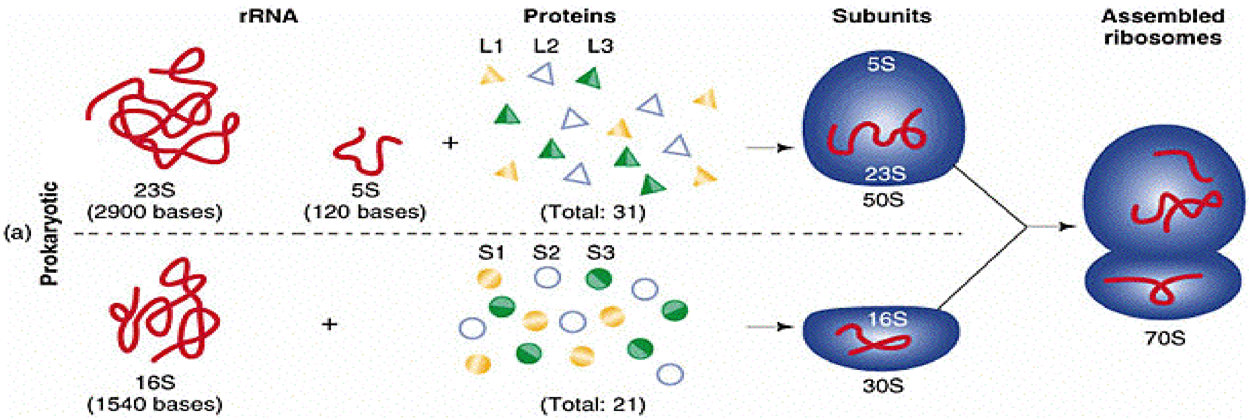
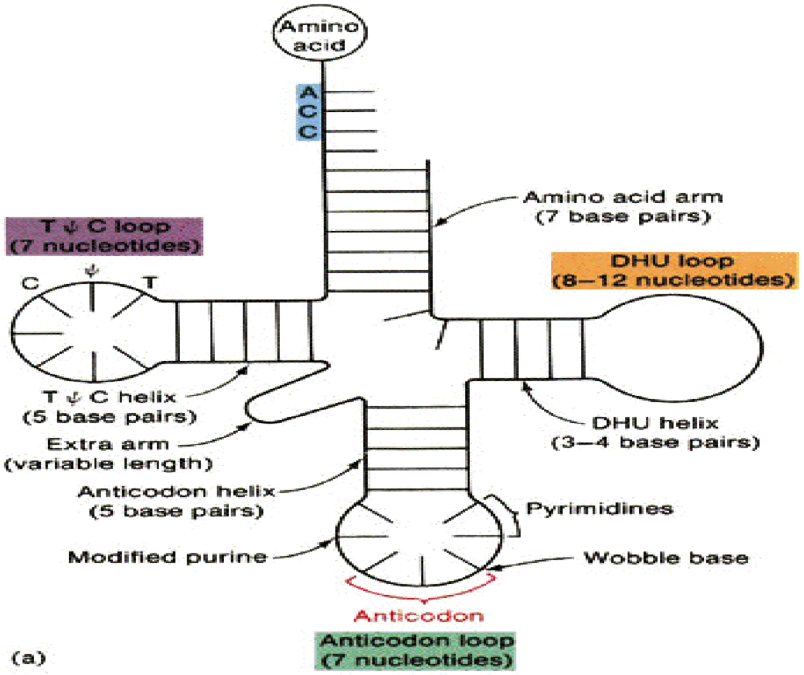
General Structure of Ribosome

PROKARYOTIC



General Structure of tRNA molecules:

- Small; approximately 75 nucleotides
- "cloverleaf" structure contains 3 characteristic loops; anticodon loop is responsible for pairing with mRNA
- 3' end of molecule associates with specific amino acid



THREE ROLES OF RNA IN PROTEIN SYNTHESIS (TRANSLATION):

