PRINCIPLES AND METHODS OF MOLECULAR BIOLOGY
LECTURE 1: INTRODUCTION TO MOLECULAR BIOLOGY

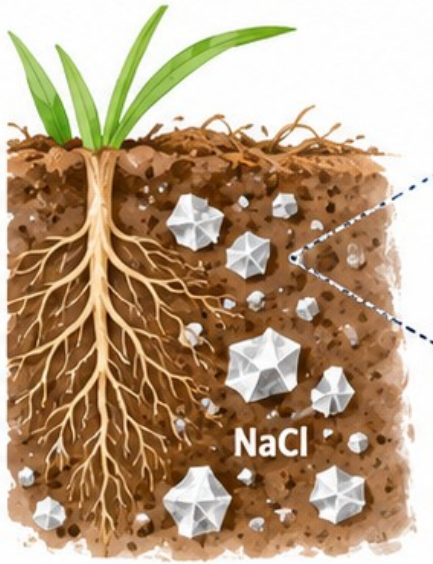
Gulzhanay K. PhD

A Molecular Biology Question

From a salty soil microbe to molecular insights for plant stress tolerance

Saline soil

High salt concentration challenges plant growth



Unknown bacterium from saline soil



Unknown bacterium (isolated from saline soil)

- **Saline soil** (e.g. coastal or arid regions)
- Rich in salt (NaCl)

Plant under salt stress (without bacterium)



- Stunted growth
- Yellowing leaves
- Reduced root development

Improves plant growth under stress

Plant under salt stress (with bacterium)



- Greater biomass
- Greener leaves
- More extensive roots
- Better stress tolerance

Key molecular biology questions:

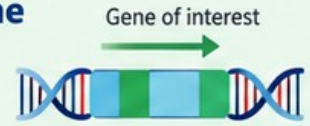
1 How can we identify this bacterium?

e.g. 16S rRNA gene sequencing, phylogenetic analysis



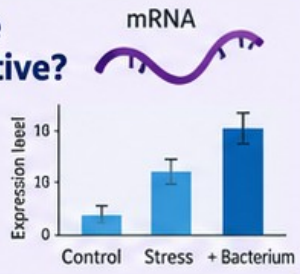
2 How can we determine whether it contains a useful gene?

e.g. genome sequencing, gene annotation, comparative analysis



3 How can we determine whether this gene is active?

e.g. RNA expression analysis (RT-qPCR, RNA-seq), protein detection



From question to answer:

Research question

Define the hypothesis

DNA / RNA

Isolate nucleic acids from the bacterium

Molecular method

e.g. PCR, sequencing, RT-qPCR, RNA-seq

Data

Sequences, expression levels, *phylogenetic trees, etc.*

Interpretation

What do the data tell us? How does it explain improved plant growth?

Principles and Methods of Molecular Biology
Lecture 1–2: Introduction to Molecular Biology



DNA, RNA and the Flow of Genetic Information

Today we will discuss:
What is molecular biology?

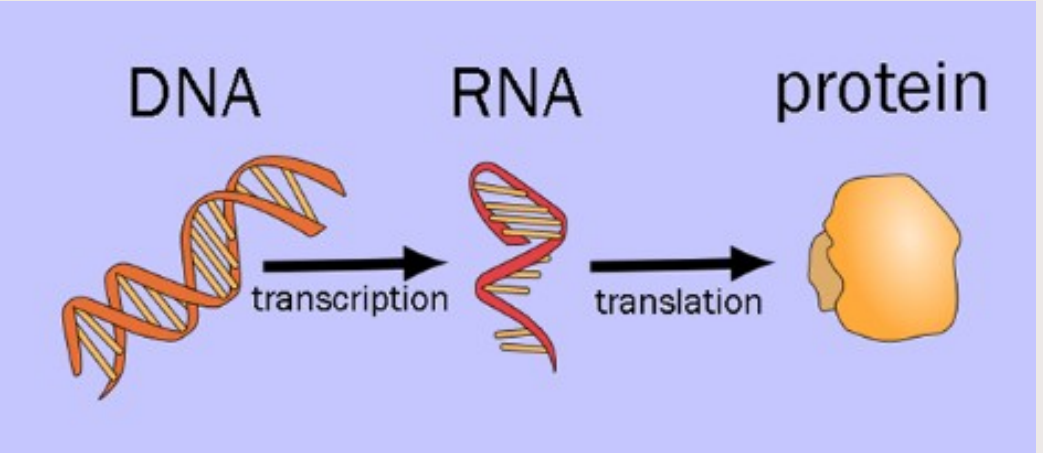
What are genes and genomes?

DNA and RNA

Central dogma

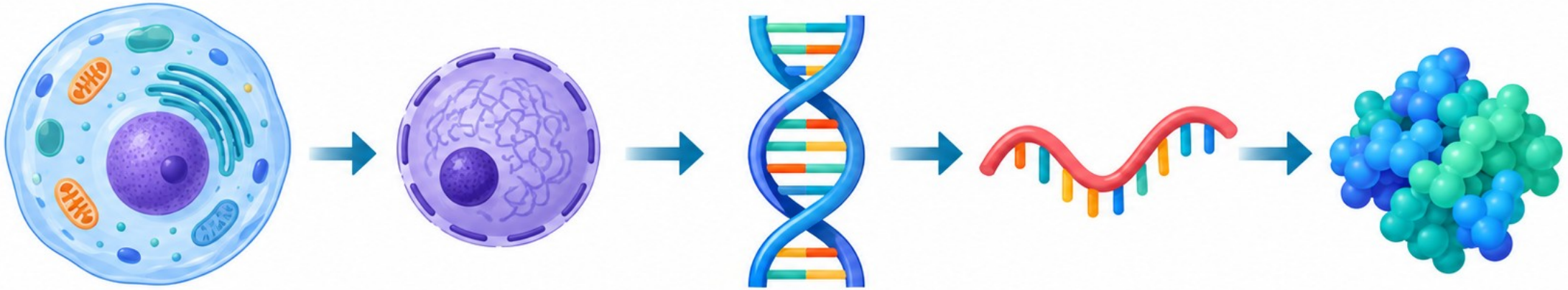
Replication, transcription and translation

How to choose a molecular method



What is Molecular Biology?

