EF-Tu: A Motor Protein

How do the structural features of the prokaryotic elongation factor EF-Tu facilitate its role as a motor protein?

EF-Tu (Figure 19A), the protein factor that positions aminoacyl-tRNA complexes in the A site of prokaryotic ribosomes, is a well-researched example of a GTP-binding motor protein. Recall that *motor proteins* (Section 2.1) use nucleotide hydrolysis to drive changes in their own conformations that promote ordered conformational changes in adjacent molecules or subunits. In other words, motor proteins, often called NTPases, function as mechanochemical transducers. These NTP hydrolysis–driven conformational changes, which principally occur in localized structural units called switches, alter the affinity of the NTPase for other molecules.

EF-Tu possesses three domains. Domain 1 contains a GTP binding site and two switch regions. Domain 2 is connected to domain 1 through a pliable peptide segment. In its active GTP-bound form (EF-Tu-GTP), the elongation factor possesses a binding site for an aminoacyl-tRNA. After aminoacyl-tRNA binding, the entire structure is referred to as the *ternary complex*. All three domains of EF-Tu are involved in tRNA binding. For example, the TψC stem of tRNA molecules (Figure 17.19) interacts with several amino acid residues in domain 3. The binding of aminoacyl-tRNA projects the anticodon away from the ternary complex so it is free to interact with mRNA codons.

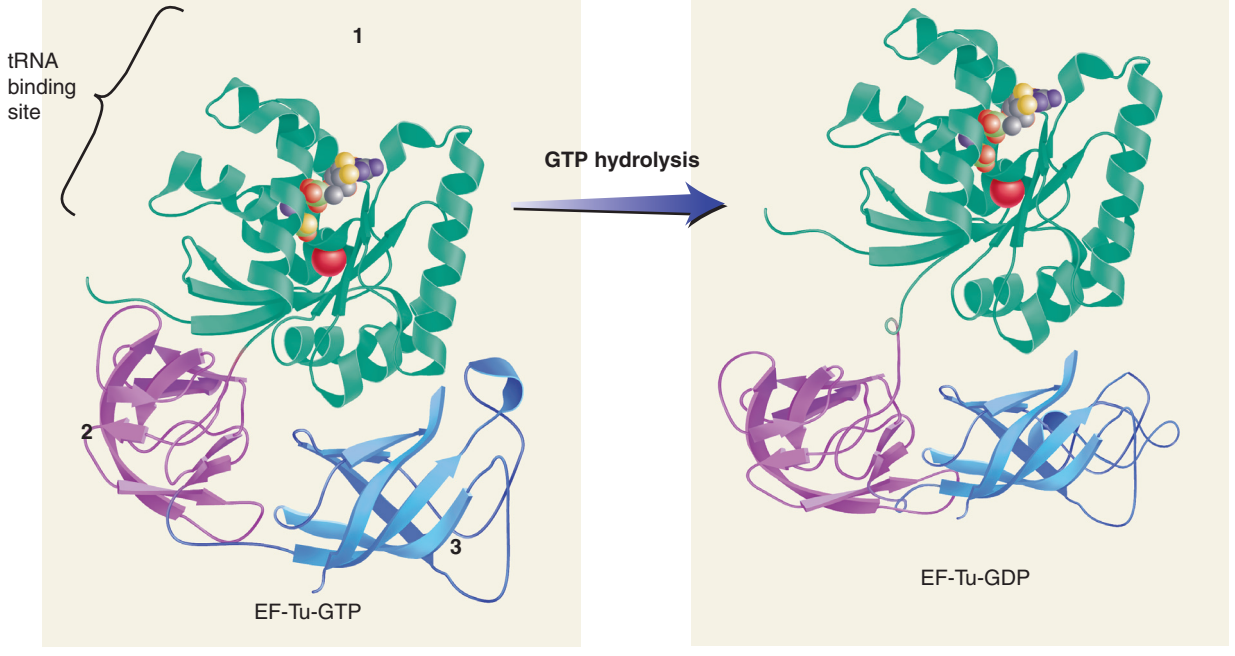

FIGURE 19A

EF-Tu

EF-Tu is a GTPase that positions aminoacyl-tRNA complexes within the A site of prokaryotic ribosomes during protein synthesis. The binding of a GTP molecule by EF-Tu causes domain conformation changes (not shown) that result in the creation of a binding cleft for an aminoacyl-tRNA complex. GTP hydrolysis causes domains 1 and 2 to move apart so that the aminoacyl-tRNA (not shown) is released. (The red sphere represents a magnesium ion.)

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During protein synthesis, the interaction of EF-Tu-GDP (the inactive form) with EF-Ts releases GDP. The subsequent binding of GTP in the domain 1 nucleotide-binding site changes the conformation in the two switch regions. These changes bring domains 1 and 2 close together, forming a binding cleft. Once an aminoacyl-tRNA has been bound in the cleft, the ternary complex enters the ribosome where the aminoacyl-tRNA anticodon binds reversibly to an mRNA codon in the A site. When a ternary complex contains a cognate aminoacyl-tRNA, a conformation change in the ribosome triggers a conformation change in the EF-Tu nucleotide-binding site. The subsequent hydrolysis of GTP causes domains 1 and 2 to move apart, thus allowing the release of the aminoacyl-tRNA.

Switch region 1, which is believed to contain a β-hairpin in the inactive EF-Tu-GDP, is converted to a helix (the "switch helix") in EF-Tu-GTP. In switch region 2 (near the GTP-binding site) the γ-phosphate of GTP causes a conserved glycine residue to flip 180°, moving it 4.6 Å. The dramatic movement of this residue forces the switch helix to migrate four residues along the polypeptide chain, a process that changes the axis of rotation of the helix by 45° and results in a 46 Å movement of the most distal portion of domain 1. This feature of EF-Tu function has been described as a timing mechanism. The timer is activated when the ternary complex binds to the ribosome. The relatively slow rate of GTP hydrolysis provides sufficient time for the dissociation of incorrect codon-anticodon pairings. In contrast, the binding of a cognate aminoacyl-tRNA is so tight that there is sufficient time for GTP to undergo hydrolysis. Thus, the functioning of the ternary complex provides another mechanism for proofreading during translation in addition to that described for the aminoacyl-tRNA synthetases.

SUMMARY: EF-Tu is an NTPase that binds and hydrolyzes GTP. The binding of GTP to domain 1 of EF-Tu causes a change in conformation of the entire protein that facilitates the binding of an aa-tRNA. Once the aa-tRNA complex is positioned in the ribosome via codon-anticodon base pairing, GTP hydrolysis causes a conformational change that results in the release of the aa-tRNA.