- **Messenger RNA (mRNA) carries information copied from DNA in the form of a series of three base "words" termed codons.**
- **Transfer RNA (tRNA) deciphers the code and delivers the specified amino acid.**
- **Ribosomal RNA (rRNA) form ribosomes, the protein-synthesizing machines.**

START AND STOP SIGNALS:

- **Start codon (initiate translation)**

AUG (encodes amino acid methionine)

- **Stop codons (terminate translation)**

- **UAA**
- **UGA**
- **UAG**

Groupings of codons:

- Of the 64 codons, 61 specify amino acids and the other 3 are signals to terminate translation
- 9 codon families.
  - *e.g.* encode Thr with:
    - ACU
    - ACC
    - ACA
    - ACG
- 13 codon pairs

- *e.g.* encode Asp with: Glu with
  - GAU GAA
  - GAC GAG

Codons for initiation of translation:

- The major codon for initiation is AUG
- For the 4288 genes identified in *E. coli* :
- AUG is used for 3542 genes.
- GUG is used for 612 genes.
- UUG is used for 130 genes.
- AUU is used for 1 gene.
- CUG may be used for 1 gene.

CODONS FOR TERMINATION OF TRANSLATION

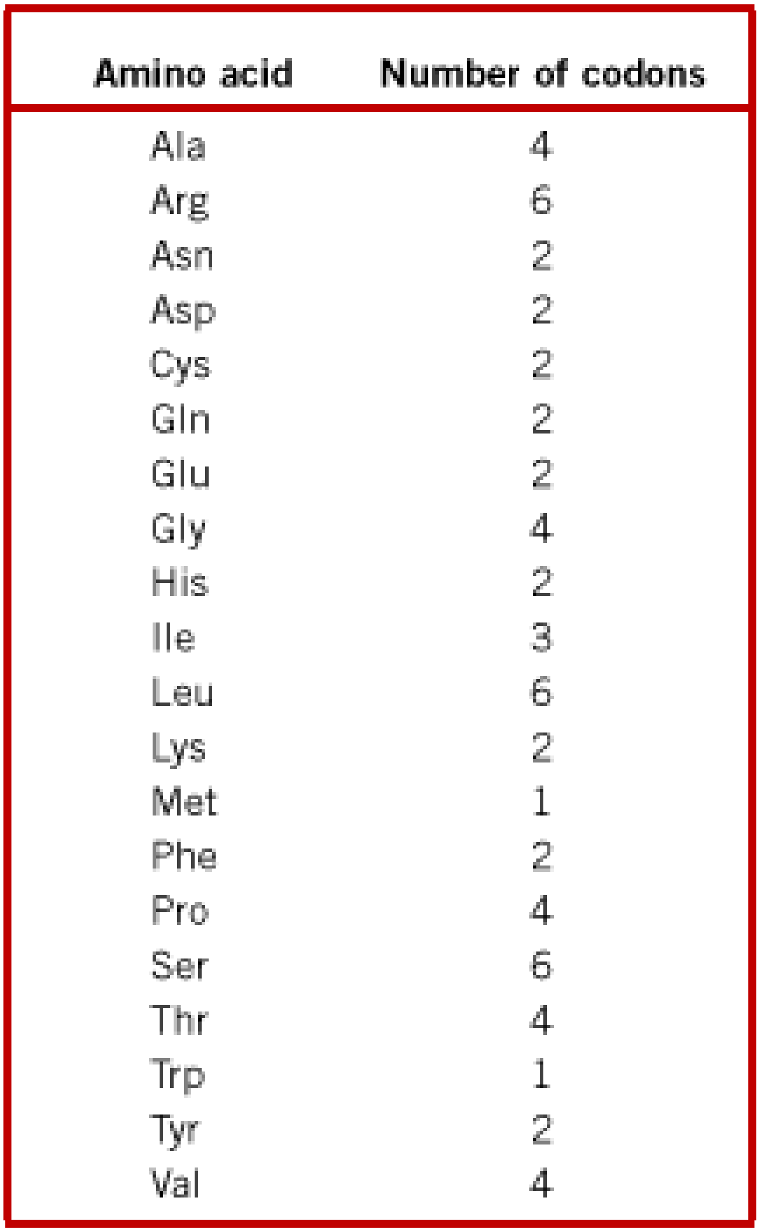
- **UAA, UAG, UGA**
- **For the genes identified in *E. coli:***
- UAA is used for 2705 genes.
- UGA is used for 1257 genes.
- UAG is used for 326 genes.