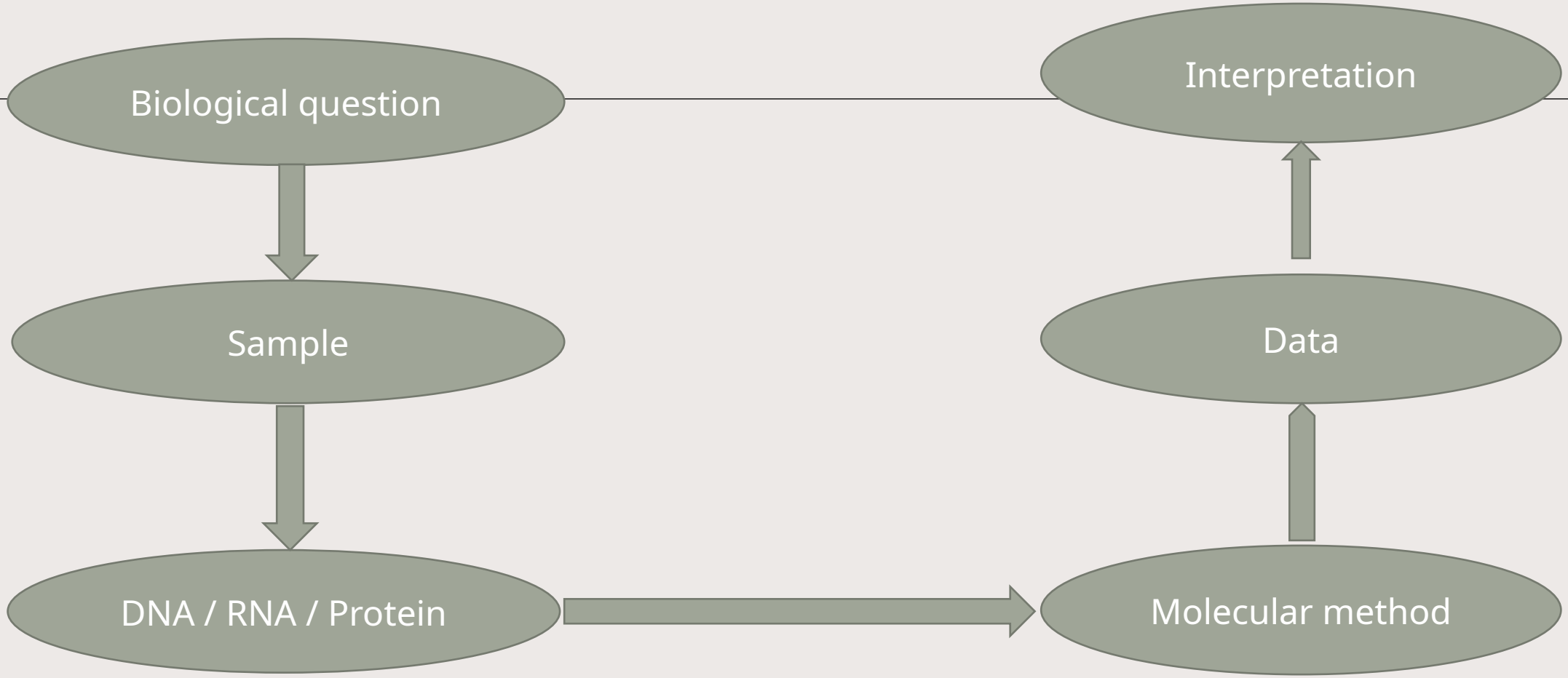
The study of DNA, RNA, proteins, and how they control life.

