Bio 102 Practice Problems
Genetic Code and Mutation

Multiple choice: Unless otherwise directed, circle the one best answer:

1. Choose the one best answer: Beadle and Tatum mutagenized Neurospora to find strains that required arginine to live. Based on the classification of their mutants, they concluded that:

   A. one gene corresponds to one protein.
   B. DNA is the genetic material.
   C. "inborn errors of metabolism" were responsible for many diseases.
   D. DNA replication is semi-conservative.
   E. protein cannot be the genetic material.

2. Choose the one best answer. Which one of the following is NOT part of the definition of a gene?

   A. A physical unit of heredity
   B. Encodes a protein
   C. Segement of a chromosome
   D. Responsible for an inherited characteristic
   E. May be linked to other genes

3. A mutation converts an AGA codon to a TGA codon (in DNA). This mutation is a:

   A. Termination mutation
   B. Missense mutation
   C. Frameshift mutation
   D. Nonsense mutation
   E. Non-coding mutation

4. Beadle and Tatum performed a series of complex experiments that led to the idea that one gene encodes one enzyme. Which one of the following statements does not describe their experiments?

   A. They deduced the metabolic pathway for the synthesis of an amino acid.
   B. Many different auxotrophic mutants of *Neurospora* were isolated.
   C. Cells unable to make arginine cannot survive on minimal media.
   D. Some mutant cells could survive on minimal media if they were provided with citrulline or ornithine.
   E. Homogentisic acid accumulates and is excreted in the urine of diseased individuals.

5. The Ames test shows that nitrous acid is a mutagen because it greatly increases the mutation rate for *S. typhimurium* strain TA1535 (in a dose-dependent manner). However, it does not increase the mutation rate for TA1537. The best conclusion is:

   A. Nitrous acid is not likely to be carcinogenic.
   B. Nitrous acid probably causes insertion mutations in DNA.
   C. Nitrous acid probably causes substitution mutations in DNA.
   D. Nitrous acid probably causes nonsense mutations in DNA.
   E. Nitrous acid is less dangerous than sodium azide

6. Which one statement is not true about DNA and RNA?

A. DNA contains deoxyribose and RNA contains ribose.
B. DNA contains T nucleotides and RNA contains U nucleotides.
C. Both DNA and RNA have a sugar-phosphate backbone.
D. Both DNA and RNA use the same purines.
E. DNA is always double-stranded and RNA is always single-stranded.

7. Which one statement is true about the genetic code?

A. The genetic code table lists tRNA sequences.
B. Every protein starts with a Pro amino acid.
C. Each amino acid is encoded by exactly one codon.
D. Only three codons have no matching anticodons.
E. The genetic code table lists anticodon sequences.

Short answer (show your work or thinking to get partial credit):

1. The RNA sequence below encodes a very short protein:

   5' ACCGUACGACCAUG-UCC-CAC-UAU-CCC-UAGGCGAUC 3'

   a. Circle the codon where translation (protein synthesis) by the ribosome will start. Put a box around the codon where it will stop.

      Start codon = AUG (shown in green), stop codon = UAG, UAA or UGA (shown in red)

   b. Use the genetic code table in your text to decode this message: what will be the sequence of amino acids in the protein?

      Codons are separated by hyphens above: AUG=methionine, UCC=serine, CAC=histidine, UAU=tyrosine, CCC=proline

   c. If the underlined base changed from U to G, what would be the effect on the protein? What do we call this kind of mutation?

      This would change the UAU codon to UAG, a stop codon! The protein will end early; we call this a nonsense mutation.

   d. If the underlined base changed from U to C, what would be the effect on the protein? What do we call this kind of mutation?

      This would change the UAU codon to UAC, which also encodes tyrosine. This wouldn't change the amino acids in the protein, so it's a silent mutation.

2. Below is the DNA sequence for a short protein:

   5' AGCTAGACGCATCCTAATG-GCC-ACT-GAA-TCC-TGAATGGACGA 3'

   a. Draw a circle around the start codon and a box around the stop codon.

