Online Supplemental Material, Shuster Genetics and Genomics Competencies for Nurses

Biol 219, Public Health Microbiology

Voluntary Genetics Pre-Test, Fall 2008

***** Note that an answer key to selected BIO and TECH questions has been provided, as has a description of the scoring for these representative questions******

Please write your name on each page of this **voluntary** pre-test. Please circle your answer for multiple-choice questions, and write answers to short-answer questions in the space provided. This **voluntary** test does **NOT** count for any points in the course. It is completely optional. Although it does not count for course points, it will allow me to assess how effective the course is at helping student meet several genetics and genomics competencies, as recently developed by the Consensus Panel on Genetic/Genomic Nursing Competencies. It is purely for informational purposes, to assess learning and teaching in the course (as explained on the Participant Informed Consent Form). All analysis will occur on an **ANONYMOUS** basis, after the course has been completed (*i.e*. results of this test and this analysis will have NO IMPACT on your course grade).

B: Biological Basis/Implications of Genetic Diversity
1 pt
1. The emergence of pandemic strains of influenza is most likely due to
a. conjugation
b. antigenic shift
c. transformation
d. antigenic drift

T: Technology & Genetics
1 pt
2. PFGE fingerprinting, or bacterial DNA profiling, is done in order to
a. compare the overall genetic relatedness of two strains of bacteria
b. determine the presence or absence of specific genes of interest in different strains of bacteria
c. determine which alleles of specific genes are present in different strains of bacteria
d. determine the antibiotic-resistance profile of different strains of bacteria

B: Biological Basis/Implications of Genetic Diversity
total of 8 pts (4 & 4)
3. Treating an HIV positive patient with a particular antiviral drug will often result in the treated patient developing an HIV population that is resistant to the drug.
a. Why does this happen? (be as specific as you can with respect to the mechanism(s) that lead to the patient developing a drug-resistant HIV population)

The elements that were considered to constitute a complete response included:

- ***a genetically-diverse population of HIV***
- ***reverse transcriptase is a major reason for the genetically diverse population***
- ***the drugs provide a selective pressure, only those viruses with a resistant allele will be able to replicate (natural selection)***
- ***putting these elements together in a cogent and coherent fashion***

Pre-test average score: 14% (0.56 out of 4 possible points)
Post-test average score: 63.3% (2.53 out of 4 possible points)

<u>Pre-Test Answer distribution</u>

- ***Don't know or Blank: 17.8%***
- ***Virus mutates: 42.2%***
- ***Virus evolves: 4.4%***
- ***Virus is "immune": 11.1%***
- ***Patient's immune system: 6.7%***
- ***Other: 17.8%***
    - ***e.g. HIV mutates its "cells"***
    - ***e.g. no known cure***
    - ***e.g. drug and virus (both) mutate***

<u>Post-Test Answer Distribution</u>

- ***Completely correct: 26.7%***
- ***Missing one or two elements: 28.9%***
- ***Only had one element (e.g. high mutation rate): 22.2%***
- ***Answers focusing on using one drug vs. a drug cocktail: 11.1%***
- ***Other: 11.1%***

b. Describe a treatment option that can help circumvent this resistance problem, and explain HOW it works to circumvent the resistance problem.
B: Biological Basis/Implications of Genetic Diversity
2 pts
c. In light of your answers to part a. and b. of this question, explain how between 11 and 27% of newly infected, untreated adolescents have strains of HIV that are resistant to nucleoside or nonnucleoside reverse transcriptase inhibitors.

B: Biological Basis/Implications of Genetic Diversity
8 pts
4. Why do retroviruses, such as HIV, tend to have a higher spontaneous mutation rate than bacteria?

B: Biological Basis/Implications of Genetic Diversity
12 pts
5. Complete the following table to compare and contrast antigenic shift and antigenic drift.

	Antigenic SHIFT	Antigenic DRIFT
a. What is it?		

	Antigenic SHIFT	Antigenic DRIFT
b. (i) Can ALL viruses do this process? (ii) If not, which viruses can/can't and WHY?		
c. Why is this process important or significant?		

Example-specific question (used ONLY in overall, but not as a B/T/M question)
6 pts (total)
6. a. What is the role of CCR5 in HIV infection?
b. Explain how a person's CCR5 genotype influences their resistance to HIV infection and progression.