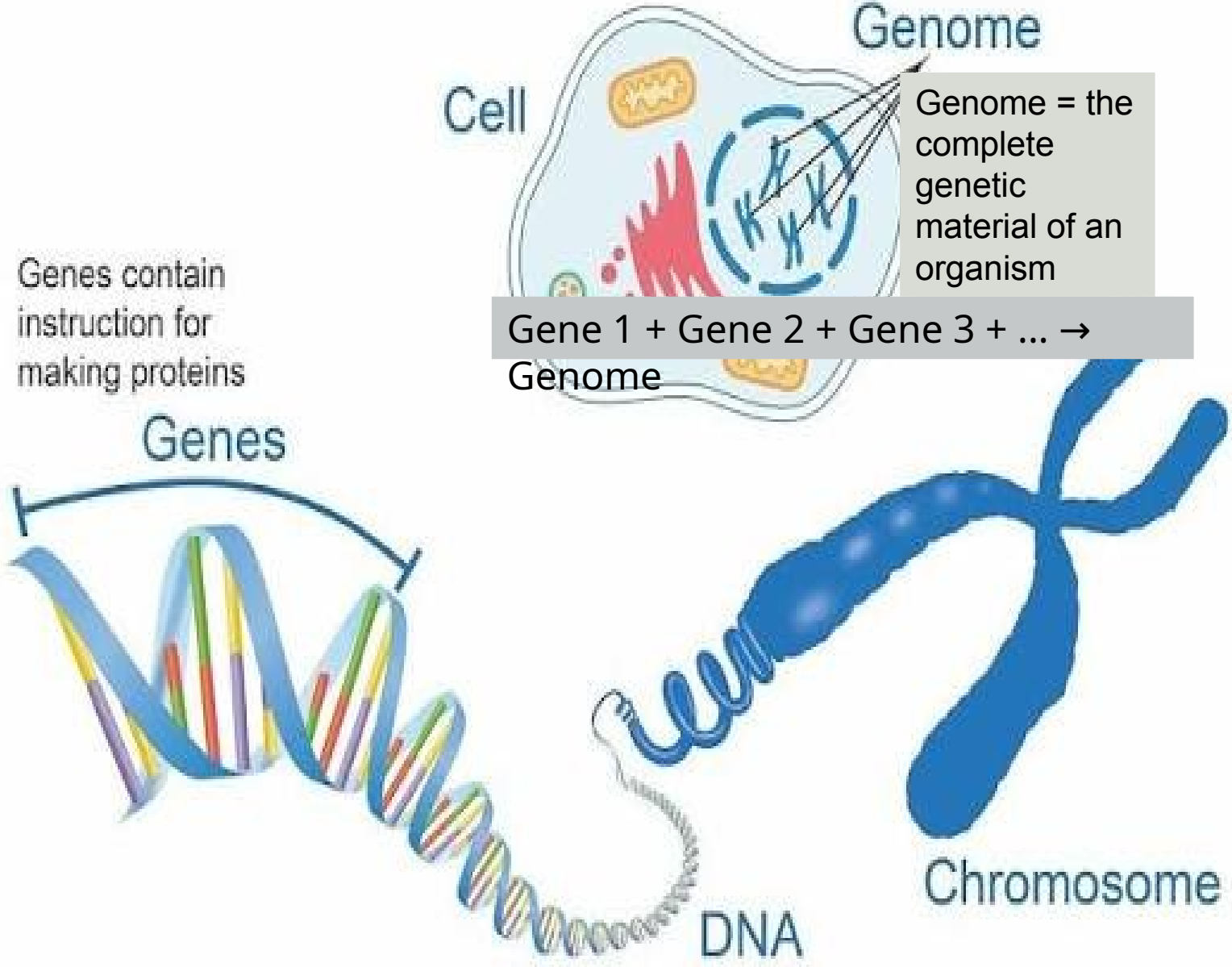


Molecular biology explains how genetic information is stored, expressed, and used in living cells.

Genetic information
Gene expression
Molecular mechanisms

FROM A BIOLOGICAL QUESTION TO AN EXPERIMENT

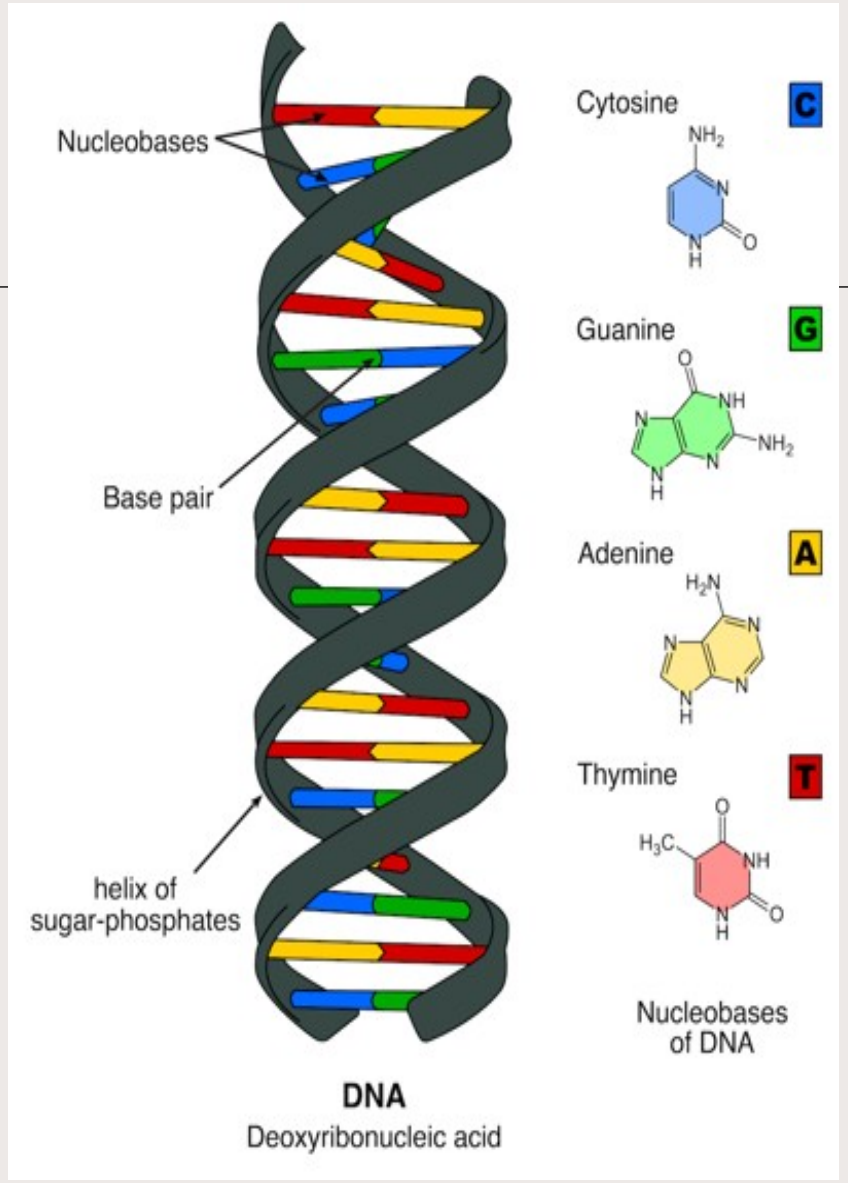




Gene → Protein
Or
Gene → Functional RNA

acdS → ACC deaminase
16S rRNA gene → ribosomal RNA

acdS is found in some beneficial bacteria and encodes ACC deaminase, an enzyme that can help plants tolerate stress by lowering ACC/ethylene levels.



