

THE RIBOSOME

Structure, Function, Eukaryotic vs Prokaryotic Ribosomes, and Ribosomal DNA (rDNA)

1. Introduction

Ribosomes are the molecular machines that carry out **protein synthesis (translation)** in every living cell. Each ribosome is a large ribonucleoprotein complex built from **ribosomal RNA (rRNA)** and dozens of **ribosomal proteins**, organised into two unequal subunits — a large subunit and a small subunit — that come together on an mRNA molecule to read its genetic code and assemble a corresponding polypeptide chain.

- First observed by George Palade (1955) using the electron microscope as dense cytoplasmic granules; he later shared the 1974 Nobel Prize in Physiology or Medicine for this work.
 - Found in all three domains of life — Bacteria, Archaea, and Eukarya — reflecting their fundamental and ancient role in the cell.
 - Not membrane-bound; ribosomes occur free in the cytosol, attached to the rough endoplasmic reticulum and nuclear envelope (eukaryotes), and inside mitochondria and chloroplasts (organellar ribosomes).
 - Ribosome size/composition is described using the **Svedberg unit (S)**, a measure of sedimentation rate during centrifugation — it reflects size and shape together, so S-values are not simply additive.
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2. Structure of the Ribosome

A functional ribosome is composed of a **large subunit** and a **small subunit**, each itself an assembly of rRNA molecules folded into an intricate three-dimensional scaffold, studded with proteins. The two subunits join only during active translation and separate again once a polypeptide is released.

- **Small subunit** — binds mRNA and is chiefly responsible for decoding: it checks that each incoming tRNA anticodon correctly pairs with the mRNA codon.
- **Large subunit** — contains the **peptidyl transferase centre (PTC)**, the catalytic site that forms peptide bonds between amino acids; the PTC is built entirely of rRNA, making the ribosome a **ribozyme** (a catalytic RNA).
- **Three tRNA-binding sites** span both subunits: the **A (aminoacyl) site** receives the incoming charged tRNA, the **P (peptidyl) site** holds the tRNA carrying the growing polypeptide, and the **E (exit) site** is where the spent, uncharged tRNA leaves.
- An **mRNA-binding groove** runs through the small subunit, and a **polypeptide exit tunnel** runs through the large subunit, through which the new protein emerges as it is made.
- Ribosomes that translate together on a single mRNA strand form a **polysome (polyribosome)**, allowing several protein copies to be made from one transcript simultaneously.

The Ribosome during Translation

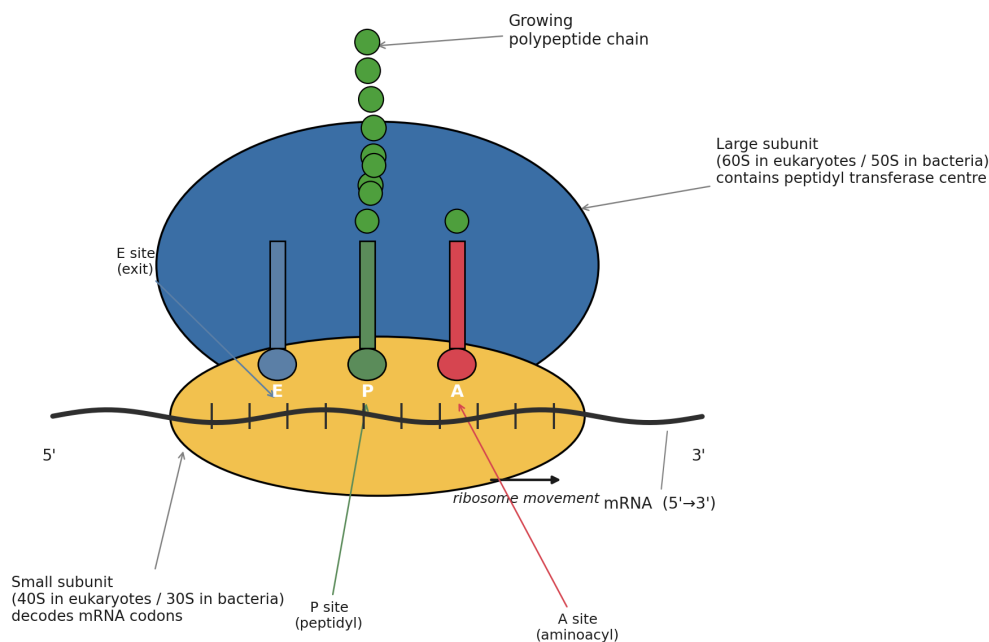


Fig. 1 — General structure of a ribosome bound to mRNA, showing the small and large subunits and the A, P, and E tRNA sites.