DEGENERACY OF THE CODE

- The code is “degenerate”: different codons can specify the same amino acid or an amino acid may be specified by more than one codon.
- Degeneracy refers to the fact that almost all amino acids are encoded by multiple codons.

- Degeneracy is found primarily in the 3rd position of the codon, *i.e.* the nucleotide in the 3rd position can change without changing the amino acid specified.

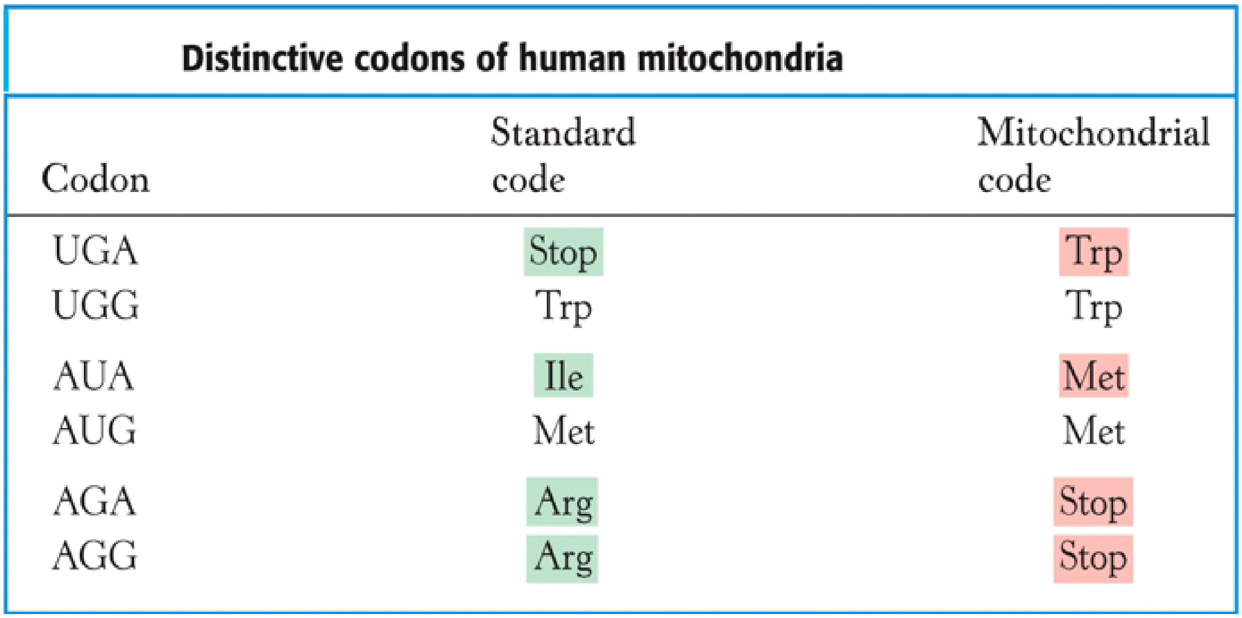
- In some cases, the 1st position is also degenerate.



Amino acid	Number of codons
Ala	4
Arg	6
Asn	2
Asp	2
Cys	2
Gln	2
Glu	2
Gly	4
His	2
Ile	3
Leu	6
Lys	2
Met	1
Phe	2
Pro	4
Ser	6
Thr	4
Trp	1
Tyr	2
Val	4

Genetic code is universal (almost)

- All organisms so far examined use the code as originally deduced (or something very close to it).
- The rare exceptions involve limited differences.
  - *e.g.* in RNA derived from mitochondrial DNA, UGA encodes Trp instead of serving as a stop codon.