DNA stores genetic information

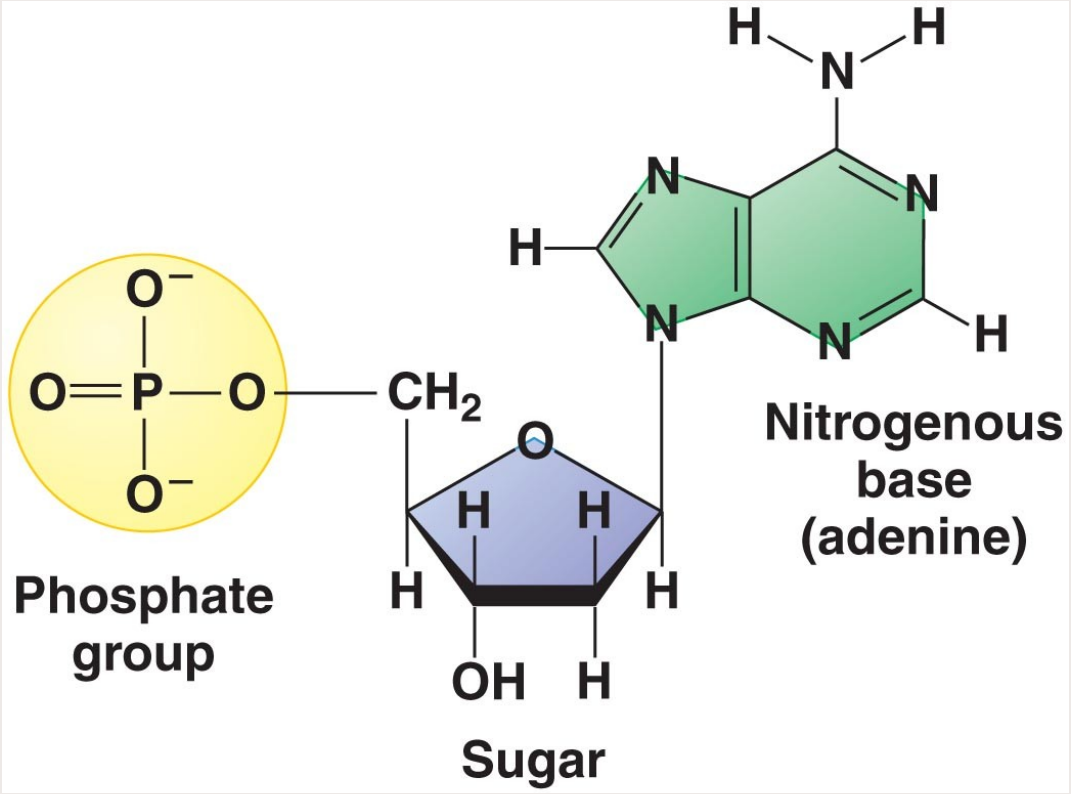
Deoxyribonucleic acid

Usually double-stranded

Made of nucleotides

Contains A, T, G and C

DNA Is Made of Nucleotides



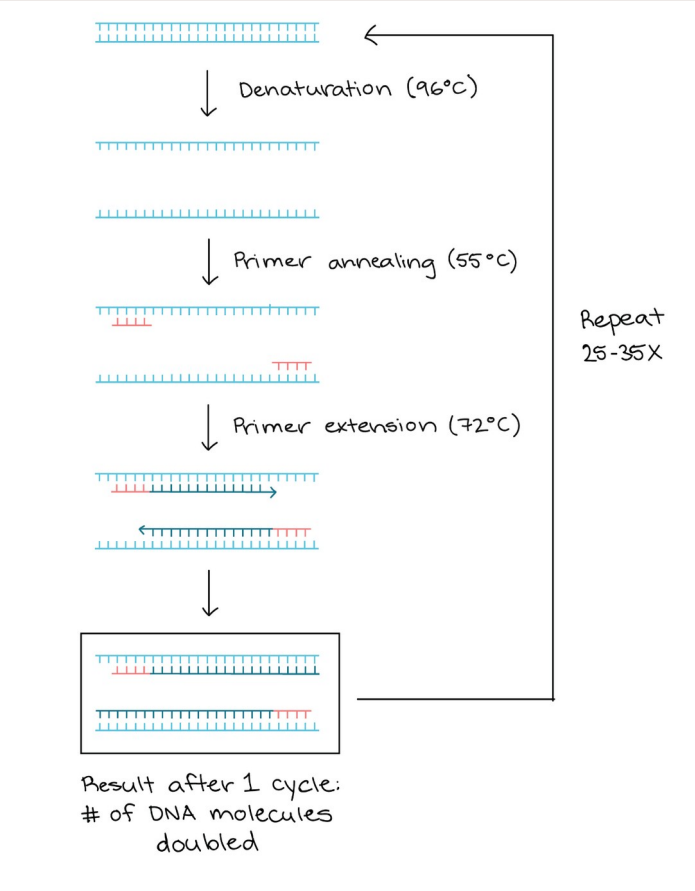
Complementarity allows specific molecular recognition