      Start codon, ATG (or AUG in RNA) is in blue; stop codon (TAG, TGA or TAA) is in purple.

   b. How many amino acids will the protein translated from this mRNA have?

      five (three-base units starting with the start codon, as shown above; no amino acid for the stop codon)

   c. Give the amino-acid sequence of the protein.

      Methionine-Alanine-Threonine-Glutamic Acid-Serine

   d. If a mutation occurs and a C nucleotide is inserted after the red T nucleotide (with a corresponding G on the bottom strand), how will this affect the protein? Be as specific as possible.

      The protein will not change, because the insertion happens before the start codon.

   e. If a mutation occurs and the green A nucleotide is changed to a C (with a corresponding change on the bottom strand), how will this affect the protein? Be as specific as possible.

      The threonine amino acid will be replaced with a proline amino acid.

3. You are investigating a rare genetic brain disorder and would like to identify the gene responsible, clone it and determine its function. A very small protein isolated from the brains of healthy individuals is not present in brain cells of individuals who have the disease. The protein has a molecular weight of about 1000 Da. If the molecular weight of a typical amino acid is 100 Da, about how long do you expect the protein to be, and how long would the corresponding DNA coding sequence be?

   A 1000 Da protein made up of 100 Da amino-acid subunits must be 1000/100 = 10 amino acids long.

   Each amino acid is encoded by a 3-base DNA codon, so for 10 amino acids, there must be 10×3 = 30 nucleotides of DNA (33 if you count the stop codon!).

4. Diseased individuals do not seem to make any of the protein described in #3 above.

   a. List two kinds of mutations that might produce this result.

      (1) A nonsense mutation that inserted a stop codon somewhere early in the protein (so the full-length protein is not made).

      (2) A 1- or 2-base insertion mutation that produces a frameshift within the coding sequence.

      (3) A 1- or 2-base deletion mutation that produces a frameshift within the coding sequence.

      (4) A larger deletion that removes the gene altogether

      (5) A mutation in a regulatory sequence that prevents the gene from being transcribed or translated

      (6) A missense mutation that happens in the start codon (other missense mutations would alter the protein but not get rid of it altogether)

   b. Would you expect these mutations to be dominant alleles or recessive alleles? Explain your reasoning carefully.

      We would expect these to be recessive alleles. Remember that a recessive allele does not show up phenotypically in a heterozygous individual. If a heterozygous person had one "good" allele that encodes a normal protein and one "bad" allele that encodes no usable product at all, that person will be able to make at least some functional protein from the one good allele and should therefore be normal or nearly normal in phenotype.

      It's possible that he or she won't be able to make enough good protein, and we might then get some kind of incomplete dominance, but in most cases, one good copy is sufficient to compensate for one non-functional copy and most non-functional genes actually are recessive mutations.

5. Intron #21 in the CFTR gene is 25 nucleotides long. Suppose that this intron fails to get removed from the mRNA. Predict the kind of mutation that this would introduce to the CFTR protein by circling your answer from the list below *and explain* your reasoning.

   - Missense Mutation
   - Nonsense Mutation
   - Silent Mutation
   - Frameshift Mutation
   - This would not change the CFTR protein

   Explanation:

   This will add 25 nucleotides to the mRNA that will be translated. 25 nucleotides is 8 1/3 codons. So this will insert 8 additional amino acids and after that everything will be out-of-frame.

6. A tRNA in a bacterial cell contains the anticodon 5'GUU3'.

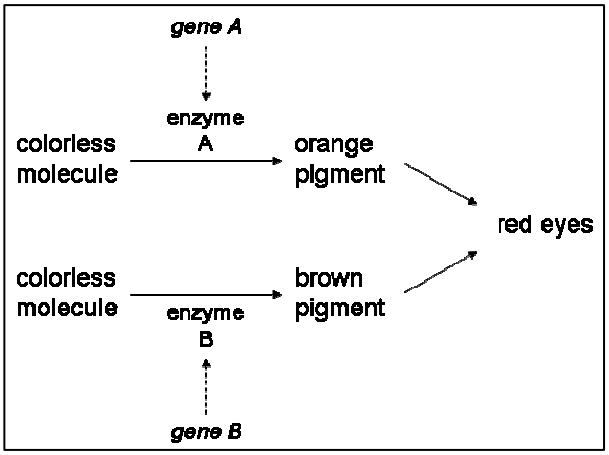
   a. During the 'charging' reaction, what amino acid should be covalently linked to this tRNA molecule?