3. Function: Protein Synthesis (Translation)

The central function of the ribosome is to translate the nucleotide sequence of messenger RNA (mRNA) into the amino acid sequence of a protein, using transfer RNAs (tRNAs) as adaptor molecules.

Translation proceeds in three stages:

(a) Initiation

- The small subunit binds mRNA near its start codon (AUG) together with an initiator tRNA carrying methionine (or N-formylmethionine, fMet, in bacteria).
- The large subunit then joins to form a complete, translation-competent ribosome, with the initiator tRNA positioned in the P site.

(b) Elongation

- A charged aminoacyl-tRNA enters the A site and its anticodon is checked against the mRNA codon.
- The peptidyl transferase centre catalyses formation of a peptide bond, transferring the growing chain from the P-site tRNA onto the A-site amino acid.
- The ribosome then translocates one codon along the mRNA (an energy-dependent step, using GTP and elongation factors): the tRNAs shift A→P→E, and the spent tRNA leaves from the E site.
- This cycle repeats rapidly — bacterial ribosomes add roughly 15–20 amino acids per second; eukaryotic ribosomes are somewhat slower.

(c) Termination

- When a stop codon (UAA, UAG, or UGA) enters the A site, no matching tRNA exists; instead, a release factor binds and triggers hydrolysis of the bond linking the polypeptide to the final tRNA.

- The completed protein, the tRNA, and the mRNA are released, and the ribosomal subunits dissociate, ready to be recycled for another round of translation.

Beyond synthesising cytosolic proteins, ribosomes docked on the rough endoplasmic reticulum (via the signal recognition particle) translate proteins destined for secretion, the plasma membrane, or membrane-bound organelles, feeding the nascent chain directly into the ER lumen or membrane.

4. Eukaryotic vs. Prokaryotic Ribosomes

Although the core mechanism of translation is conserved, prokaryotic (bacterial) and eukaryotic ribosomes differ in size, rRNA and protein content, and several details of how translation is initiated — differences that are also the basis of selective antibiotic action.

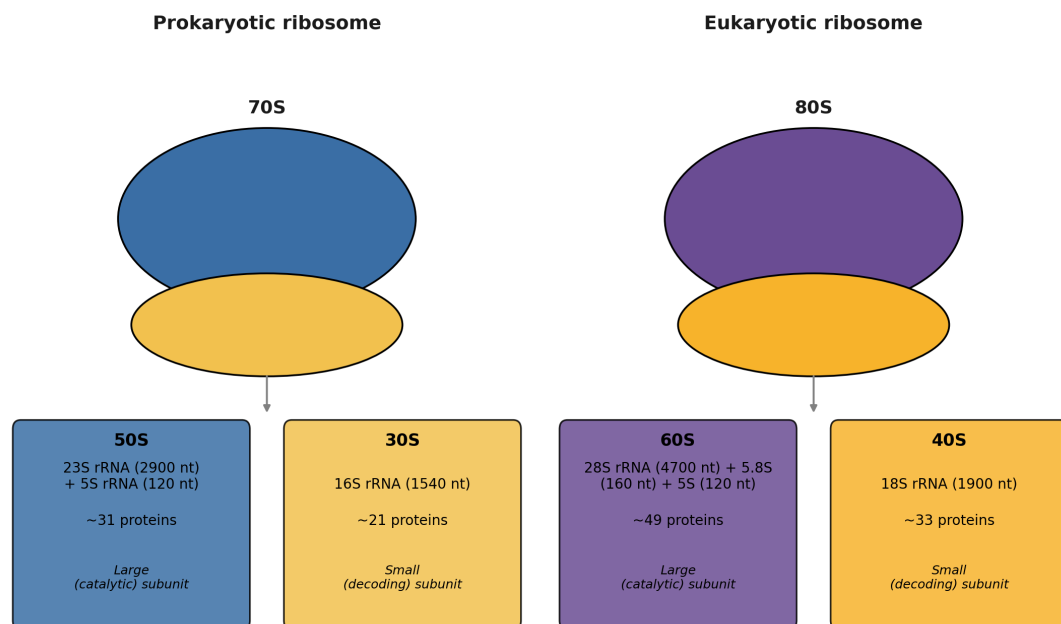


Fig. 2 — Subunit composition of the prokaryotic 70S ribosome and the eukaryotic 80S ribosome.