Distinctive codons of human mitochondria

Codon	Standard code	Mitochondrial code
UGA	Stop	Trp
UGG	Trp	Trp
AUA	Ile	Met
AUG	Met	Met
AGA	Arg	Stop
AGG	Arg	Stop

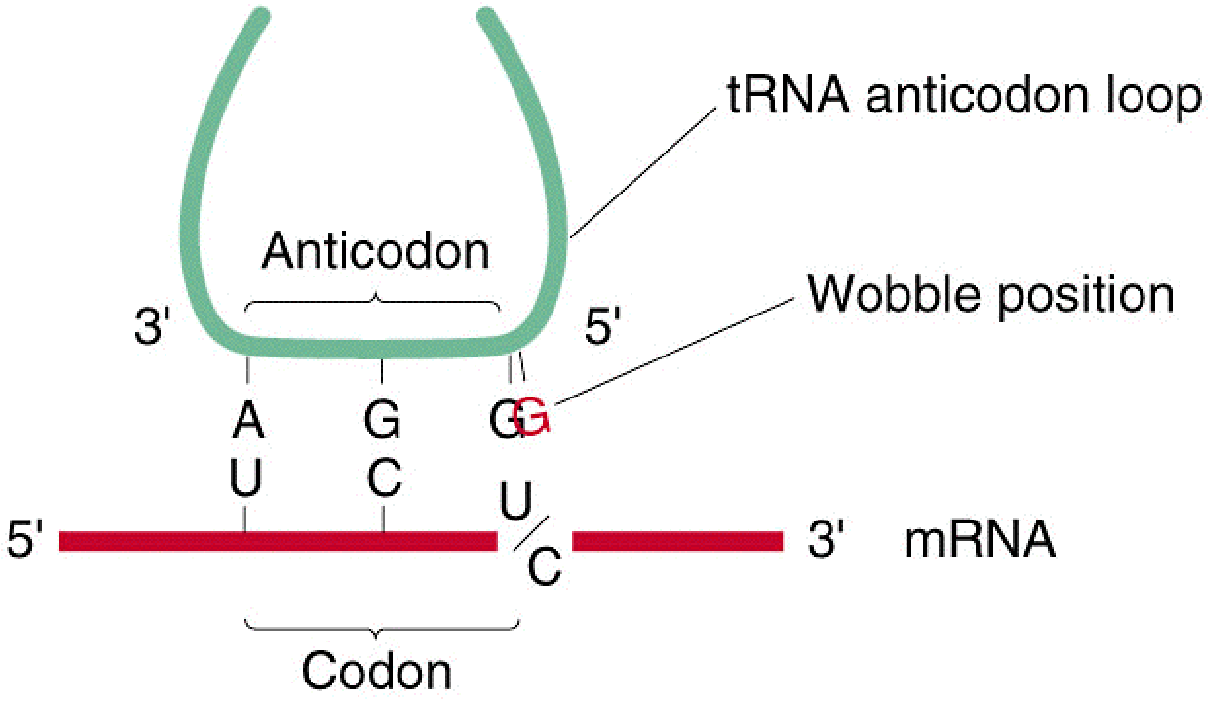
Wobble in anticodon-codon pairing

- Some nucleotides in the 1st position of the anticodon (in tRNA) can pair with >1 nucleotide in the 3rd position of the codon.
- G can pair with U and I can pair with U, C or A.

