

## SSN SBPM Workshop Exam One

### Short Answer Questions & Answers

#### 1. Describe the effects of DNA damage on the cell cycle.

ANS : DNA damage causes cell cycle arrest at a G2 checkpoint. This arrest allows time for DNA damage to be repaired before mitosis. DNA damage also slows progression through the S phase and arrests the cell cycle at a G1 checkpoint, to repair DNA before errors would be replicated. G1 arrest is mediated by the protein, p53.

#### 2. Describe in general the activities of cyclin-dependent kinases (cdk's). How are they regulated?

ANS : Cdk proteins associate with specific cyclins (proteins that are synthesized or degraded during different parts of the cell cycle) to drive progression through stages of the cell cycle. Cdk activities are regulated via:

- association of cdk with specific cyclin partner.
- activation by phosphorylation of conserved threonine 160 residue by CAK (cdk activating kinase).
- Inhibitory phosphorylation of tyrosine residues near amino terminus by Wee1 protein kinase. Inhibition is removed by dephosphorylation by Cdc 25 family of protein phosphatases.
- Binding of CKIs (Cdk inhibitors) to Cdk/cyclin complexes.

#### 3. What does it mean to say that insulin is made as a pre-pro-insulin molecule? Describe the steps involved in going from pre-pro-insulin to functional insulin. Where does each step occur?

ANS: "Pre-pro" indicates that two protein cleavages must occur in order to create a functional insulin hormone. The "pre" portion of the molecule is a signal peptide that brings the ribosome and emerging protein to the rough ER and targets the protein to the lumen of the rough ER. This "pre" peptide is cleaved from pro-insulin in the lumen of the rough ER. The "pro" portion of the molecule, also known as the C-peptide (C for connecting), assists in folding insulin so that the appropriate disulfide bonds can be made. The C-protein is cleaved within the secretory granule and secreted with the insulin molecule.

*Fun Fact: When folks with Type I diabetes are getting exogenous insulin, it can be hard to tell whether they are still producing insulin. Looking for C-peptide is a way to measure endogenous insulin production.*

**4. The eukaryotic ribosome is 80S. It is made of up of two subunits: 60S and 40S. The 60S subunit is made of three rRNAs: 28S, 5.85S, and 5S.**

**A recent report from the National Institute of Standards and Technology indicates that  $60+40=100$ , making 80S seem 20% too low. Similarly, according to the Congressional Budget Office  $28+5.85+5=38.85$ , making 60S seem more than 50% too high. Are the government math experts wrong? Give two explanations.**

ANS:

- 1) “S” stands for sedimentation coefficient, a measure of how fast suspended particles move when they are spun in a centrifuge. It is not a measure of mass or length and it is not additive. So  $60S + 40S$  can equal 80S.
- 2) Ribosomal subunits are not composed solely of rRNA. There are also protein components. Therefore, the three pieces of rRNA (28, 5.85, 5) and a bunch of proteins together can have a sedimentation coefficient of 60S. If you are wondering how those ribosomal proteins are first made, contact City Council Speaker, Gifford Miller or dial 311.

#### **5. Gene Frequency Statistics.**

**Alpha 1-antitrypsin deficiency is an autosomal recessive disorder with an incidence of 1/2000 in Denmark**

- a. **What does the pattern of inheritance of an autosomal recessive disorder look like? How does it differ from an X-linked recessive disorder pattern?**
- b. **What is the difference between incidence and gene frequency?**
- c. **What is the probability that an individual from Denmark with no phenotypic expression of the disease is a carrier for the disorder?**

ANS:

**a.** An autosomal recessive trait is (almost always, with exceptions due to environmental mutagens) expressed only in homozygotes (ie, aa), and thus must have received this recessive allele from each parent. Please see Figure 3 in the syllabus lecture “Patterns of Inheritance” for an illustration of a recessive autosomal inheritance pattern. Note the infrequency of the expressed trait.

Autosomal recessive inheritance differs from X-linked recessive in that an X-linked mutation is expressed in all males who receive it, but only in females who are homozygous for the trait, which is rare. Note that if a male is affected the X-linked recessive trait often skips a generation and re-emerges if there was a daughter carrier who has a son. See Figure 5 from the “Patterns” lecture.