B: Biological Basis/Implications of Genetic Diversity
1 pt
7. How do antibiotics contribute to antibiotic resistance?
a. by killing ALL the bacteria in a population
b. by causing mutations in bacterial genes (i.e. antibiotics are mutagenic)
c. by providing a selective environmental pressure
d. by generating bacterial diversity by inducing antigenic shift

Correct answer: c. by providing a selective environmental pressure

Pre-test % correct: 4.4%
Post-test % correct: 84.9%

Pre-test distribution of responses:
a. 8.9%
b. 53.4%
c. 28.9%
d. 28.9%
Blank. 4.4%

Post-test distribution of responses:
a. 0%
b. 9.4%
c. 84.9%
d. 5.7%

B: Biological Basis/Implications of Genetic Diversity
1 pt
8. When bacteria acquire an R plasmid transferred by a phage, it is a type of horizontal transfer known as
a. transformation
b. natural selection

c. transduction
d. conjugation

B: Biological Basis/Implications of Genetic Diversity
1 pt
9. What properties does a bacterial cell acquire when it acquires an R plasmid by horizontal transfer?
a. the ability to produce catalase
b. the ability to be resistant to one or more antibiotics
c. the ability to sporulate
d. the ability to produce a capsule

T: Technology & Genetics
4 pts total (2 & 2)
10. Pathogenic strains of *Vibrio cholerae* carry a toxin gene- it is this toxin gene that makes these strains pathogenic. Non-toxigenic strains of *V. cholerae* do not cause cholera.
a. What genetic-based test can be carried out to detect the presence of the toxin gene?
b. What type of test can be carried out to detect the actual toxin protein itself?

B: Biological Basis/Implications of Genetic Diversity
1 pt
11. You have been taking an antibiotic but are still sick. What might be going on?
a. you have become immune to the antibiotic
b. you have become resistant to the antibiotic
c. the bacteria causing the infection are resistant to the antibiotic
d. all of the above are possible

Correct answer: c. the bacteria causing the infection are resistant to the antibiotic

Pre-test % correct: 42.2%
Post-test % correct: 90.5%

Pre-test distribution of responses:
a. 4.4%
b. 11.2%
c. 42.2%
d. 42.2%

Post-test distribution of responses:
a. 0%
b. 3.8%
c. 90.5%
d. 5.7%

12. AIDS specialists will order genetic tests for HIV-positive patients prior to initiating anti-retroviral therapy.

T: Technology & Genetics
4 pts
(a) What **SPECIFICALLY** is being tested, and what type of genetic method/technique is used to do this test?

The components of a complete answer included:

- ***naming both reverse transcriptase and protease as the HIV genes being investigated (in a genotyping assay)***
- ***identifying PCR (amplification) and sequencing as genetic-based tests***
- ***being able to make the connections and explain the answer clearly***

Average Pre-test score: 0.8% (0.03 points/4 possible points)
Average Post-test score: 24.7% (0.99 points/4 possible points)

Distribution of Pre-Test Responses

- ***Don't Know or Blank: 68.9%***
- ***Invoking patient genes/genetics/blood work: 20%***
- ***Some aspect of HIV in the patient (but not specific): 4.4%***
- ***Other: 6.7%***

Distribution of Post-Test Responses:

- ***Responses that included the HIV gene names: 4/45***
- ***Responses that included the human CCR5 (HIV co-receptor) gene: 7/45***
- ***Responses that included PCR and/or sequencing (of any gene): 23/45***
- ***Responses that included (incorrectly) PFGE/DNA Profiling: 24/45***
- ***Responses that included (incorrectly) MIC or an ELISA: 13/45***
- ***Blank responses: 1/45***

Note: Individual responses on the post-test generally included more than one element. For example: PCR and sequence analysis of the CCR5 gene of the patient. While this does include PCR and sequencing, it does not relate these methods to the relevant HIV genes.

B: Biological Basis/Implications of Genetic Diversity
4 pts
(b) WHY does the doctor order this test prior to initiating the anti-retroviral therapy? That is, how will the doctor use the results of this test in terms of patient care?

13. Strains of Salmonella are referred to as serotypes. Some of these serotypes include *Salmonella* Dublin and *Salmonella* Saintpaul (the latter has been somewhat notorious this past spring and summer due to its involvement in a multi-state outbreak). The serotypes are based on O antigens, H antigens and K (capsular) antigens.

M: General Microbiology
2 pts
13.a. What kind of test is used to determine the serotype of a strain? State the general type of test, and a specific example of this type of test.

M: General Microbiology
4 pts
13.b. Given that Salmonella strains have O and H antigens, what can you infer about their cell wall structure and motility? Explain your answer.

13.c. The following is from a July 23, 2008 CDC Update:

> "An FDA laboratory detected Salmonella Saintpaul with the outbreak strain fingerprint pattern in a sample of jalapeño pepper obtained from a distribution center in McAllen, Texas."

T: Technology & genetics
4 pts
13.c.(i) What type of test is a "fingerprint" test? What is it looking at? Is the same or different than a serotype test?

Used in overall as part of 13.c., but not as a specific (B, T, G type)
13.c.(ii) Why was the finding of the outbreak strain on a jalapeño significant? (i.e. why was it important to find this particular strain on this particular pepper?)