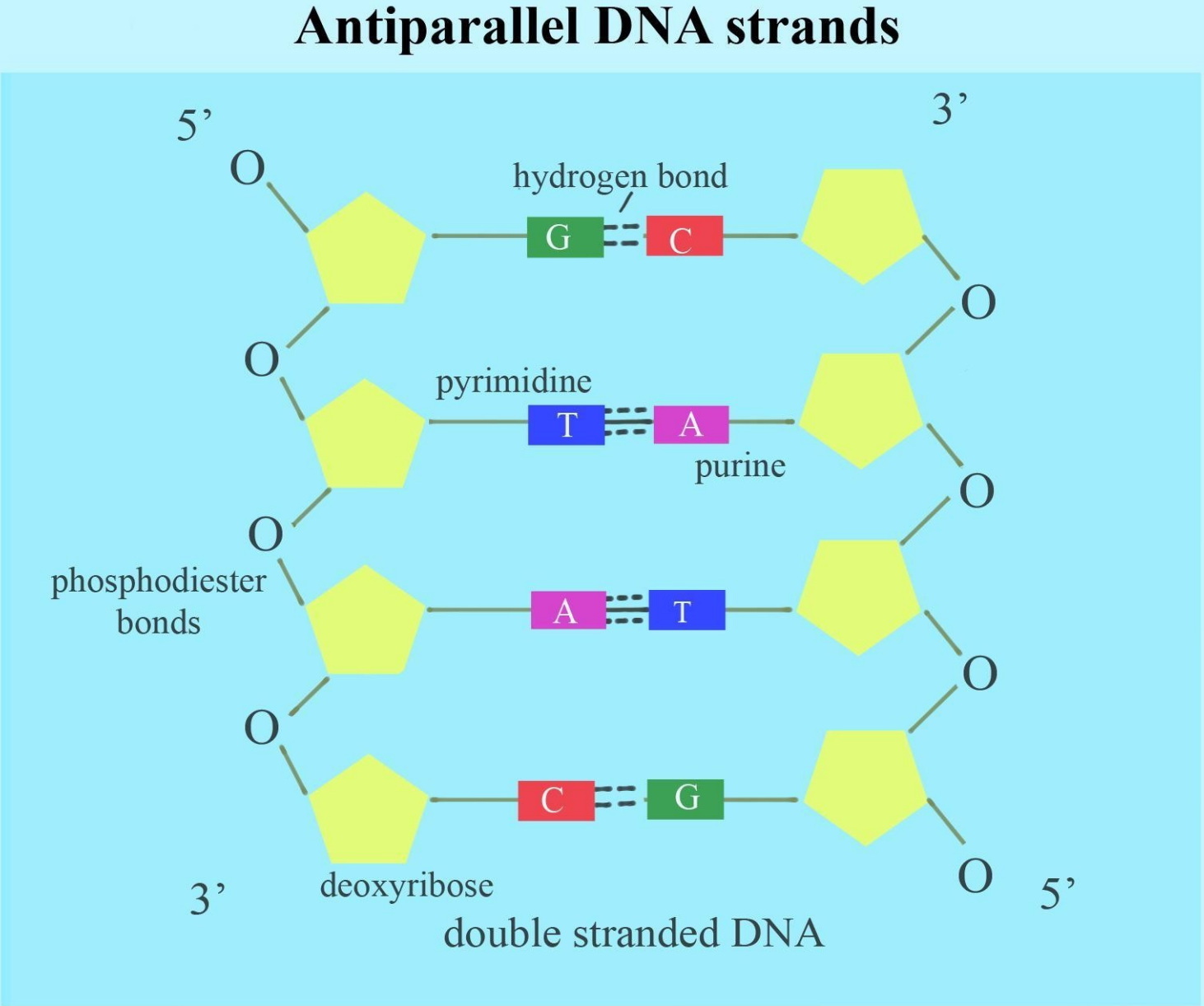
PCR

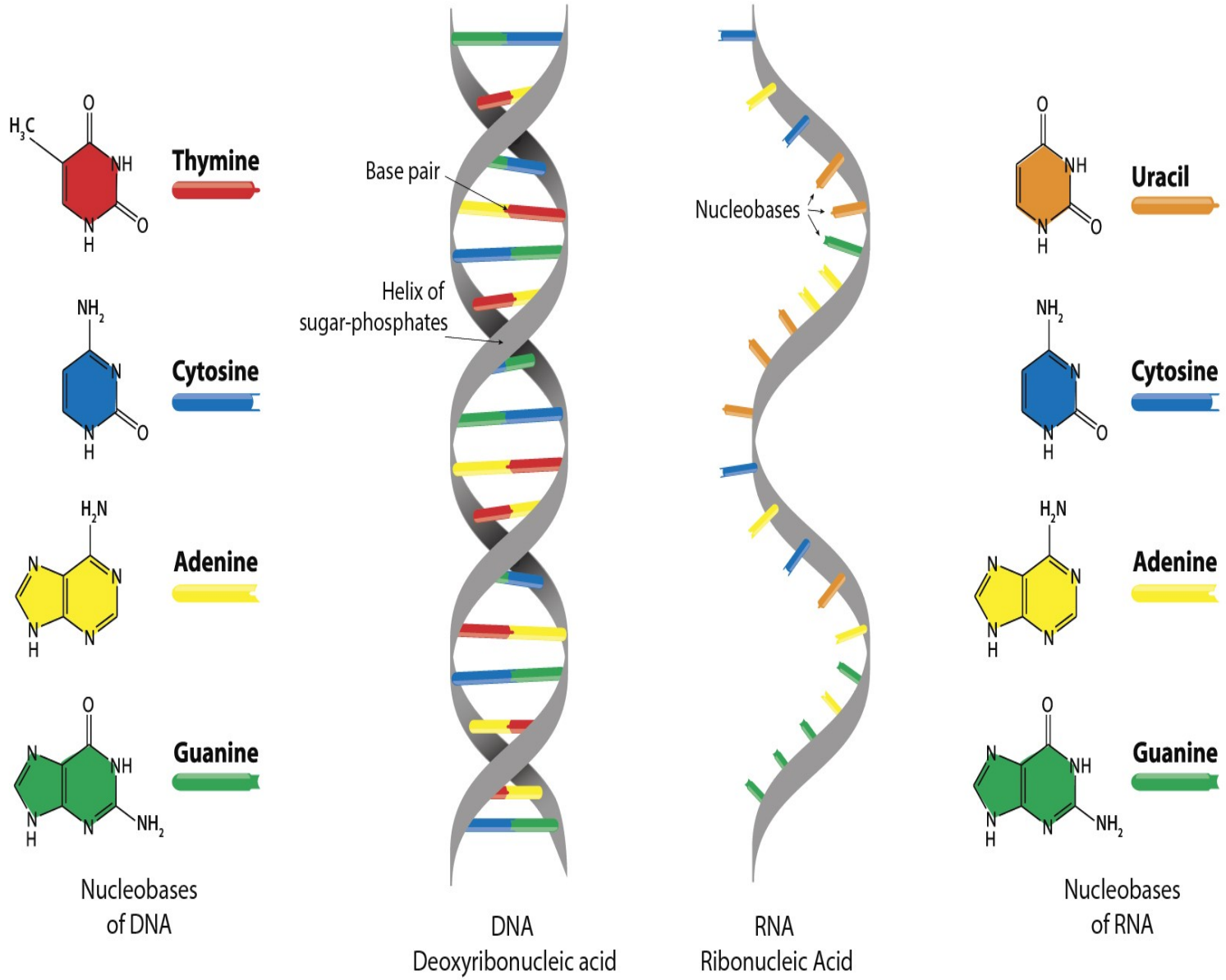
Primers

Sequencing

Hybridization



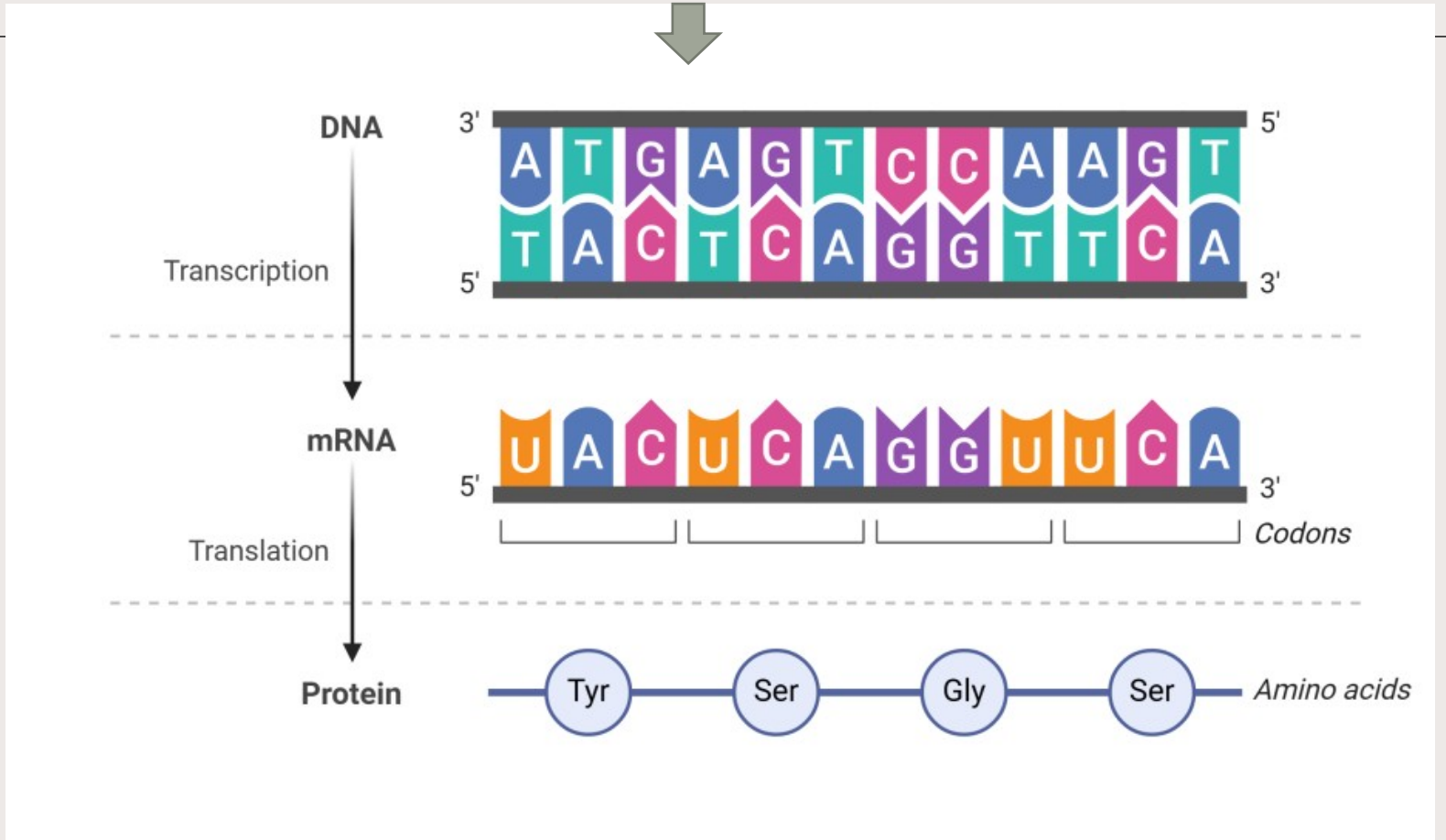




mRNA — messenger RNA
Carries genetic information
rRNA — ribosomal RNA
Part of the ribosome
tRNA — transfer RNA
Carries amino acids

