The following is a logical categorization of the enzymes and proteins mentioned in the lectures, organized by their biological processes: DNA replication, topology, repair, recombination, transcription, and biotechnology applications.

1. DNA Replication Enzymes and Proteins

These proteins are responsible for duplicating the genetic material within the cell.

- **DnaA:** The initiator protein in *E. coli*. It binds to specific repeats (9-mers) at the origin of replication (oriC), forms a helical protein complex that twists the DNA, and aids in unwinding the 13-mer A-T rich region to allow replication to begin,. It functions as a licensing factor that is active when bound to ATP.
- **DnaB (Helicase):** An enzyme (hexamer) that moves along the lagging strand template in the 5' to 3' direction, separating the DNA double helix using energy from ATP hydrolysis,.
- **DnaC (Helicase Loader):** A protein that helps load the DnaB helicase onto the DNA at the replication origin; it inhibits the helicase until it is released via ATP hydrolysis,.
- **SSB (Single-Strand Binding protein):** A tetramer that binds cooperatively to single-stranded DNA exposed by the helicase to stabilize it, prevent re-annealing, and protect it from degradation.
- **DnaG (Primase):** An enzyme that synthesizes short RNA primers (11-12 bases) on the DNA template, providing the necessary free 3'-OH end for DNA polymerase to initiate synthesis,.
- **DNA Polymerase III (Pol III):** The primary replicase in *E. coli*, responsible for synthesizing the leading and lagging strands. It is a large holoenzyme with high processivity (can add tens of thousands of bases without falling off) and a fast polymerization rate,,. It contains multiple subunits:
    - **Core enzyme ($\alpha, \varepsilon, \theta$):** The catalytic unit. The α subunit performs polymerization, and the ε subunit performs 3'-5' exonuclease proofreading.
    - **Sliding Clamp (β dimer):** A ring-shaped protein that surrounds the DNA and tethers the core enzyme to the template, conferring high processivity,.
    - **Clamp Loader (γ complex):** A multi-subunit complex that uses ATP to load the sliding clamp onto the DNA.
    - **Tau (τ):** Dimerizes the two core enzymes.
- **DNA Polymerase I (Pol I):** An enzyme used primarily for replacing RNA primers with DNA during lagging strand synthesis and for DNA repair. It has three activities: 5'-3' polymerization, 3'-5' proofreading, and a unique 5'-3' exonuclease activity (nick translation) used to remove RNA primers or damaged DNA,,.
- **DNA Ligase:** An enzyme that seals "nicks" (breaks in the sugar-phosphate backbone) by creating a phosphodiester bond between a free 3'-OH and a 5'-phosphate. It connects Okazaki fragments and seals repair patches,.

- **SeqA:** A negative regulator of replication initiation. It binds to hemimethylated GATC sequences at the origin of replication, sequestering the origin to the cell membrane and preventing DnaA from binding, thus delaying re-initiation,.
- **HU Proteins:** Proteins that bind to and stabilize single-stranded DNA during the pre-priming stage of replication.

Viral/Alternative Replication Proteins:

- **Terminal Protein:** Used by Adenovirus; it binds covalently to a Cytosine nucleotide and serves as a primer for DNA polymerase at the very end of linear DNA,.
- **Protein A:** A cis-acting protein in phage ϕX174 rolling circle replication. It nicks the origin, binds to the 5' end, and later ligates the displaced strand to form a circle,.

2. DNA Topology Enzymes

These enzymes manage the supercoiling and topological state of DNA.

- **Topoisomerase I:** Relaxes negative supercoils by nicking one strand of the DNA, passing the intact strand through the break, and resealing it. This process does not require ATP,.
- **Topoisomerase II (DNA Gyrase):** Introduces negative supercoils into DNA using ATP (2 molecules per cycle). It relieves positive supercoiling ahead of the replication fork and aids in condensing the bacterial genome,. It consists of GyrA (DNA cutting) and GyrB (ATP hydrolysis) subunits.
- **Topoisomerase III:** A type I topoisomerase in *E. coli*.
- **Topoisomerase IV:** A type II topoisomerase responsible for decatenating (separating) the two circular daughter chromosomes after replication.

3. DNA Repair and Modification Enzymes

Enzymes involved in maintaining genome integrity and epigenetic marking.

- **Dam Methylase:** Adds a methyl group to the Adenine in GATC sequences. This methylation helps the cell distinguish between the "old" (methylated) template strand and the "new" (unmethylated) strand for mismatch repair and replication control,.
- **Photolyase:** Directly repairs UV-induced pyrimidine dimers by breaking the covalent bonds between bases (found in *E. coli*).
- **MGMT (Methylguanine-DNA Methyltransferase):** Directly repairs methylated guanine by transferring the methyl group to a cysteine residue on the enzyme itself (suicide enzyme).
- **Glycosylases:** Enzymes involved in base excision repair (e.g., Uracil DNA Glycosylase) that recognize specific damaged or incorrect bases and "flip" them out of the helix to cleave them,.
- **UvrABC Endonuclease:** A complex involved in Nucleotide Excision Repair (NER). UvrA and UvrB scan for distortions; UvrC cuts the DNA on both sides of the damage,.