1st position in anticodon	**3rd position in codon**
C	G
A	U
U	A or G
G	C or U

I (inosinic acid)   U, C or A

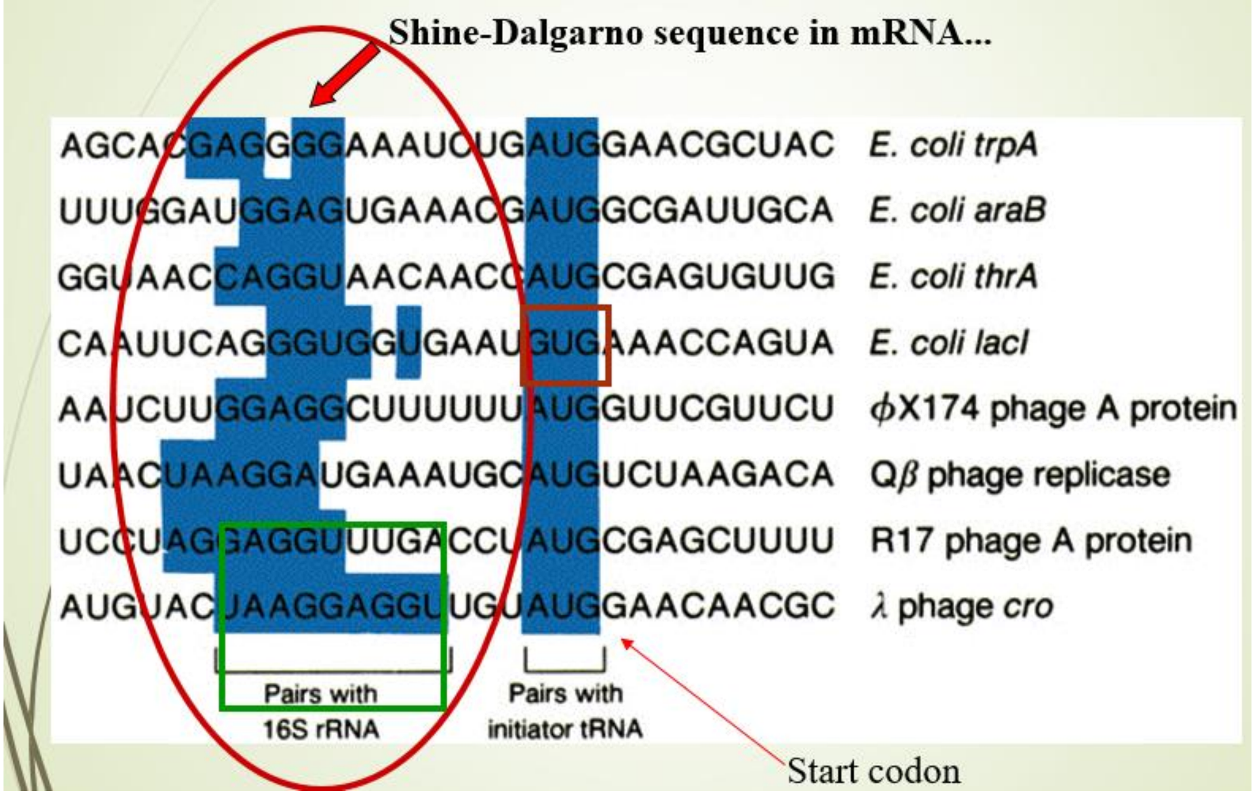
- Result: 61 codons can read by as few as 50 tRNAs.



Shine-Dalgarno Sequences:

- Help to align ribosomes on mRNA to properly start translation.
- Can base-pair with a sequence (ACCUCCUUA) contained in the ribosomal RNA.
- mRNA has also regions that do not encode for a protein
- **Ribosome Binding Site (RBS) = Shine-Dalgarno sequence (SD)**
- The first AUG after SD-sequence is interpreted as the start site of translation.

- **How does translation start? <u>AUG</u> is the Start Codon**



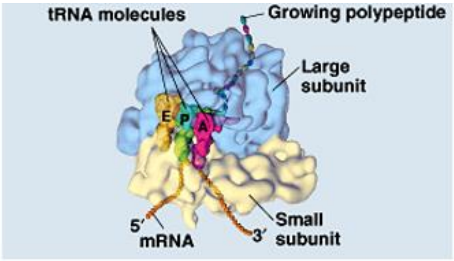
Shine-Dalgarno sequence in mRNA...