The central dogma

Replication DNA → DNA

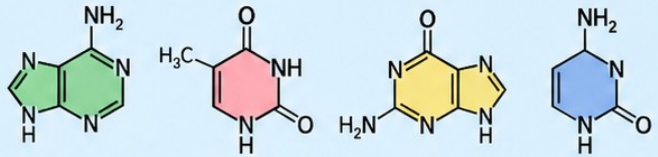


From Base to Nucleotide to DNA, and What Is a Codon?

Key concepts and how they are related

1. Nitrogenous bases

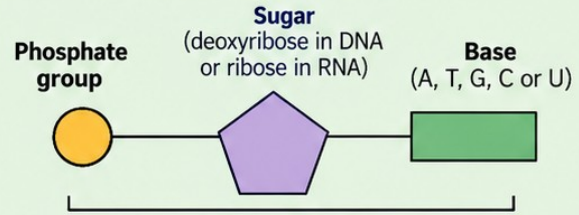
There are four bases in DNA (and four in RNA).



Adenine (A) | **Thymine (T)** (DNA) | **Guanine (G)** | **Cytosine (C)**

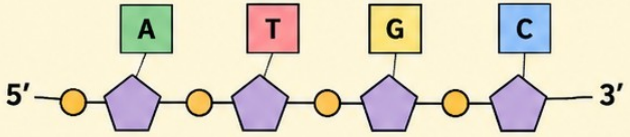
2. Nucleotide

A nucleotide consists of three parts.



3. DNA strand

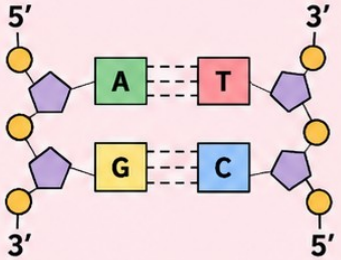
Nucleotides are linked together to form a strand of DNA.



Each nucleotide contains one base (A, T, G or C).

4. Base pairing

In double-stranded DNA, bases pair specifically.



A pairs with **T**
G pairs with **C**

The two strands are antiparallel.

5. Codon (in mRNA)

A codon is a sequence of three nucleotides in mRNA.



Codon AUG = Methionine (start codon)

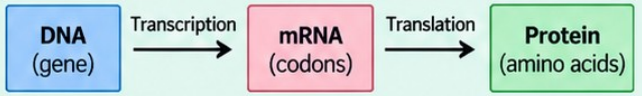
Codon UUU = Phenylalanine

Codon GCU = Alanine

Each codon consists of three nucleotides and usually codes for one amino acid or a stop signal.

6. From gene to protein

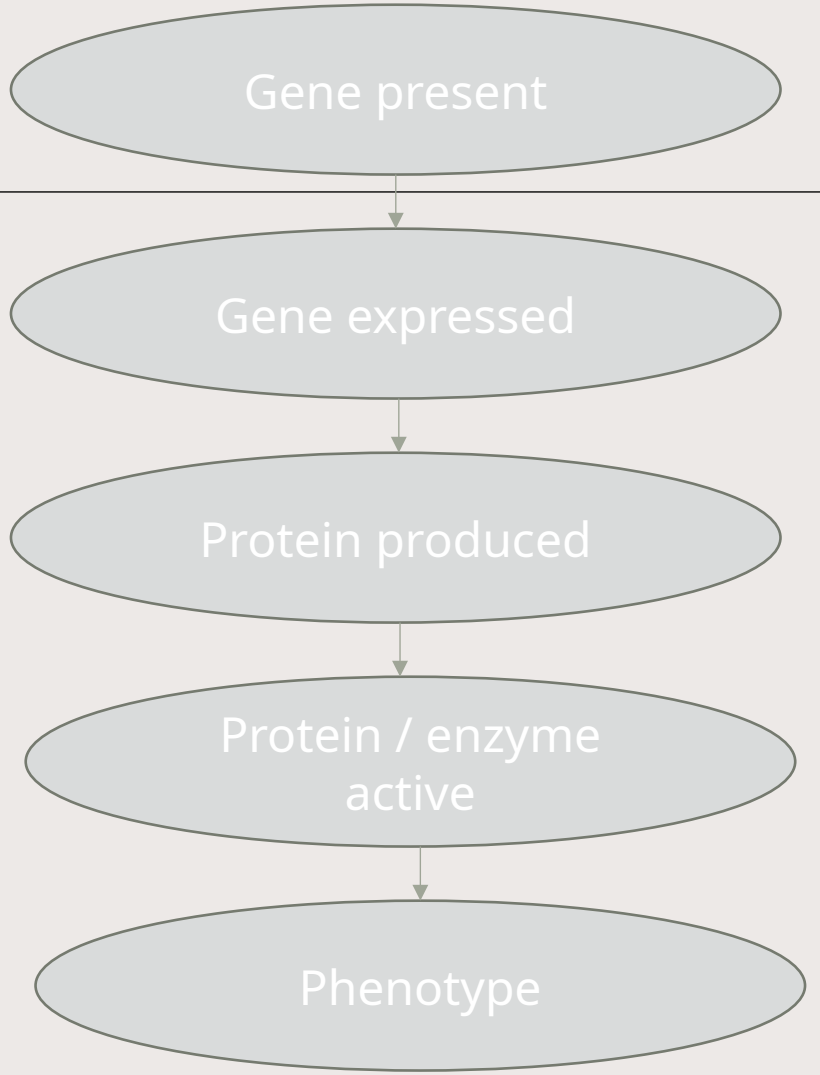
The sequence of nucleotides in a gene determines the sequence of amino acids in a protein.