M: General Microbiology
7 pts
14. For each of the following tests, mark them as G (genetic-based test), S (serological-based test) or B (biochemical/metabolic-based test)

_____ ELISA

_____ MIC test

_____ PCR to detect *N. gonorrhoeae* in urethral exudate

_____ catalase test

_____ testing to determine if a *V. cholerae* isolate is O1 or O139

_____ PFGE fingerprinting/profiling

_____ coagulase test

15. The figure below is from the article "Update to CDC's Sexually Transmitted Diseases Treatment Guidelines, 2006: Fluoroquinolones No Longer Recommended for Treatment of Gonococcal Infections". MMWR (2007) 56(14): 332-336. It shows ciprofloxacin resistance in *N. gonorrhoeae* isolates over time. Resistance was based on MIC tests.

M: General Microbiology
9 pts
15.a. Describe

(i) what a MIC test is

(ii) what information it gives you

(iii) how you would set one up (you may use words or a diagram).

B: Biological Basis/Implications of Genetic Diversity
3 pts
15.b. Based on the information shown in the figure, why are fluoroquinolones, such as ciprofloxacin, no longer recommended in the treatment of gonococcal infections?

M: General Microbiology
15.c. Describe the appearance of *Neisseria gonorrhoeae* on a Gram stain (include color, cell shape and arrangement)

M: General Microbiology
16. For each of the following pathogens, describe (i) what they would look like on a Gram stain (color, shape, arrangement of cells), (ii) a brief description of their cell wall structure and (iii) the disease that they cause.

Staphylococcus aureus

(i)

(ii)

(iii)

M: General Microbiology
4 pts
17. *Vibrio cholerae* is a marine organism (*i.e*. it grows in salt ocean water), and can grow on culture medium containing 2-3 % NaCl (a relatively high salt concentration).
a. What happens to a typical bacterial cell is a high salt environment (and why does this happen)?
b. What compensatory mechanisms might *V. cholerae* use to prevent this from happening to their cells?

Biol 219, Public Health Microbiology
Take-Home Assignment #3

This assignment will help you master several of the following learning objectives for Unit 3:

By the end of this unit you will be able to

- Explain several mechanisms by which pathogens generate genetic diversity
- Explain the role of genetic diversity in resistance to therapy and treatment failure
- Describe the relationship between a genotype and a phenotype, using several bacterial, viral and human loci as examples
- Predict the outcome of an infection and/or treatment protocol, based on the results of genotyping tests
- Use the results of genetic-based tests to determine whether or not pathogens involved in different infections are related, and explain the significance of knowing the relatedness of pathogens

1. In your own words, explain why HIV can mutate so quickly, relative to other viruses and relative to bacterial populations. (2 points)
2. Now explain (in your own words) why/how a population of HIV in a treated patient will respond to antiretroviral therapy, both in terms of amount of virus (viral load) and in terms of the predominant genotypes and phenotypes present in the population before the therapy is initiated, and after the therapy has been underway for an extended period of time. (4 points)
3. You are working as a nurse, and one of your nursing colleagues sustains a needle stick (through their gloves) from a needle that had been used to draw blood from an HIV-infected patient. The patient has been on AZT, Atazanavir and Rescriptor.

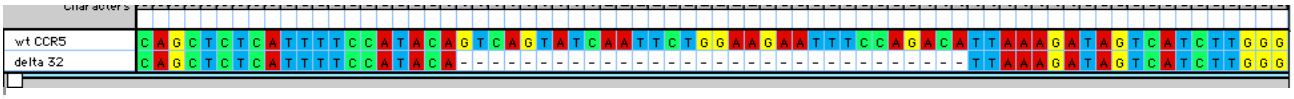
a. For each of the three anti-retroviral drugs listed above, figure out what the general type of drug they are, and what that means about its function. For example, if penicillin was on the list (which it obviously wouldn't be, as it is not an anti-retroviral drug), I would say it's a beta-lactam antibiotic, and it binds to and inhibits the activity of PBPs, thereby preventing cross-linking of the peptidoglycan cell wall (and of course give a citation for a reference, which I would then annotate in my annotated bibliography!) (6 points)

b. Given that the patient whose blood infected your colleague had been on anti-retroviral medications, is it possible for your colleague to have acquired drug-resistant HIV (even though your colleague has never tested positive for HIV or had antiretroviral therapy)? How does this relate to your answer to question 2, and what are the practical implications of this? (2 points)

c. Based on our discussions, and your work on the previous questions, should AZT, Atazanavir or Rescriptor be part of the post-exposure prophylaxis for your colleague? Why or why not? (2 points)