AGCACGAGGGGAAAUCUGAUGGAACGCUAC	*E. coli trpA*
UUUGGAUGGAGUGAAACGAUGGCGAUUGCA	*E. coli araB*
GGUAACCAGGUAACAACCAUGCGAGUGUUG	*E. coli thrA*
CAAUUCAGGGUGGUGAAUGUGAAACCAGUA	*E. coli lacI*
AAUCUUGGAGGCUUUUUUAUGGUUCGUUCU	φX174 phage A protein
UAACUAAGGAUGAAAUGCAUGUCUAAGACA	Qβ phage replicase
UCCUAGGAGGUUUGACCUAUGCGAGCUUUU	R17 phage A protein
AUGUACUAAGGAGGUUGUAUGGAACAACGC	λ phage *cro*

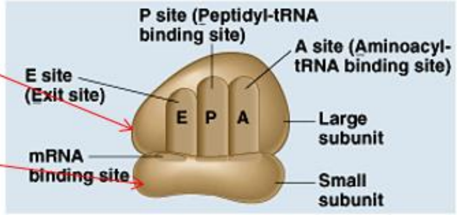
The anatomy of a functioning ribosome:

Ribosomes – complex of proteins and ribosomal RNA (rRNA)

- Large subunit
    - Binding sites for tRNAs
        - E (exit site)
        - P (peptidyl-tRNA site)
        - A (aminoacyl-tRNA site)
- Small subunit
    - Binding site for mRNA through a special binding site that recognizes a specific sequence.