Key points to remember

1. A base (A, T, G, C or U) is part of a nucleotide.
2. A nucleotide = phosphate + sugar + base.
3. DNA strands are made of many nucleotides.
4. In DNA, A pairs with T and G pairs with C.
5. A codon is three nucleotides in mRNA.
6. The sequence of nucleotides determines the sequence of amino acids in a protein.

Different Levels of Biological Evidence



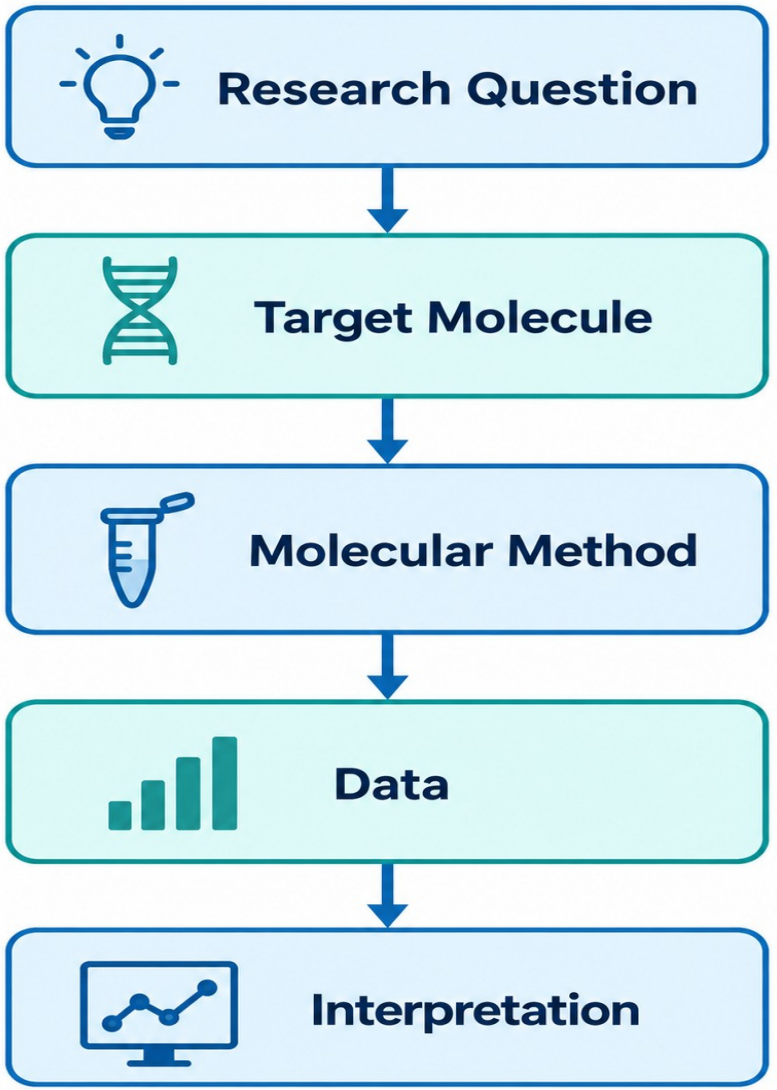
acdS → ACC deaminase
16S rRNA gene → ribosomal RNA

But

Presence ≠ Expression ≠ Function

Research question	Analyze
Is the gene present?	**DNA**
Is the gene expressed?	**RNA**
Is the protein produced?	**Protein**
Is the enzyme active?	**Activity**

FROM RESEARCH QUESTION TO METHOD



Is acdS present?
→ DNA
→ PCR
→ Band
→ Gene detected

CASE STUDY 1: UNKNOWN BACTERIUM

Research question:

What bacterium is this?

Workflow:

Bacterial isolate

↓

DNA extraction

↓

16S rRNA gene PCR

↓

Sequencing

↓

Database comparison / BLAST

↓

Identification

16S rRNA sequencing helps determine taxonomic identity or closest relatives, but it does not always reliably distinguish between closely related species or strains.

CASE STUDY 2: THE ACDS GENE

Question 1

Does the bacterium contain acdS?

DNA → PCR

Question 2

Is acdS expressed under stress?

RNA → cDNA → RT-qPCR

Question 3

Is ACC deaminase active?

Enzyme activity assay

CHALLENGE: CHOOSE THE METHOD

Which approach would you choose?

1. Does a bacterial isolate contain nifH?

2. Does salt stress increase expression of a target gene?

3. Which bacteria are present in a soil sample?

TAKE-HOME MESSAGES

DNA — information storage
RNA — gene expression
DNA → RNA → Protein
Question → Molecule → Method → Data → Interpretation
Presence ≠ Expression ≠ Function

References / Figure Sources

MERO Foundation. *Molecular Biology* educational illustration.

Pearson Education, Inc. (2009). Nucleotide structure illustration.

BioRender. *Central Dogma* template/illustration.

Introductory infographic “A Molecular Biology Question” — created for this lecture.