- **UvrD:** A helicase that unwinds and releases the damaged DNA segment cut by UvrC.
- **Mfd:** A protein that displaces a stalled RNA polymerase at a damage site and recruits the Uvr repair system.
- **MutS, MutL, MutH:** Proteins involved in mismatch repair. MutS binds the mismatch; MutL links MutS to MutH; MutH is an endonuclease that cleaves the unmethylated (new) strand at a GATC site,.

4. Recombination Enzymes

Proteins that facilitate the exchange of DNA strands between molecules.

- **RecBCD:** A complex with nuclease and helicase activities. It prepares double-strand breaks for recombination by degrading DNA until it reaches a "Chi" sequence, where it generates a 3' single-stranded tail,.
- **RecA:** A protein that coats single-stranded DNA and catalyzes strand invasion (assimilation) into a homologous double-stranded DNA molecule,.
- **RuvA and RuvB:** Proteins that process Holliday junctions. RuvA forms a tetramer that binds the junction, while RuvB is a hexameric helicase that drives branch migration,.
- **RuvC (Resolvase):** An endonuclease that resolves Holliday junctions by cleaving the DNA strands.
- **RecG:** An enzyme that can also drive branch migration.
- **HO Endonuclease:** Specifically cuts yeast DNA at the *MAT* locus to initiate mating-type switching.
- **Integrase (Int):** A site-specific recombinase (from Phage Lambda) that functions like a Topoisomerase I to integrate viral DNA into the host genome,.
- **IHF (Integration Host Factor):** A host protein required for the integration and excision of Phage Lambda; it bends the DNA,.
- **Xis (Excisionase):** A phage protein required (along with Int and IHF) to excise the phage genome from the host chromosome,.

5. Transcription Enzymes and Factors

Proteins involved in synthesizing RNA from a DNA template.

- **RNA Polymerase (Bacterial):**
    - **Core Enzyme ($\alpha_2 \beta \beta' \omega$):** Synthesizes RNA but lacks promoter specificity. The β and β' subunits form the active site and channel. The α subunits are involved in assembly and interaction with regulatory proteins.
    - **Holoenzyme:** The core enzyme plus a Sigma factor.
- **Sigma Factors (σ):** Proteins that guide RNA polymerase to specific promoters.
    - **σ^{70}:** The housekeeping sigma factor for general gene expression.
    - **σ^{32}:** Activated during heat shock to transcribe chaperones.
    - **σ^{E}:** Activated by extracytoplasmic stress (misfolded proteins in the periplasm).

    - **σ^{54}:** Activated during nitrogen starvation.
    - **σ^{28} / σ^{F}:** Involved in flagellar synthesis.
    - **gp28:** A viral sigma factor (Phage SPO1) that displaces the host sigma factor to transcribe "middle" viral genes.
- **T7 RNA Polymerase:** A single-subunit viral polymerase that is very fast and specific to its own promoters.
- **FtsH:** A protease that degrades σ^{32} under normal conditions.
- **RseA:** An anti-sigma factor that binds σ^{E} to the membrane, preventing its activity until stress occurs.
- **DegS and RseP:** Proteases that cleave RseA in response to stress, releasing σ^{E}.
- **Chaperones:** Proteins (transcribed by σ^{32}) that stabilize other proteins during heat stress.
- **EBP (Enhancer Binding Protein):** A protein that helps activate σ^{54} via phosphorylation and ATP hydrolysis.

6. Biotechnology and Laboratory Tools

Enzymes and proteins specifically noted for their utility in research methods.

- **Restriction Enzymes (Endonucleases):** Bacterial enzymes that cut DNA at specific palindromic sequences (e.g., **EcoRI**, **HindII**, **EcoRV**, **PstI**, **BamHI**, **NotI**, **HaeIII**). They act as a defense against viruses and are used for cloning,,.
- **Klenow Fragment:** The large fragment of DNA Pol I (created by protease digestion) that retains polymerization and proofreading activity but lacks the 5'-3' exonuclease activity. Used for labeling DNA probes,.
- **Taq Polymerase:** A thermostable DNA polymerase from *Thermus aquaticus* used in PCR because it withstands high temperatures.
- **Pfu Polymerase:** A thermostable polymerase with higher fidelity (proofreading activity) than Taq, but slower.
- **Alkaline Phosphatase:** An enzyme used to remove phosphate groups from the 5' ends of DNA (e.g., plasmids) to prevent self-ligation during cloning.
- **Beta-lactamase (implied):** Enzymes encoded by plasmids that degrade antibiotics (e.g., ampicillin), conferring resistance.
- **Endolysin:** An enzyme used by phages to cleave the bacterial cell wall and release progeny virions.