Ps- It is a good idea to review Mitochondrial Inheritance as well.

b. In the discussion of genetics in this lecture, **Incidence** refers to the rate of phenotypic expression of a trait in a population and gene frequency refers to the frequency of an allele in the population.

c. The probability that a person from Denmark is a carrier of the recessive trait is equal to  $2pq$  from the Hardy-Wienberg equation,  $p^2+2pq+q^2$ . Thus,

$$\begin{aligned}q^2 &= 1/2000 \\q &= .02 \\p+q &= 1 \\ \text{and } 1-p &= .98 \\p &= .98\end{aligned}$$

and  $2pq = 2 \times .98 \times .02 = .039$ , or about 4% carriers of the recessive allele in the population.

## 6. Multifactorial Inheritance

a) **What differentiates multifactorial inheritance from Mendelian inheritance? Define each, then note the primary difference.**

b) **Are you more likely to have CAD, a disease which displays multifactorial inheritance patterns, if your mother or father has CAD? First, state whether your mother or father is more likely to have the disease based on syllabus information. Second, explain why it is important to your risk calculation which parent carries the disease based on the principles of multifactorial inheritance.**

ANS:

**A.** inheritance of single genes in which the traits segregate within families and generally occur in fixed proportions among the offspring of different combinations of alleles. There are dominance patterns and recessive patterns based on the dominance or recessivity of the allelic expression.

**Multifactorial Inheritance** is a non-classical pattern of single gene inheritance in which the single gene disorders that appear to run in families are neither single-gene or chromosomal in origin. This type of inheritance is from a combination of genetic and non-genetic factors, they recur within families, but they Do Not show any particular pedigree pattern in an individual family.

**B.** You are more likely to have CAD if your mother has it. This is because CAD is more prevalent in men, and the risk is higher for relatives of patients of the less susceptible sex (rule #5 of multifactorial inheritance, review all rules ☺).

Sons of the female patient are more likely to express the trait than daughters, because they are the more susceptible sex for this disease. Review how sibship risk differs in multifactorial inheritance vs. mendelian.

**7. The common feature of all cancers is uncontrolled cell growth. However, there are several mechanisms by which uncontrolled growth is achieved in a cell. Experimental evidence indicates that retinoblastoma, a cancer, is caused by a mutation in the RB gene. However, loss of one retinoblastoma (*Rb*) gene does not lead to cancer.**

**a) How can you explain this?**

This must mean that some active RB is still being produced by the other Rb gene, and that is adequate to prevent cancer.

**b) Based on this evidence, is Rb a proto-oncogene or a tumor suppressor gene?**

Because mutation in 1 gene locus is not sufficient to cause cancer, RB is a tumor suppressor gene. For tumor suppressor genes, only total loss of function both of the gene loci (a knockout of the gene on both alleles) will lead to cancer.

**c) Knowing that “two hits” are required, you are surprised to see a 4-year-old patient in your office one day with an apparent tumor in one eye. How can you explain the onset of the cancer at such a young age?**

Most people inherit two normal copies of the Rb gene. However, it is possible to inherit one chromosome with the Rb locus deleted, effectively knocking out one copy of the Rb gene right from the start (13q14 deletion). In these patients, only one additional “hit” in any eye cell is necessary to deactivate the other Rb gene. This is termed an “inherited predisposition” to that cancer. For people with no germline mutation of the Rb gene, retinoblastoma is extremely rare, because any one cell would have to receive two hits over its lifetime- this is termed a “sporadic” development of the cancer.

**d) How would the situation be different for a proto-oncogene?**

Proto-oncogenes cause problems by being “turned on,” unlike tumor suppressors, where problems are caused when they are “turned off.” For that reason, only one “hit” is necessary to begin cancer with a proto-oncogene.

**8. You are an obstetrician and deliver a child to a 35-year-old woman. The child is her first, but is diagnosed with Down syndrome.**

**a) What type of error leads to Down syndrome, and what is one risk factor?**

Down syndrome is trisomy of chromosome 21 (3 copies instead of 2), and usually arises through meiotic nondisjunction. This nondisjunction error can occur in either parent and in either meiosis I or meiosis II although the mechanisms are slightly different. In the case of trisomy 21, 95% of cases are of maternal origin, and the main

risk factor is maternal age, with the rate of trisomy 21 rising exponentially with maternal age after 32. Amniocentesis is how we test in utero for trisomy 21.