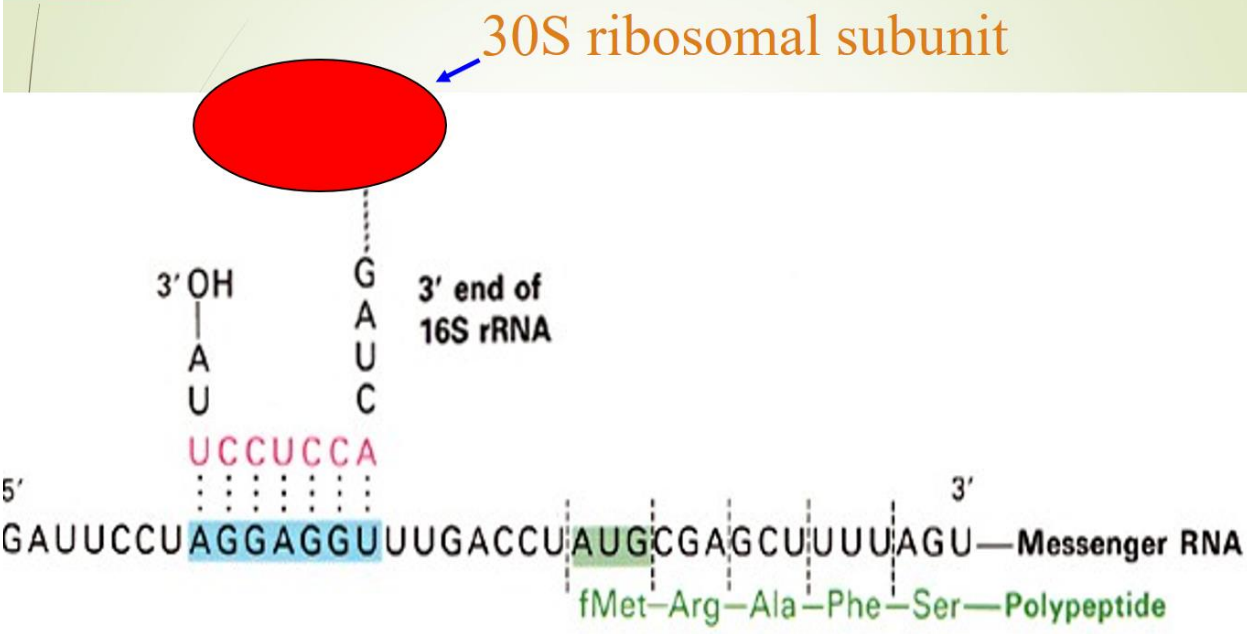


(a) Computer model of functioning ribosome



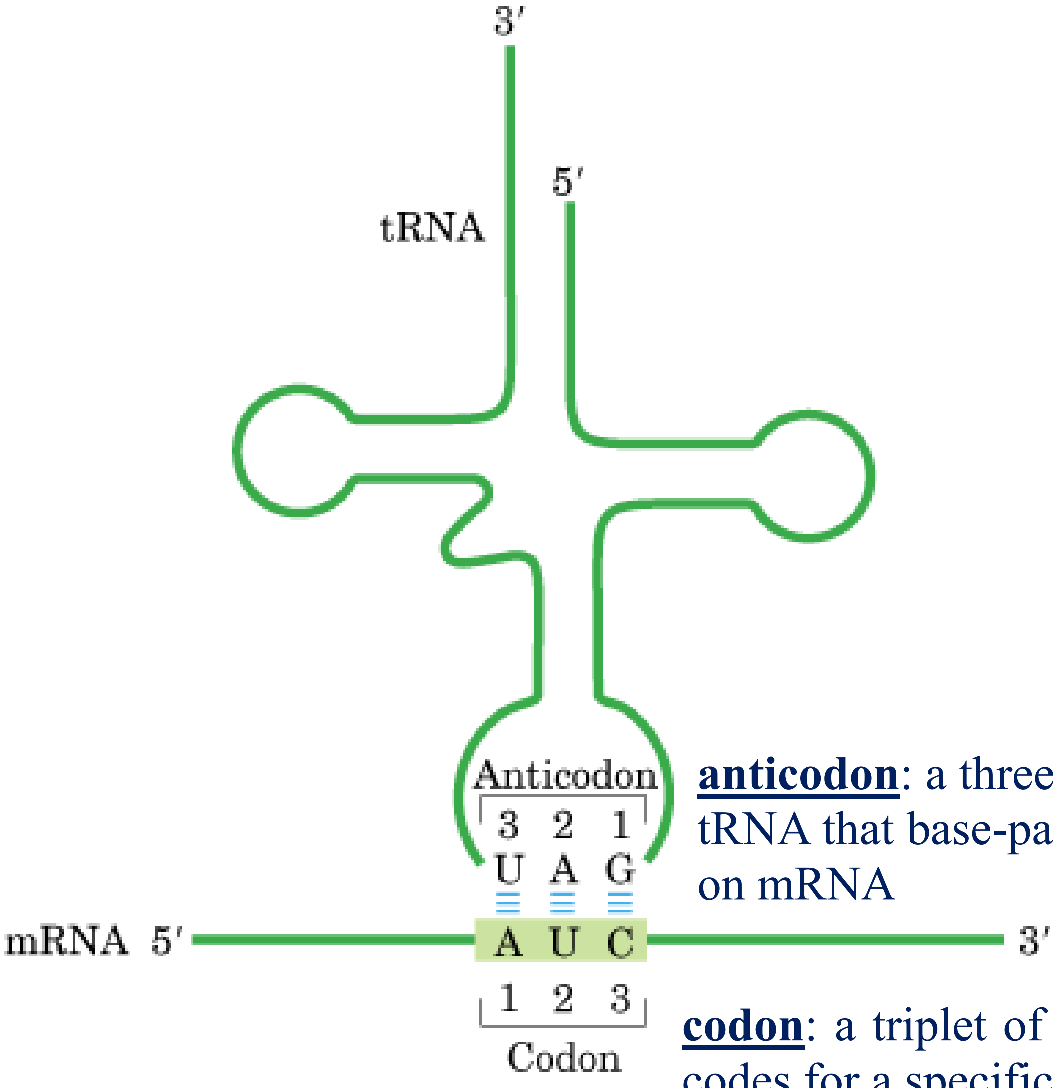
(b) Schematic model showing binding sites

How is the Shine-Dalgarno Sequence Recognized?