      The 5'GUU3' anticodon will base-pair to a 5'AAC3' codon. From the genetic code, AAC encodes the amino acid Asn (asparagine)

   b. If this same tRNA with the same anticodon was in a mouse brain cell, should the same amino acid be attached? Please explain.

      Yes. The same amino acid will be attached because the genetic code is universal (ie the same in essentially all organisms from bacteria to mice).

7. Fruit flies (*Drosophila*) normally have bright red eyes. However, the red color that you see is really a combination of an orange pigment produced by one enzyme and a brown pigment produced by a second enzyme, as shown at right.



   a. What would be the genotype and phenotype of a pure-breeding fruit fly carrying a nonsense mutation in gene **A**, assuming gene B functions normally?

      A = normal gene→functional enyzme (dominant)

      a = mutated gene→no enzyme (recessive)

      genotype: aaBB

      phenotype: brown eyes (no orange pigment)

   b. What would be the genotype and phenotype of a pure-breeding fruit fly carrying a nonsense mutation in gene **B**, assuming gene A functions normally?

      B = normal gene→functional enyzme (dominant)

      b = mutated gene→no enzyme (recessive)

      genotype: AAbb phenotype: orange eyes (no brown pigment)

   c. What would be the genotype and phenotype of the F_1 offspring resulting from a mating between these two flies?

      aaBB x AAbb → F_1 offspring all AaBb, red eyes (a functional allele of each gene)

   d. Give the phenotypes and expected ratios of the F_2 offspring resulting from a mating between two of the F_1 flies.

      Each AaBb parent can produce AB, ab, aB and Ab gametes. So, it's an ordinary F_2 cross, with a 4x4 Punnett square. The results are:

      9/16 A-B- (red eyes)

      3/16 A-bb (orange eyes)

      3/16 aaB- (brown eyes)

      1/16 aabb (white eyes – can't make either pigment)

8. A particular strain of *Salmonella typhimurium* is his^- because a TAC codon was mutated to a stop codon.

   a. What would have been the anticodon sequence of the tRNA that would have recognized the original codon?

      3' AUG 5' – note that if you don't indicate which end is 3', then it's assumed that the 5' end is on the left!

```
DNA  5' TAC 3'
          ↓
mRNA 5' UAC 3'
        |||
tRNA 3' AUG 5'
```

   b. What mutation might have occurred to produce the his^- strain?

      Possible single-base mutations:

      (1) TAC → TAG (UAG stop codon)

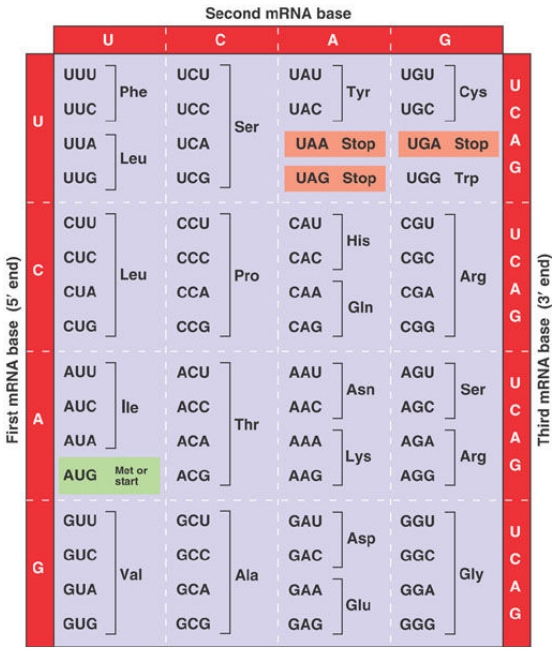
      (2) TAC → TAA (UAA stop codon)

      (3) TAC → TAGC (insertion gives UAG)

      (4) TAC → TAAC (insertion gives UAA)

   c. What is the term for this type of mutation?

