# BIOL 240 *GENERAL MICROBIOLOGY* (Part III) – Important Terms & Concepts 4/14/2019

8. **MICROBIAL GENETICS:** Genetics, Gene, Genome, Genotype, Phenotype. E. coli as Model Organism. ***Central Dogma***: **DNA (replicates) → (transcription) RNA → (translation) Proteins**; DNA: A=T, G≡C. **Semi-Conservative & Bidirectional Replication:** Antiparallel Strands**, Origin of Replication, 5'→3' synthesis**, **DNA Polymerase III**, **RNA Primase**, RNA Primers, template strands, **DNA Polymerase I, DNA Ligase.** Continuous leading strand, discontinuous Lagging Strand Synthesis. **Transcription: RNA Polymerase; mRNA, tRNA**, **rRNA**, 5'→3' from **PROMOTER** on DNA template, unidirectional synthesis! **Codons/Genetic Code – stop (UAA, UAG, UGA) and start (AUG) codons**. **Translation:** mRNA + tRNA with Amino Acid attached (and ***AntiCodon***) + Small Ribosomal subunit + Large Ribosomal subunit. **Ribosome** moves 5'→3' along mRNA template, transfers growing strand in **P-Site** to new Amino acid on tRNA in **A-site** (Peptidyl Transferase); Peptide synthesized **Amino (N) Terminus → Carboxyl (C) Terminus** according to mRNA codons starting with AUG/methionine. ***Constitutive Enzymes, Repressible Enzymes, Inducible Enzymes*.**

- ***OPERONS***: = ***promoter + operator + structural genes, regulated by a Repressor protein*. Lac Operon. (catabolic)** = ON if Lactose present (repressor OFF when BOUND by lact. signal) AND glucose absent; **Trp Operon (anabolic/ biosynthetic)** = ON if Trp end-product NOT in excess & bound to Trp Repressor (repressor OFF when UNBOUND by Trp signal).
- **Horizontal Gene Transfer: Transformation** (Griffiths: R→S *pneumococcus*), recombination/ crossover into chromosome ("homologous gene replacement"), **Conjugation** (**F factor**/ ***Plasmid***, sex pilus, ***Hfr*** Cells; ***F+*** donor cells, F- recipient cells). Bacteriophage **Transduction**. R-Factors (MDR), Transposons. Recombination Required: linear DNA vs. circular DNA with *Ori*.
- **Mutations**: chemical and radiation ***mutagens***. ***DNA Excision Repair*** (Endonuclease, DNA Polymerase I, Ligase). **Missense, Nonsense, Frameshift mutations.** (& "Silent" mutations). Positive/Direct and Negative/Indirect Selection, Replica Plating.
- **AMES TEST** for Chemical Mutagens/Carcinogens.

9. **BIOTECHNOLOGY & RECOMBINANT DNA:** **Biotechnology, Recombinant DNA Technology, Genetic Engineering**; Select microbe, Mutate microbe, Site-Directed Mutagenesis; **Restriction Enzymes**; **Vectors** (*Ori*, antibiotic-resistance (selectable marker), disruptable gene -- with multiple-cloning site), **Polymerase Chain Reaction (PCR)** – Taq DNA Polymerase, specific DNA primers, ***lacZ***-insertions for cloning in plasmids. Denature DNA, **Hybridize, Probe. *Agrobacterium*** – Genetic Engineering in plants; Ti Plasmid → **T-DNA**.

11. **PROKARYOTES:** Domains = Bacteria; Archaea. **rRNA gene (rDNA) analysis – PCR, Hybridization**. **Proteobacteria (Gram -)** = *Agrobacterium*, *Rhizobium*, Enterobacteria (*E. coli, Salmonella, Shigella, Serratia, Klebsiella; Helicobacter*), *Pseudomonas*;
    - **Non-proteobacteria Gram neg's** = Cyanobacteria, Purple & Green Photoautotrophs, Spirochaetes;
    - **Firmicutes** (Low GC gram +) = ***Clostridium***, *Bacillus, Staph.*, ***Mycoplasma***;
    - **Actinobacteria** (High GC gram +) = Corynebacterium, diptheria, ***Mycobacterium***, ***Streptomyces***.
    - **ARCHAEA** = extremophiles! (Hyperthermophiles, Halophiles, Methanogens) – very DIVERSE domain!!! ***No PG cell wall; ether-linked, branched membrane lipids.*** Prokaryotic Diversity: bacterial "giants" = *Thiomargarita, Epulopiscium*.

12. **EUKARYOTES: FUNGI** – chemoheterotrophic Chitin cell wall, sterols in membrane, sexual and asexual spores. Molds & Yeasts; Hyphae, Mycelia. Fungal life cycle – Zygomycete – fusion of haploid hyphae produces **zygospore**, meiosis → release spores → **asexual growth** and **asexual spores** from sporangium. (Plasmogamy followed by Karyogamy). Zygomycota, Ascomycota, Basidiomycota. **LICHENS**: mutualistic. **ALGAE**: Cellulose cell walls and chlorophylls, single and multi-cellular, photoautotrophs. Red, Green, Brown algae (seaweeds). **Diatoms**, **Dinoflagellates**, **Oomycota**, **PROTOZOA**: (chemoheterotrophs, cysts, unicellular euk., asexual or sexual reprod. by "conjugation". **Archaezoa**; **Apicomplexa**. **Ciliates**.. **Euglenozoa**, **Slime molds** – cellular and plasmodial. **HELMINTHS**: **Platyhelminthes** (flat worms) **Nematodes** (round worms, eg: trichinosis, hookworms). **Arthropod Vectors:** Insects, Arachnids.