Pairing relationships of codon and anticodon

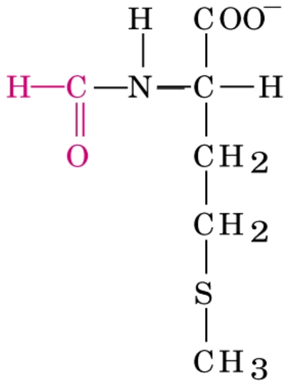
Cells must have at least one kind of tRNA for each amino acid. At least 32 tRNAs are required to recognize all the amino acid codons (some recognize more than one codon).



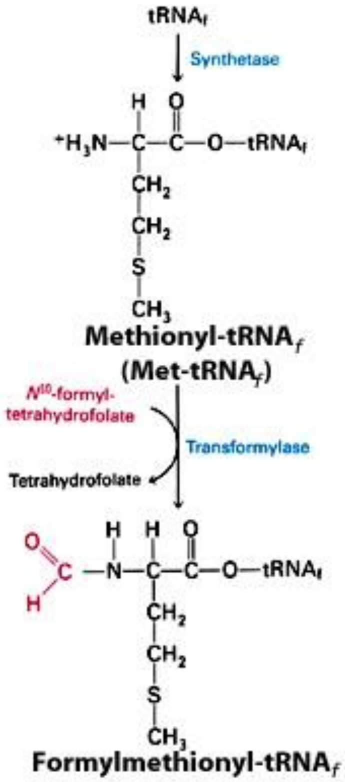
anticodon: a three base sequence on tRNA that base-pairs with the codon on mRNA

codon: a triplet of nucleotides that codes for a specific amino acid

- In bacteria, the first amino acid used to initiate protein synthesis is N-formylmethionine.
- There are two separate types of tRNAs in bacteria that are specific for methionine: $tRNA^{Met}$ and $tRNA^{fMet}$
- Both tRNAs are charged with methionine. However the methionine on $tRNA^{fMet}$ is subsequently formylated by a transformylase.
- There is only one methionine aminoacyl tRNA synthetase.



N-Formylmethionine



- **TRANSLATION CAN BE DIVIDED INTO THREE STAGES:**

- **INITIATION**
- **ELONGATION**
- **TERMINATION**

LECTURE 4 QUESTIONS SHEET

1. A codon contains how many nucleotides?
a) 1
b) 2
c) 3
d) 4

The initiation codon is ____________
a) AUG
b) UAA
c) UAG
d) UGA

. The termination codon is not ____________
a) AUG
b) UAA
c) UAG
d) UGA

9. The genetic code translated the language of ____________
a) Proteins into that of RNA
b) Amino acids into that of RNA
c) RNA into that of proteins
d) RNA into that of DNA

1. Which of the following is not a feature of the genetic code?
a) Triplet
b) Degenerate
c) Non – overlapping
d) Ambiguous

The codon is a _____________

a) Singlet
b) Duplet
c) Triplet
d) Quadruplet

Which of the statements below is false?

A) The genetic code is overlapping.
B) The genetic code is universal.
C) Degenerate codons specify the same amino acids.
D) The genetic code is triplet.

The first mRNA codon to specify an amino acid is always

A) TAC.
B) UAA.
C) UAG.
D) AUG.

Transfer RNA's bind during translation by the

A) codon.
B) anticodon.
C) template

Of the ____ different possible codons, ____ specify amino acids and ____ signal stop.

A) 20, 17, 3
B) 180, 20, 60
C) 64, 61, 3
D) 61, 60, 1

WITH OUR BEST WISHES