      A mutation that produces a stop codon where it doesn't belong is a nonsense mutation. If you chose an insertion as your way of producing the stop codon, then it could also be classified as a frameshift.



d. Is there a different mutation that would have produced the same result?
See “b” above for four different possibilities.

9. The Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) protein is a chloride (Cl^-) facilitated transporter made up of 1480 amino acids and whose gene is located on the long arm of chromosome 7. In healthy humans, the protein is found in the plasma membranes of the cells that line the lungs, allowing the movement of Cl^-. In people with cystic fibrosis, no functional CFTR is made.

   a. Do you expect cystic fibrosis to be inherited in a dominant or recessive fashion? Autosomal or sex-linked?

      An allele which results in synthesis of no protein would probably be recessive to a normal allele (it wouldn't do anything to block or interfere with the normal allele). Chromosme #7 is not the X chromosome, so this trait will be autosomal.

   b. The CFTR gene is approximately 82,500 bp long. However, CFTR mRNA isolated from the cytoplasm is only 6,500 nucleotides in length. Please explain what happened to the missing 76,000 bp.

      The gene probably contained introns, which were spliced out before the mRNA left the nucleus.

   c. A variety of mutations can lead to a defective CFTR protein and the disease. For each listed mutation, describe why this leads to a nonfunctional protein.

      Insertion of a U nucleotide at position 3905.

      This would cause a frameshift mutation: it alters the reading frame so that not only one codon but all subsequent codons are affected. This certainly would produce a non-functional protein.

      Codon 542 is changed from GGA to UGA.

      UGA is a stop codon: this is a nonsense mutation. So, the protein will terminate prematurely and will certainly not be functional.

      A deletion of amino acid 508 means that the CFTR protein never leaves the Golgi.

      Although this protein presumably would be made, if it is trapped in the Golgi, it can't get to the cell membrane where it needs to be to function.

10. The beak of the rare purple-beaked gooney bird is purple because of two enzymes encoded by two separate genes. Enzyme A, the product of Gene *A*, converts a white molecule into a red pigment. Enzyme B, the product of Gene *B*, then converts the red pigment into a purple pigment. The recessive alleles of these two genes produce non-functional enzymes.

   a. What would be the phenotype of an *AAbb* gooney bird? Of an *aaBB* bird?

      An *Aabb* bird has no enzyme B to make red pigment into purple, so its beak will be red. The *aaBB* bird has no enzyme A to make red pigment in the first place, so is beak is white.

   b. In a cross between two *AaBb* gooney birds, what offspring phenotypes would you expect, and what fraction of the offspring would show each phenotype?

      The genotypes will come out in a 9:3:3:1 ratio as always—we just have to remember which genotype goes with which phenotype. As noted above, any bird that is homozygous recessive for *a* (*aaB-*, *aabb*) will be white. So:

      9/16 *A-B-*, purple

      3/16 *A-bb*, red

      4/16 *aaB-* or *aabb*, white (3/16 *aaB-* and 1/16 *aabb*)

11. The year is 2015, and our first manned mission to Mars has just returned with samples of bacteria-like organisms that live in the Martian soil. Chemical analysis shows that they contain nucleic acid—dubbed "MNA" (Martian nucleic acid)—fairly similar to earthly DNA.

    a. MNA is double-stranded but is made up of six different nucleotides, none of which appear in earthly DNA. The table below shows the amount of each nucleotide in MNA. Based on this analysis, what are the base-pairing rules?