Feature	Prokaryotic ribosome	Eukaryotic ribosome
Overall size	70S	80S
Large subunit	50S — 23S rRNA + 5S rRNA, ~31 proteins	60S — 28S rRNA + 5.8S rRNA + 5S rRNA, ~49 proteins
Small subunit	30S — 16S rRNA, ~21 proteins	40S — 18S rRNA, ~33 proteins
Location	Cytoplasm (also chloroplasts/mitochondria — see note below)	Cytoplasm; also bound to rough ER and nuclear envelope
rRNA genes	Single <i>rrn</i> operon (16S + 23S + 5S together); few copies per genome	Separate 45S/47S precursor locus (18S+5.8S+28S) in NOR + independent 5S rDNA array
Start codon / initiator tRNA	AUG, read with N-formylmethionine (fMet) tRNA	AUG (usually the first from the 5' cap), read with methionine (Met) tRNA

Feature	Prokaryotic ribosome	Eukaryotic ribosome
mRNA recognition	Shine–Dalgarno sequence pairs with 16S rRNA to position the start codon	5' cap recognised by initiation factors; ribosome scans to the start codon (Kozak context)
mRNA structure	Often polycistronic (one mRNA, several genes); no cap or poly-A needed	Typically monocistronic; capped (5') and polyadenylated (3')
Antibiotic sensitivity	Inhibited by streptomycin, tetracycline, chloramphenicol, erythromycin	Generally insensitive to those antibiotics (basis of selective toxicity)
Ribosome biogenesis site	Cytoplasm (no nucleolus)	Nucleolus (rRNA processing and subunit assembly) then exported to cytoplasm

Note — organellar ribosomes

Mitochondria and chloroplasts contain their own ribosomes, reflecting their endosymbiotic bacterial origin (endosymbiont theory).

Mitochondrial ribosomes (mitoribosomes) and chloroplast ribosomes are more similar in size, rRNA content, and antibiotic sensitivity to prokaryotic 70S ribosomes than to the cytoplasmic 80S ribosomes of their own eukaryotic cell.

5. Ribosomal DNA (rDNA)

Ribosomal DNA (rDNA) refers to the DNA sequences that encode ribosomal RNA. Because a cell needs huge numbers of ribosomes, rRNA genes are present in many copies and are transcribed at a very high rate — unlike most protein-coding genes, which exist as a single copy per haploid genome.

(a) Organisation in eukaryotes

- The genes for 18S, 5.8S, and 28S rRNA lie together in a single repeating transcription unit, arranged in **tandem arrays of hundreds of copies** at one or more chromosomal loci called **Nucleolar Organizer Regions (NORs)** — in humans, on the short arms of the five acrocentric chromosomes (13, 14, 15, 21, 22).
- Each repeat unit contains, in order: a non-transcribed spacer (NTS/IGS) → external transcribed spacer (5' ETS) → 18S gene → internal transcribed spacer 1 (ITS1) → 5.8S gene → internal transcribed spacer 2 (ITS2) → 28S gene.
- RNA polymerase I** transcribes the whole unit as a single long precursor, the **45S (or 47S) pre-rRNA**, inside the **nucleolus** — the site where the NORs cluster and where rRNA processing and ribosome assembly occur.
- The pre-rRNA is then cleaved and trimmed, removing the spacers, to yield the mature 18S, 5.8S, and 28S rRNAs.
- The **5S rRNA gene** is the exception: it lies in a separate tandem array outside the nucleolus and is transcribed independently by **RNA polymerase III**; the mature 5S rRNA is then imported into the nucleolus to join 5.8S and 28S rRNA in the large (60S) subunit.