4. The first part of this assignment has focused on viral genetic diversity. Let's now look at host (human) genetic diversity.
Here is some background information about what the virus needs to enter cells (see your text and other sources for more information): As a virus, HIV requires a host cell in order to complete its life cycle, and both a receptor (CD4) and a co-receptor (CCR5) to bind to on the surface of its host cells. It turns out that not everyone in the human population has the same amount of CCR5 on their cell surfaces, due to a mutation that deletes 32 nucleotides from the middle of the CCR5 coding region (CCR5 delta-32). The CCR5 delta-32 mutation ends up preventing the CCR5 protein from being localized to the cell surface.
Here is some background on genetics/genotype terminology: humans have two copies of every gene (one from mom and one from dad), and these alleles can be the same or different. To be sure we are all on the same page with respect to terminology, the CCR5 delta-32 allele (the allele with the deletion) is RECESSIVE- that means its effect on the phenotype is not apparent if a "wild-type" or "normal" (undeleted) allele is also present.
a. Predict the phenotypes with respect to the ability of HIV to infect the cells with each of the following genotypes (3 points):
(i) homozygous wild-type (or dominant)
(ii) heterozygous
(iii) homozygous recessive
b. Search for what proportion of the population (or sub-groups) of the population may have heightened resistance to HIV infection based on their CCR5 genotype. (2 points)

5. The diagram below shows a portion of the gene (nucleotide) sequence of a wild-type CCR5 allele (TOP), and the same portion of the delta-32 CCR5 allele (BOTTOM). There is only one DNA strand shown for each allele. SO THIS IS NOT SHOWING a DOUBLE-STRANDED DNA MOLECULE!!!! It is showing how the nucleotide sequences of the TWO DIFFERENT alleles align with one another. Note that the white boxes with dashes in them represent nucleotides that are MISSING from the delta-32 CCR5 allele. So to read the nucleotide sequence of the delta-32 allele (bottom), you would just read the actual nucleotides (colored boxes) in order, skipping over the blanks-so from the first C to the last G. In order to practice with how a genotype corresponds to a protein sequence (and phenotype), let's practice by comparing these two CCR5 alleles.



Use the DNA-based genetic code (one is available at http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/C/Codons.html) to translate codons 1-6 (again, start with the first marked codon- the CAT) of each CCR5 allele (the CCR5 wild-type and the delta-32 CCR5 allele). You'll need to write out the FULL NAMES of the amino acids (don't just use their one-letter codes).
WT amino acid sequence (2 points):

CCR5 delta-32 amino acid sequence (2 points):

5 points will be deducted if there is no annotated bibliography.
1 point will be deducted if there are no in-text reference citations.

Excerpts from the Fall 2008 Biol 219 (Public Health Microbiology) Syllabus

Overall Course Goals (Why you are here…)

By the end of this 3-credit lecture course, you will be able to

- *analyze infection-related scenarios from a microbiological perspective (i.e. you will be able to "think like a microbiologist") and*
- *meet several of the essential competencies under the professional practice domain of the recently established nursing competencies for genetics and genomics*

Unit 1: Structure, Identification and Treatment of Pathogens

Unit 2: Bacterial Growth and Reproduction in Different Environments

Unit 3: Microbial Genetics

This unit will address the significance of genetic diversity in bacterial and viral populations, mechanisms by which bacteria and viruses generate genetically diverse populations and the implications of this genetic diversity on pathogenesis and treatment. We will also consider the implications of genetic diversity of the human host population on infections. Finally we will outline some common methodologies to assess genotypes and genetic relatedness.

By the end of this unit you will be able to

- Explain several mechanisms by which pathogens generate genetic diversity
- Explain the role of genetic diversity in resistance to therapy and treatment failure
- Describe the relationship between a genotype and a phenotype, using several bacterial, viral and human loci as examples
- Predict the outcome of an infection and/or treatment protocol, based on the results of genotyping tests
- Use the results of genetic-based tests to determine whether or not pathogens involved in different infections are related, and explain the significance of knowing the relatedness of pathogens

Monday	Wednesday	Friday
	10/28 Why are genetically diverse populations important? (Natural Selection)	10/10 Exam 2 (see above)
10/13 How do pathogens	10/15 How else can bacteria	10/17 More horizontal transfer

Monday	Wednesday	Friday
generate genetically diverse populations? (Mutation)	generate genetically diverse populations? (Horizontal transfer)	
10/20 How do viruses generate genetically diverse populations?	10/22 Resistance and treatment failure	10/24 Genotyping In-class GRADED activity
10/27 DNA profiling	10/29 In-class GRADED case study	10/31 Intro to Unit 4 **ASSIGNMENT 3 DUE**
11/3 **EXAM 3**		

Unit 4: Infectious Disease (pathogenesis, defenses and vaccination)