Solutions to 7.014 Problem Set 3

Question 1

Hypothetical organism X has the following DNA sequence. Part of the promoter is indicated by the boxed sequence. Transcription starts at and includes the bold A/T base pair.

5′ TATTTGATAG CTCTATGCAT GCATGGGTCC TGAAGTTCAG ATCTTTGAGT CATAGGAGTC 3′
3′ ATAAACTATC GAGATACGTA CGTACCCAGG ACTTCAAGTC TAGAAACTCA GTATCCTCAG 5′

a) Give the RNA sequence of the first 25 bases following transcription.

5′ AGCUCUAUGCAUGCAUGGGUCCUGA 3′

b) What are the first 5 amino acids of the resulting protein?

Met HIS ALA TRP VAL LEU

c) Finish translating the following mRNA sequence.

1

5′ AUA UUU AUG CAU GGG ACU UAU AGC GAU AGC UAC UAA CAU AAG 3′

ile phe met his gly thr tyr ser asp ser tyr

d) The organism that makes the above RNA sequence is exposed to a mutagen. What does a mutagen do?

A mutagen alters the DNA sequence of an organism.

e) For each of the following mutations, identify the type of mutation (insertion, deletion, point, or silent) that occurred in the DNA and the affect on the resulting protein. Consider each mutation independently.

i) C10 → A10  
If the RNA now has an A instead of a C, then the mutation in the DNA was G ← T. Protein has one amino acid different his ← ile.

ii) A40 → U40  
If the RNA now has a U instead of an A, then the mutation in the DNA was T ← A. This change is after the stop codon, so no change in the protein.

iii) T12 → C12  
If the RNA now has a C instead of a T, then the mutation in the DNA was A ← G. This results in CAU ← CAC, but both encode his. No change in the protein.
Question 1, continued

f) You purify protein made from different mutant organisms. Each new protein sequence results from a single change (insertion, deletion, or substitution) in the sequence given. Modify the RNA sequence given such that it would encode each new protein.

i) Ile Phe Met His Gly Thr Tyr Ser Glu Ser Tyr

5’ AUA UUU AUG CAU GGG ACU UAU AGC GAU AGC UAC UAA CAU AAG 3’

A or G

ii) Ile Phe Met Gln Trp Asp Leu

5’ AUA UUU AUG CAUGG GAC UUA UAG C GAU AGC UAC UAA CAU AAG 3’

A or G

Question 2

You are studying replication in your favorite bacteria when you notice the replication fidelity has decreased by a factor of 100.

a) You suspect this is caused by a mutation in the gene for the replicative enzyme. Circle the activity that you suspect is altered and results in this decrease in fidelity.

5’ → 3’ polymerase  5’ → 3’ exonuclease  3’ → 5’ exonuclease
mismatch repair helicase ligase

b) You purify the above protein and find that it is approximately half the size you expect it to be given DNA sequence data. Explain what kind of mutation could cause this result.

Any mutation that causes a premature stop such as UGG, UGU, or UGC → UGA.

c) Your roommate in course 5 has designed a unique amino acid and creates a tRNA charged with this new amino acid. This new tRNA recognizes the codon 5’ UGA 3’. You add this charged tRNA to the replication deficient mutant cells from above and find that now the fidelity is increased. Speculate why the addition of the novel tRNA results in this increased fidelity.

The unique amino acid is now inserted at the UGA created by mutation that was described above. This produces a protein that is full length, restores the function of this protein, and therefore we see an increase in fidelity.
Question 3

Martians, tired of hiding from our spacecraft, send their own spacecraft to Earth with some single-celled Martian organisms. They are willing to give these to NASA on the condition that we leave Mars alone.

Instead of having only 4 bases, their DNA has 8 different bases: B, C, D, E, W, X, Y, and Z. Chemical analysis of a single-celled Martian organism reveals that its genome is composed of:

B: 13%  C: 7%  D: 13%  E: 19%  W: 19%  X: 11%  Y: 7%  Z: 11%

a) Identify which bases pair with each other.
   B - D
   C - Y
   E - W
   X - Z

b) Martian DNA replicates in a manner analogous to that seen with earthly DNA. Given this Draw a replication fork and label the following:

   leading strand
   lagging strand
   RNA primer
   2 Okazaki fragments
   5’ and 3’ ends (10 total)
Question 4

a) On the above graphic...
   i) Label the N (amino) terminal amino acid of the protein being made.
   ii) Box the 3 bases encoding the first amino acid of the protein being made.
   iii) Circle the part of the schematic where tRNAs would bind.

b) Draw the tRNA encoding trp, include the sequence for the anticodon. Be sure to label the 5’ and 3’ ends.

   ![tRNA Diagram]

   3’
   \[\text{A C C}\]
   5’

   5’
   3’

   ![tRNA Diagram]

   3’
   \[\text{A C C}\]
   5’

   c) Would a substitution within a codon for trp always change the resulting protein sequence? Explain your answer.
   Yes, there is only one codon for trp, so any substitution will change the protein.

   d) Would a substitution within a codon for thr always change the resulting protein sequence? Explain your answer.
   No, there are 4 codons for thr, so even with a substitution the codon could still encode thr.