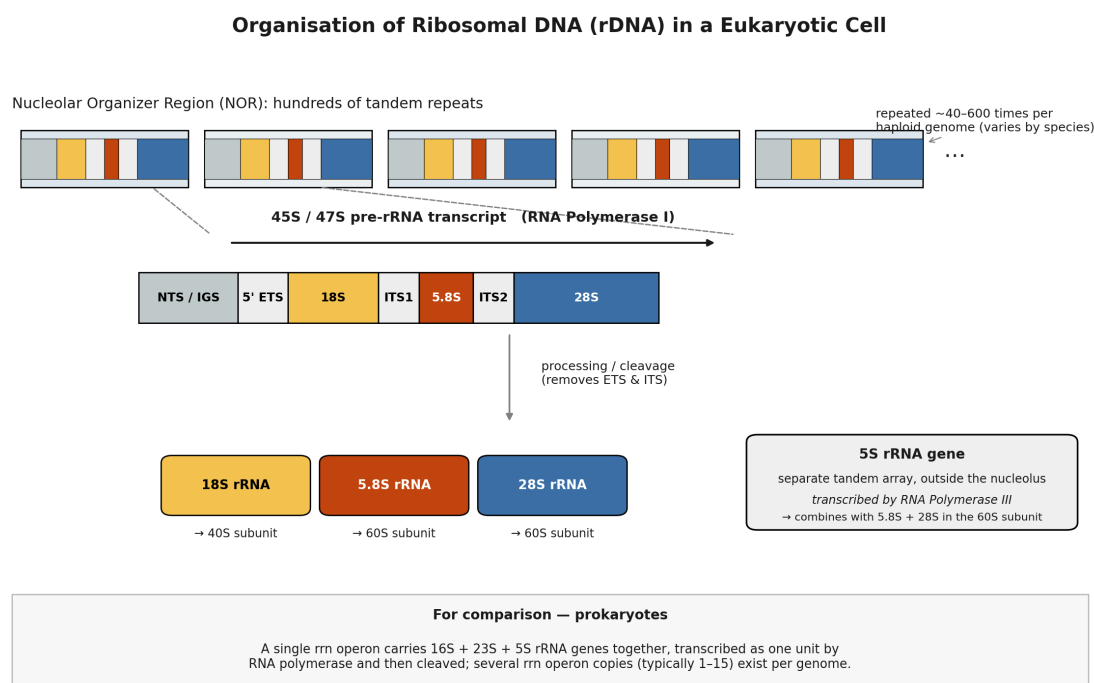


Fig. 3 — Tandem organisation of rDNA repeats at the Nucleolar Organizer Region, the 45S/47S transcription unit, and its processing into mature rRNAs.

(b) Organisation in prokaryotes

- Bacterial rRNA genes (16S, 23S, and 5S) are arranged together as a single **rrn operon** and transcribed as one polycistronic precursor by RNA polymerase, then cleaved into the three mature rRNAs.
- Most bacterial genomes carry several copies of the rrn operon (commonly around 1–15, e.g. seven in *E. coli*) scattered at different genomic locations, rather than one large tandem array — far fewer copies than the hundreds typical of eukaryotic rDNA, consistent with prokaryotes lacking a nucleolus.

(c) Why it matters

- High copy number and dedicated, highly active RNA polymerases allow cells to produce the very large quantities of rRNA needed to build thousands of ribosomes per generation.
- Because rDNA sequences (especially 16S rRNA genes in prokaryotes, and 18S/28S/ITS regions in eukaryotes) are highly conserved yet contain variable regions, they are widely used as molecular markers for identifying species and reconstructing evolutionary relationships (phylogenetics, DNA barcoding).
- The nucleolus itself is essentially defined by rDNA activity — it forms around actively transcribing NORs and disassembles when rRNA transcription is switched off (e.g., during mitosis).

6. Quick Revision Summary

Aspect	Key point
Composition	rRNA + ribosomal proteins, forming a ribonucleoprotein particle
Subunits	One large + one small subunit, joining only during translation
Catalytic site	Peptidyl transferase centre in the large subunit — an RNA-based catalyst (ribozyme)
tRNA sites	A (entry) → P (peptide bond formation) → E (exit)
Prokaryotic ribosome	70S = 50S (23S + 5S rRNA) + 30S (16S rRNA)
Eukaryotic ribosome	80S = 60S (28S + 5.8S + 5S rRNA) + 40S (18S rRNA)
rDNA (eukaryotes)	Tandem 45S/47S unit (18S–5.8S–28S) at the NOR, transcribed by Pol I; 5S rDNA separate, transcribed by Pol III
rDNA (prokaryotes)	Single rrn operon (16S+23S+5S), several copies per genome

Compiled study note — cell biology / molecular biology reference. Figures are original schematic diagrams prepared for this note and are not to scale.