GENETICS 603
Exam 1
Fall, 2004

I. A) Give 2 reasons that people were certain that protein would prove to be the genetic material.

-the information content (20 amino acids) was too great
-chromosomes carried genes and contained protein

B) What were the most convincing aspects of the Avery/Macleod/McCarty experiments in showing that DNA is responsible for heredity?

the use of proteases and nucleases; treatment with protease did not eliminate transformation but treating with DNAase did

C) If radioisotopes had been available to them, they might have convinced even more investigators. What isotopes would they use, what would they label, and how would they be used to enhance their results?

-the best bet would be the ability to prove that pure DNA was not contaminated with ^{35}S (protein)

(If they could get labeled T they could specifically label DNA, but feeding ^{32}P would not end up just in DNA like it did in the virus experiments of Hershey and Chase. In addition, unlike the viruses, bacteria have more than just protein and DNA. They can degrade proteins, incorporate the amino acids, have multiple molecules that include P, so measuring incorporation of a small amount of ^{32}P in progeny would unlikely convince many.

II. Natural transcripts (mRNAs) of eukaryotic genes that encode proteins are translated very poorly or not at all in prokaryotic systems. List and describe features that contribute to low efficiency.

Initiation signals differ; the leader of eukaryotic messages lacks the sequence that hybridizes to the ssrRNA (AGGAGG or Shine-Delgarno sequence) so would not attach. Codon preferences could also slow reading a great deal.

III. Some temperature sensitive mutants in *E. coli* stop DNA replication almost instantly when shifted to the restrictive temperature, others lead to a gradual halt of DNA replication and still others allow completion of active replicons but no new starts

a) Predict the kinds of mutations (in general) that lead to the temperature sensitive responses.

missense

b) Predict whether temperature sensitive mutations in the following components of DNA replication would cause instant or delayed halt or would just slow DNA replication:

1) the 5' to 3' activity of DNA polymerase III - instant halt

2) the 5' to 3' activity of DNA polymerase II slow (repair defect)

3) the 3' to 5' activity of DNA polymerase III slow (repair defect)

4) the ori-binding protein delayed (could finish any already started)

5) Helicase -instant or delayed (if can't unwind, cant proceed)

IV. We tend to think of the 3 types of RNA required for protein synthesis, but in fact many other types of RNA play important cellular roles. Examples mentioned during lecture included the following: What is the role of each?

snRNPs	intron removal
gRNA	guide for mRNA editing
SRP	signal receptor particle for membrane trafficing
Telomerase	maintain telomeres (ends of chromosomes)

V. Students in GENE 604, a companion lab course that has not been offered recently, performed the Ames test with a variety of test compounds. One of these was a food coloring that contained red dye #2. When the dye was added to a disc on a test plate, nearly confluent growth surrounded the disk. A) Give 3 conceptually different explanations for their observation.

1) the dye contains a mutagen
2) the dye contains a source o histidine
3) the dye contains his' bacteria (this was the actual case!)

B) Three test strains used were designed to detect different types of mutations, specifically transitions, transversions and frameshifts. Give an example or define each of these kinds of mutation.

transition: AT <-> GC (purine replaces purine)
transversion AT <-> TA; GC <->CG, AT <->CG
frameshift add or subtract 1 or 2 bases

C) If the same results described above were seen on plates with a) all 3 strains, b) with only one strain, which of your explanations would be most likely?

a) 2 or 3 b) 1 (mutagen present)

VI In 1995, Eisensmith and Woo listed known nucleotide substitutions that lead to premature termination in the human PAH gene (encodes the enzyme that converts phenylalanine to tyrosine).
a) complete the table below by listing the original codon(s) and tell whether the mutation is a transition or transversion. If more than one codon is listed, circle the one that is most likely present.

mutation	original codon(s)	type of mutation
trp to UGA	UGG	transition
gln to UAG	CAG	transition
arg to UGA	CGA(most likely) or AGA	transition / transversion
trp to UAG	UGG	transition
ser to UGA	UCA	transversion
gly to UGA	GGA	transversion
tyr to UAG	UAU/C	transversion
gln to UAA	CAA	transition
tyr to UAA	UAU/C	transversion

b) List any <u>other</u> codons that could lead to a nonsense codon by a single base substitution.

ser: UCG to UAG; leu: UUA/G to UAA/G; lys: AAA/G to UAA/G;
cys: UGU/C to UGA

c) What is characteristic of the list in part b?

all are transversions

d) Predict the phenotype of the mutants. How would affected individuals be detected and treated?

Since this is the enzyme that converts phe to tyr, nonsense mutations would make it nonfunctional, meaning that homozygous individuals would suffer from "PKU" (phenylketonuria) (low IQ, pale etc.)
they would be detected by the Guthrie test in which the high levels of phe in the blood protect bacteria from a toxic analog and treated by a low phe diet

e) Give examples of two chemicals with different modes of action (include in answer) that could cause the majority of mutations in part a.

5BU- base analog; HNO2, oxidative deamination; EMS, methylating agent, etc

VII. What is the GT -- AG rule? How do we know that it is not sufficient to explain intron removal?

Introns begin with GT and end with AG; there are other GT and AT sequences that are not removed

VIII. What is the primary reason for degeneracy in the genetic code?

wobble (pairing precision of 3rd base in anticodon is less than normal)