13. **VIRUSES**: DNA or RNA, double or single-stranded; protein coat, some enveloped, specific host range receptors (spikes). Helical, polyhedral; Obligate intracellular parasites – must be grown in host cells → **Plaques**. Identify – cytopathic effects, serology, PCR. **Bacteriophage Lytic cycle:** Attachment, penetration – inject DNA, biosynthesis, Maturation/ assembly, Release – lysis of host…. **Lysogenic cycle**: ***prophage*** genome integrated into host chromosome. **Animal Viruses:** attachment, penetration by endocytosis or fusion, uncoating, biosynthesis of viral DNA and proteins, Maturation/ assembly, release – budding or rupture. **Retrovirus: *reverse transcriptase***, **Provirus**; **Budding of enveloped viruses**. **PRIONS** --- spongiform encephalopathies, resistant proteins that convert normal cellular proteins to parasitic. **VIROIDS** – small, stable infectious RNAs in plants.

# Microbiology Midterm 3 (Spring 2019): Study Questions
## Possible Short Essay Topics *(be prepared to draw diagrams as well!)*:

1. ***Ch. 8:*** **Distinguish between the starting sequences and ending sequences and enzymes used to initiate, polymerize (elongate), and terminate Replication, Transcription, and Translation. *Define each process, including directions of synthesis, and the type of molecule produced.***
2. **Define and explain how the Central Dogma of molecular genetics illustrates the process by which hereditary Genotype becomes hereditary Phenotype in an organism** *(include **3 processes** involved in **Heredity** and "**Gene Expression**")*.
3. Distinguish between the three types of significant **Point Mutations** in DNA, and compare the possible severity of each type of mutation on an organism's phenotype.
4. **Define and diagram the structure of an Operon, and label and explain the function of at least 6 protein and DNA components involved in its function and in both positive and negative regulation.**
5. **Compare and contrast regulation of the *LAC* Operon and the *TRP* Operon. When is each turned ON or OFF? *Draw each operon in the PRESENCE of its own ligand (signal molecule).* What controls the activity of the regulatory proteins involved? *Explain how each type of regulation is appropriate for an operon encoding catabolic or anabolic enzymes*** *[HINT: How does each contribute to greater **efficiency**, by **conservation of energy and materials** for the cell??]*.
6. Using diagrams, compare and contrast the **three mechanisms of horizontal gene transfer** in bacteria. If DNA is transferred from donor to recipient cell, does that DNA always become a hereditary component of the recipient cell? Explain.
7. ***Ch. 9:*** Distinguish between ***Biotechnology*** and *Recombinant DNA Technology (Genetic Engineering)*. Describe a specific case of Biotechnology that does NOT require recombinant DNA, and one that does use recombinant DNA.
8. Describe the **4 main ingredients** of a **PCR** reaction, and diagram and describe the **three steps in a cycle**. How do these work to amplify (make many copies of) a SPECIFIC segment of DNA out of a whole genome?? How many PCR cycles would you need to amplify 1 DNA segment _____ (number to be given at exam) times? *(be prepared to do the math!)*
9. **Diagram the 4 essential components of a bacterial Cloning Vector (eg: a plasmid). How would you insert your DNA/gene of interest (such as a human or other mammalian gene) into this bacterial DNA carrier? How will you know that recipient bacteria have taken-up your vector? How will you know that vector taken up by bacteria contains your insert?**
10. **Outline and describe TWO methods of identifying whether or not a DNA fragment you have cloned into a vector has the desired specific sequence (gene of interest).** *[Hint: Apply what we have learned about DNA properties and binding specificity.]*
11. ***Ch. 11:*** **You have isolated a previously undiscovered species of prokaryote. Assuming you have a very "high-tech" laboratory, how could you identify whether this organism is Bacteria or Archaea** *(3 ways – consider cell structure, biochemistry, and genetics)***? How would you determine whether the organism, if a Bacterium, is Firmicutes or Actinobacteria or Proteobacteria** *(2 methods distinguish between all 3 phyla)***?** ***[think about Chs. 9 and 11 methods, and our BASIC techniques from lab too!]***
12. ***Ch. 12:*** Describe and diagram two differences between each of the three major Fungal divisions/phyla. Give an example of a representative fungal species from each division.
13. **Compare and contrast the cellular, nutritional, and life cycle (sexual/asexual, haploid/diploid) characteristics of protozoan, algal, and fungal microbes and helminths. Give an example of each that is important to humans or to the environment.**

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14. ***Ch. 13:*** Describe & diagram a basic bacteriophage reproductive cycle. How does this compare to infection by animal viruses? Describe at least 3 special adaptations, including latent phases, that some animal viruses have to avoid the immune system?
15. Diagram and Describe enveloped retrovirus structure and its reproductive cycle. Compare and contrast this with the structure and reproductive cycle of an unenveloped Animal DNA virus. Describe 3 special adaptations, including proviral stages, that retroviruses (and some other viruses) have to avoid the immune system.

***** All questions are important study guides, but BOLDed questions are the most likely essays on the exam.***
***** Remember: A good strategy when answering compare/contrast questions is to make a TABLE of characteristics.***
******* REVIEW and PRACTICE these topics and questions MANY times each, EVERY weekday!!! *******