**b) What is the mother's risk of having a second child with Down syndrome?**

After one trisomic delivery, even at a young age, a woman has two times the age-related risk of another trisomic pregnancy. Assuming she has her second child at 37, her risk is then  $2 \times 6.1$  per 1000 = 12.2 per thousand, or 1.22%.

**9. You are working in a genetic counseling clinic, where you see people who are at an increased risk for having children with genetic diseases. You have a blatantly clueless medical student with you for their first clerkship. You ask him to explain the difference between physical mapping and genetic mapping to determine the distance between genes.**

**a) Assuming he isn't a deer caught in the headlights, what does he say?**

Physical mapping provides the locations and distances between genes in physical measurements, such as DNA base pairs. Genetic mapping uses a different method, linkage analysis, to arrive at gene distances using the observed amount of recombination between genes.

**b) What is the definition of two genes that are linked?**

The two genes travel together in a pedigree more often than expected by chance.

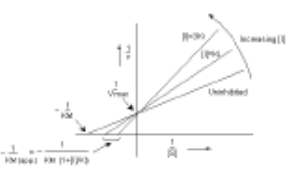
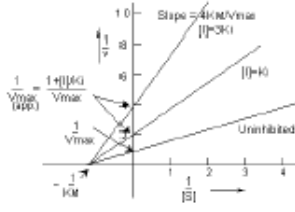
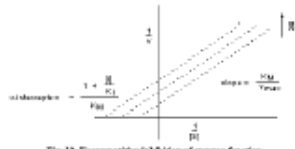
**c) How do we study linkage in a clinical setting?**

By studying a series of families and calculating (through laboratory analysis) what percentage of offspring of informative meioses are recombinant. 50:50 recombinant:nonrecombinant is what is expected by chance for independent loci. A higher ratio than 50:50 may suggest linkage, and a value can be assigned based on the percentage recombination to calculate the physical distance between the loci.

**10. Describe briefly the major differences between enhancer and promoter sequences:**

Promoters' position and orientation in relation to the gene are essential for proper function. The position and orientation of enhancers can be changed significantly without loss of their enhancing function. (Other responses are acceptable)

**11. Contrast the effects of competitive, non-competitive, and uncompetitive inhibitors on  $V_{max}$  and  $K_M$ .**

	Competitive Inhibitor	Noncompetitive Inhibitor	Uncompetitive Inhibitor
<b>Binding</b>	<ul style="list-style-type: none"> <li>• Binds E, not ES</li> <li>• Binds active site</li> <li>• Inhibitor resembles S</li> </ul>	<ul style="list-style-type: none"> <li>• Binds E and ES</li> <li>• Binds away from active site</li> <li>• Inhibitor doesn't resemble S</li> </ul>	<ul style="list-style-type: none"> <li>• Binds ES, not E</li> </ul>
<b>Effect on <math>K_M</math></b>	Increases	No change	Decreases
<b>Effect on <math>V_{max}</math></b>	No change	Decreases	Decreases
<b>Effective Range</b>	Low [S] <i>Can overcome with high [S]</i>	High and low [S] <i>Cannot overcome with high [S]</i>	High [S] <i>Effect not noticed at low [S]</i>
<b>Completely Unrigorous Explanation</b>	Since S must compete with inhibitor for E, a higher [S] is required to reach $\frac{1}{2}V_{max}$ (so $K_M$ is higher). $V_{max}$ is unchanged, however, because a very high [S] can swamp out the effects of inhibitor.	Assuming the inhibitor binds E and ES with equal affinity, the presence of inhibitor doesn't bias the distribution between E and ES, so $K_M$ is unchanged. The inhibitor does, however, effectively decrease $[E]_T$ , so $V_{max}$ is reduced.	By binding a fraction of ES, the inhibitor favors E + S association, <i>increasing</i> the apparent affinity (decreasing $K_M$ ). However, even at high [S], where all E is bound up as ES, some fraction of ES is still bound by inhibitor, so $V_{max}$ is less. More important for multisubstrate enzymes.
<b>Lineweaver-Burk Plot</b>	 <p>Fig. 8 Competitive inhibition of enzyme function.</p>	 <p>Fig. 9 Non-competitive inhibition of enzyme function.</p>	 <p>Fig. 10 Uncompetitive inhibition of enzyme function.</p>