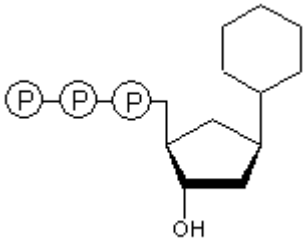
nucleotide	percent of all nucleotides in MNA
Thioguanine (T)	24.3
Orotidine (O)	9.8
Methylcytidine (M)	15.5
Aminoinosine (A)	10.2
Xanthine (X)	25.5
Tetrahydrouridine (U)	14.7

T pairs with X
O pairs with A
M pairs with U

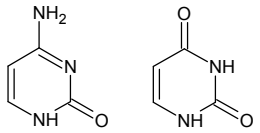
    b. If Martian proteins were composed of the same 20 amino acids used in earthly proteins, what would be the smallest number of MNA bases that could make up a Martian codon? Explain.

Two bases. There are six bases total, so there are 36 combinations of two bases (AA, AT, AO, AM, AX, AU, TT…): $6^2 = 36$. This is more than enough to handle the 20 amino acids.

    c. The bases in MNA are all deoxyribonucleotides. Methylcytidine is a pyrimidine. Sketch a methylcytidine nucleotide.



12. Cytosine (C), a base used in DNA, is very similar to uracil (U), a base used only in RNA: as shown at right, they are different only by one amino (NH_2) group. Sometimes, the amino group is spontaneously removed by an unwanted chemical reaction, converting C to U.



C U

    a. The DNA sequence below is the first part of a gene from a yeast cell. The bottom strand is the template, and the mRNA starts with the first base shown. If the underlined C were converted to U (with a corresponding change on the other strand), what effect would this have on the protein?

```
5′  CGTACCACGCACCAGGATGCCAGACC…
3′  GCATGGTGCGTGGTCCTACGGTCTGG…
```

This would convert the ACG sequence on the non-template strand to AUG. This means that the mRNA would contain an extra AUG before the original AUG start codon. Because eukaryotes start translation at the first AUG after the 5′ cap, this means translation would start in the wrong place. Where the normal cell would read:

AUG|CCA|GAC…

The mutant cell will now read:

AUG|CAC|CAG|GAT|GCC|AGA…

producing an entirely different protein.

    b. If the mRNA above were from *Salmonella typhimurium*, what effect would the same mutation have on the protein?

This mutation should have no effect in this prokaryotic organism. The correct AUG to start translation would be marked by a Shine-Dalgarno sequence, so an extra AUG won't change anything.

True or False? Read carefully: a question is <u>false</u> unless it is <u>completely</u> true!

T■ F☐ 1. Beadle and Tatum isolated many arg- mutants, all of which could grow on minimal media supplemented with the amino acid arginine.

T☐ F■ 2. A mutation is a change in the sequence of DNA that alters the amino acid sequence of the corresponding protein.

T☐ F■ 3. A mutation that substitutes one nucleotide for another can change the amino-acid sequence of a protein but is very unlikely to produce any significant effect on protein function.

T■ F☐ 4. Beadle and Tatum hypothesized that one gene is responsible for one enzyme, and thus for one step in a metabolic pathway.

T■ F☐ 5. There are no tRNA molecules with the anticodons 5′ CUA, 5′ UCA or 5′ UUA.

T■ F☐ 6. Recessive alleles often encode non-functional proteins, or no protein.

T☐ F■ 7. A nonsense mutation is very likely to affect the function of a protein, but a missense mutation is unlikely to have a major effect.

Matching:

1. Below are descriptions of several different mutations. Match each with the <u>one</u> term that would be the best classification for this mutation. You may use the terms once, more than once, or not at all.

D	A mutation changes a G to a C within an intron (no change in protein)
D	A mutation changes a TCA codon to TCG (TCA = Ser, TCG = Ser, no change in protein)
A	A mutation changes a TCA codon to CCA (TCA = Ser, CCA = Pro; 1 amino acid changed)
B	A mutation changes a TCA codon to TAA. (TCA = Ser, TAA = stop)
D	A mutation occurs between the +1 nucleotide and the start codon of a gene (no change in protein in this untranslated region)
F	This kind of mutation changes every amino acid in the protein beyond the point where it occurs (insertion or deletion produces a frameshift)

A. Missense mutation
B. Nonsense mutation
C. Antisense mutation
D. Silent mutation
E. Coding mutation
F. Insertion mutation
G. Stabilizing mutation