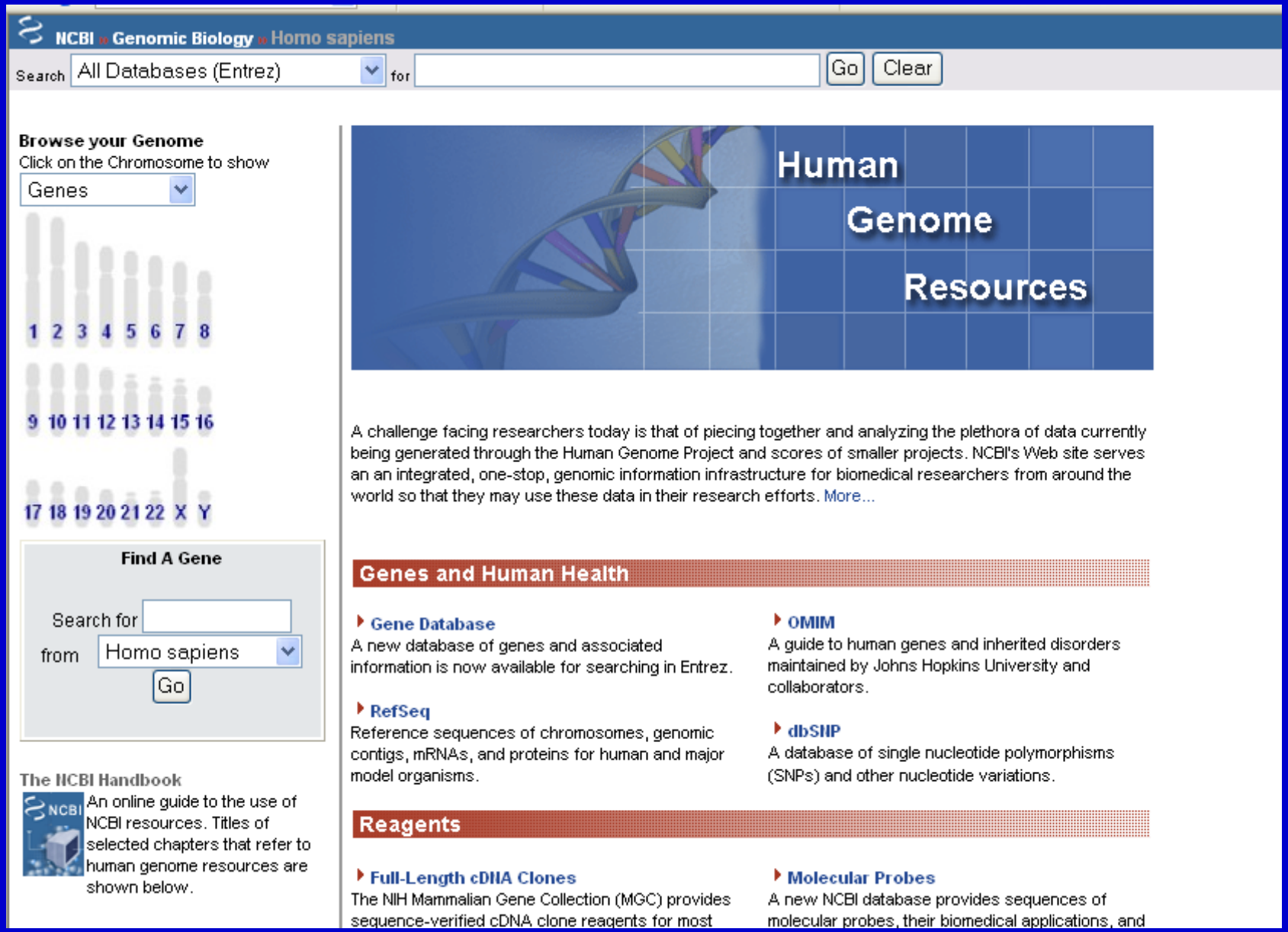
# Sequências de DNA são submetidas ao NCBI




National Center for Biotechnology Information

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

# NCBI: recurso para pesquisar doenças genética



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

- 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 re eutils) will no longer be supported.

## OMIM® - Online Mendelian Inheritance in Man®

Welcome to OMIM®, Online Mendelian Inheritance in Man®. OMIM is a comprehensive, aut human genes and genetic phenotypes. The full-text, referenced overviews in OMIM contain inf disorders and over 12,000 genes. OMIM focuses on the relationship between phenotype and ge 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 mende Mendelian Inheritance in Man (MIM). Twelve book editions of MIM were published between OMIM, was created in 1985 by a collaboration between the National Library of Medicine and t at Johns Hopkins. It was made generally available on the internet starting in 1987. In 1995, OM


## BLAST Assembled RefSeq Genomes

Choose a species genome to search, or **list all genomic BLAST databases**.

