

Highlights Translation 2

1. Protein synthesis uses GTP as an energy source (not ATP).
2. Translation initiation requires 1) an mRNA; 2) a start codon in the mRNA - AUG; 3) the 30S ribosomal subunit; 4) the 50S ribosomal subunit; 5) proteins called initiation factors; and 6) GTP.
3. During initiation of translation, the first tRNA (linked to fmet) forms base pairing between its anti-codon and the codon of the mRNA. A sequence known as the Shine-Dalgarno sequence in the mRNA (in prokaryotes only) helps the ribosome to properly align the mRNA in the ribosome. The Shine-Dalgarno sequence forms a base paired region with the 16S rRNA of the 30S subunit of the ribosome.
4. Elongation of translation occurs by binding of an incoming tRNA (which will base pair with the next codon) in a region of the ribosome known as the 'A' site. The previous tRNA is found on the adjacent 'P site' of the ribosome. A third site of the ribosome where tRNAs are found is the 'E site', which is where tRNAs exit the ribosome after they have been used.
5. Translation involves linking amino acids properly together via peptide bonds. Note that the growing polypeptide chain is held on a tRNA. After the peptide bond forms, the tRNA with the polypeptide chain moves to the P site and a new tRNA with an amino acid comes into the A site. This process also involves the mRNA sliding one codon down the chain to put a new codon in the A site.
6. After the tRNA properly binds in the A site, a peptide bond is formed between the peptide on the tRNA in the P site and the amino acid on the tRNA in the A site. The catalyst for the peptide bond is the 23S rRNAs in the ribosome. Thus, peptide bonds are formed by ribozymes (catalytic RNAs). The result is a peptide linked to the tRNA in the A site.
7. The next step in elongation occurs when the tRNA with the polypeptide in the A site moves over to the P site (displacing the tRNA previously there to the E site, from which it exits the ribosome). The A site is thus left open. The ribosome slides three nucleotides along the mRNA, bringing a new complementary sequence into the A site. Movement of the ribosome along the mRNA is facilitated by EF-G and requires GTP.
8. Two other EF proteins are required for elongation. EF-Tu is the most abundant E. coli protein and it functions to carry the tRNA to the A site of the ribosome. EF-Tu makes sure that the properly base-paired tRNA is in the A site. EF-Tu also protects the bond between the amino acid and the tRNA from water.
9. Proper termination of translation is the last step of the process of making a protein. The process occurs when a stop codon appears in the 'A' site of the ribosome. Since there are no tRNAs with a complementary anti-codon to the stop codons, release factors bind to the stop codon. They cause the bond between the polypeptide and the tRNA in the P site to be broken because they carry water to it and this breaks the bond. Thus, the final polypeptide is released from the ribosome and the entire complex then comes apart.
10. Translation is a target for action of antibiotics. This works because the phenomenon of translation in prokaryotes is different enough from that of eukaryotes that specific inhibitors of prokaryotic translation can be found that have no effect on eukaryotic translation. As a result, prokaryotes can be killed without any effect on eukaryotes taking said antibiotics. One translational inhibitor of prokaryotic translation is the antibiotic puromycin, which acts to prematurely terminate prokaryotic translation at the elongation phase. This happens because puromycin looks like a tRNA and fits into the A (or P) site of the ribosome. In the A

site, it can be attached to the growing polypeptid chain, but it is soon released, causing premature termination. In addition, puromycin can bind to the P site of ribosomes and prevents anything from binding there.

11. Eukaryotic mRNAs have a 5' cap and a 3' polyA tail. Eukaryotic translation occurs much like prokaryotic translation, but the factors are differently named. One important difference of eukaryotic translation is that the initiation complex in eukaryotes involves both the 5' cap and the 3' polyA sequence in a looped structure. The 5' cap and 3' poly-A sequence both play roles in helping to increase the stability of eukaryotic mRNAs.

12. Post-translational (= after translation has occurred) processing of proteins gives them their final structure and properties. One such modification was discussed for insulin. This involves proteolytic cleavage at two places in the insulin precursor, giving rise to a final structure that has two chains held together by disulfide bonds.

13. Proteins do not have long lifetimes in the cell. They are broken down in cells in cellular structures called proteasomes. Proteins targeted for degradation are tagged with the peptide known as ubiquitin.

14. Selenocysteine is a rare amino acid occasionally incorporated into proteins. It is sometimes called the 21st amino acid. It is the only modified amino acid in proteins that gets there by direct incorporation rather than post-translational modification. It is because of this that trace amounts of selenium are needed in the diet.