- **Human**
- **Mouse**
- **Rat**
- ***Arabidopsis thaliana***
- ***Oryza sativa***
- ***Bos taurus***
- ***Danio rerio***
- ***Drosophila melanogaster***
- ***Gallus gallus***
- ***Pan troglodytes***
- ***Microbes***
- ***Apis mellifera***

## Basic BLAST

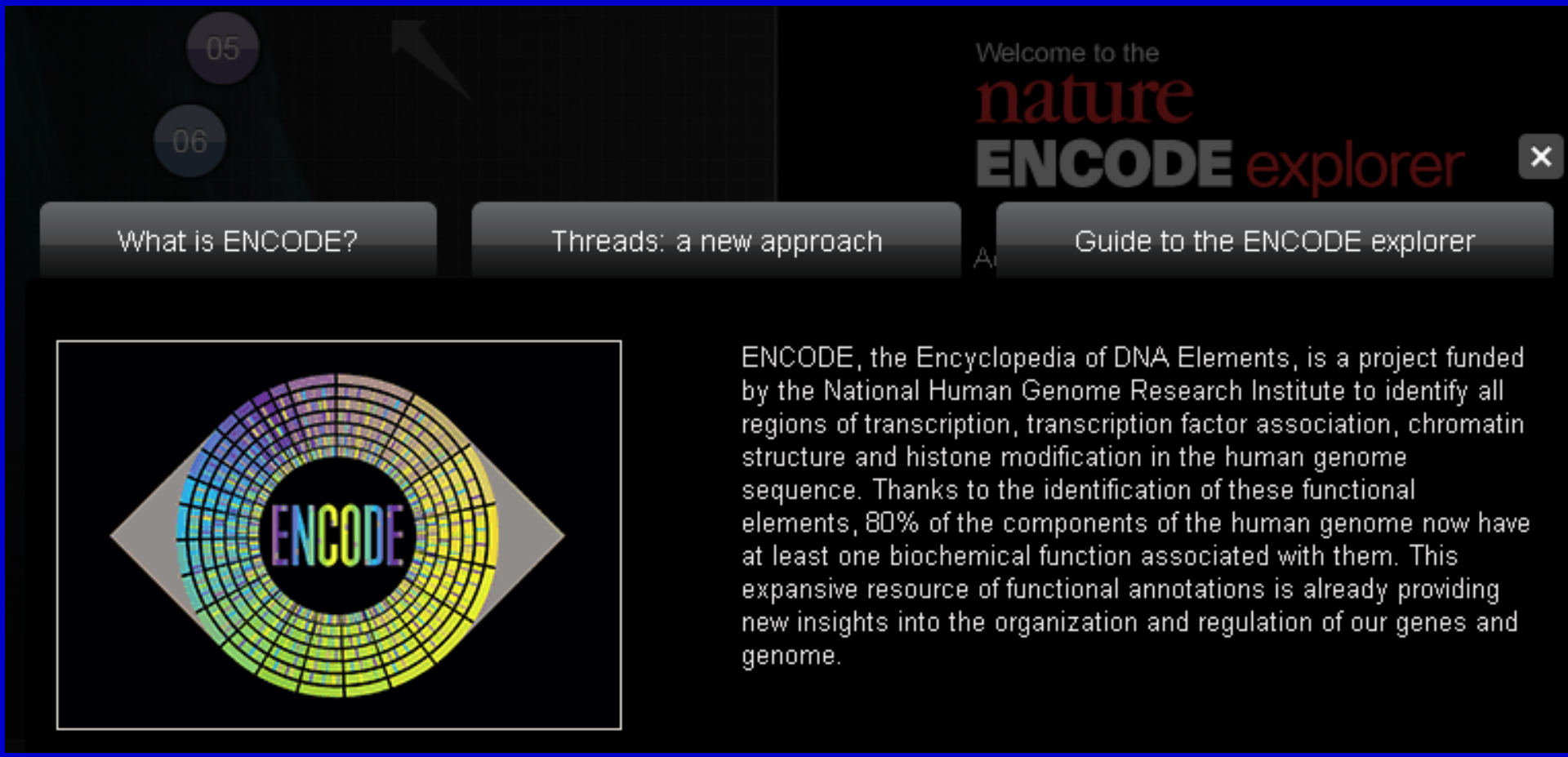
Choose a BLAST program to run.

| | |
|---|---|
| **nucleotide blast** | Search a **nucleotide** database using a **nucleotide** query<br>*Algorithms:* blastn, megablast, discontiguous megablast |
| **protein blast** | Search **protein** database using a **protein** query<br>*Algorithms:* blastp, psi-blast, phi-blast, delta-blast |
| **blastx** | Search **protein** database using a **translated nucleotide** query |
| **tblastn** | Search **translated nucleotide** database using a **protein** query |
| **tblastx** | 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




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

# Bibliografia

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.