The 7.014 Final Exam will be:

Wednesday, May 24
9:00 am - 12:00 noon

Johnson

*YOU MUST BRING YOUR ID.

This will be a cumulative, closed book exam.

Please bring your PRS devise—we will collect them at the exam.

Additional Office hours will be held in the coming week. Times and locations will be posted.
Question 1

Your ongoing interest in bioluminescent organisms has lead you to study the Hawaiian squid, *Euprymna scolopes*. Most nocturnal creatures cast shadows under the bright Hawaiian moonlight, and are easy prey to predatory fish. This type of squid can shine light downwards to match the moonlight and avoid casting a shadow thereby decreasing predation.

a) In their native ocean environment, the squid feed on a wide variety of small fish and invertebrates. You suspect that the squid is the keystone species in this ecosystem. What experiment could you do to test this theory? What result do you predict?

To provide squid for your studies, you maintain a large, self-supporting squid tank. Surprisingly, your tank-raised squid are not luminous. In fact, the glow seen in free living squid is due to a species of bacteria, *Vibrio fischeri* that live in a specialized light organ within the squid.

b) When juvenile squid hatch, ambient water enters the light organ. *V. fischeri* and the many other bacterial species in this ambient water also enter the light organ and colonize it. All the bacteria initially flourish in this nutrient rich environment, but after 10 hours, only *V. fischeri* remain in the light organ.

   i) The interaction between *V. fischeri* and the other bacterial species initially colonizing the light organ can best be described as ________________.

   ii) The interaction between the squid and *V. fischeri* can best be described as ________________.

   iii) Is the light organ of the squid the fundamental niche of *V. fischeri*. Why?

   iv) Is the light organ of the squid the realized niche of *V. fischeri*. Why?
Question 2

As the semester comes to a close, you discover some interesting fruit fly mutants around your dorm’s kitchen. Blue eyes and green antennae are two phenotypes new to these mutants. When you cross males with both traits to wild type females you find that all the F1 male offspring have wild type eyes and antennae while all the female offspring have both blue eyes and green antennae:

Males with blue eyes and green antennae  X  normal females

All males are normal
All females have blue eyes and green antennae

a) What is the most likely mode of inheritance for both of these traits?

When you cross F1 flies to each other you find the following results for the males in the F2:

F1 normal males  X  F1 blue eyes and green antennae females

F2: Out of 250 males:  113  blue eyes, green antennae
                        112  wild type
                        12   blue eyes, normal antennae
                        13   normal eyes, green antennae

b) What is the recombination frequency between the genes for the blue eyes and green antennae traits?

c) Would you expect to see F2 female flies with blue eyes and normal antennae? Why or why not?
a) Match each of the following structures to the type of molecule it represents:

1) protein       2) RNA       3) DNA

[Diagram of a protein structure]
[Diagram of an RNA structure]
[Diagram of a DNA structure]
**Question 3, continued**

b) Fill in the blanks in this representation of the central dogma.

Choose from:

- nucleus, translation, RNA, membrane, DNA,
- cytoplasm, polysaccharide, transcription, or protein

This process is ____________________ .

In eukaryotes, this process occurs in the ____________________ .

This process is ____________________ .

In prokaryotes, this process occurs in the ____________________ .

**Question 4**

In analyzing differences between star squash players and armchair warriors, you have discovered a protein that exists only in the squash players. You design substrates that will bind to the "star protein".
Question 4, continued

a) Give the name for the strongest intermolecular interaction between the substrate and the following amino acids on the star protein. Choose from ionic bond, covalent bond, hydrogen bond, and van der Waals forces.

<table>
<thead>
<tr>
<th>Amino Acid</th>
<th>Strongest interaction</th>
</tr>
</thead>
<tbody>
<tr>
<td>i) Val</td>
<td></td>
</tr>
<tr>
<td>ii) Glu</td>
<td></td>
</tr>
<tr>
<td>iii) Asp</td>
<td></td>
</tr>
<tr>
<td>iv) Ala</td>
<td></td>
</tr>
</tbody>
</table>

b) You make the following additional substrates

Does the type of interaction between the Ala of the star protein and the substrate change with substrate 2 as compared to substrate 1?

Does the type of interaction between the Glu of the star protein and the substrate change with substrate 3 as compared to substrate 1?

c) Which substrate would you expect binds the most tightly to the star protein?

substrate 1  substrate 2  substrate 3

Why?
STRUCTURES OF AMINO ACIDS
at pH 7.0

ALANINE (ala)

ARGININE (arg)

ASPARAGINE (asN)

ASPARTIC ACID (asp)

CYSTEINE (cys)

GLUTAMIC ACID (glu)

GLUTAMINE (glN)

GLYCINE (gly)

HISTIDINE (his)

ISOLEUCINE (ile)

LEUCINE (leu)

LYSINE (lys)

METHIONINE (met)

PHENYLALANINE (phe)

PROLINE (pro)

SERINE (ser)

THREONINE (thr)

TRYPTOPHAN (trp)

TYROSINE (tyr)

VALINE (val)
Question 5

To investigate the yeast metabolic pathway for serine biosynthesis, you screen for serine auxotrophs (mutants which are unable to grow without serine supplied in their growth medium). You isolate four such mutants, and test them for growth on medium supplemented with several intermediates (A, B and C) known to be part of the pathway. The results are shown below ("+" represents growth, "-" represents no growth).

<table>
<thead>
<tr>
<th>Strain</th>
<th>minimal medium</th>
<th>minimal + A</th>
<th>minimal + B</th>
<th>minimal + C</th>
<th>minimal + serine</th>
</tr>
</thead>
<tbody>
<tr>
<td>wild type</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>m1</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>m2</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>m3</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>m4</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
</tbody>
</table>

You then mate the haploid m1 strain with the haploid m4 strain to create a diploid yeast carrying both the m1 and the m4 mutations. You test the diploid for growth on the same conditions as above and observe that the diploid exhibits the same growth requirements as m1 or m4 haploid.

a) Are the m1 and m4 mutations in the same gene or different genes? Briefly explain your reasoning.

b) Draw the metabolic pathway for the synthesis of serine, consistent with the data given above. Include the intermediates (A, B, and C) and serine, and indicate which mutants (m1, m2, m3, m4) are defective at each step in the pathway.

c) You create a haploid strain that has both the m1 and m3 mutations.

   i) This haploid mutant will grow on media supplemented with which of the following intermediate(s) (A, B and/or C)?

   ii) Which of the following intermediate(s) (A, B and/or C) will accumulate when this haploid mutant is grown on minimal medium?
Question 6

a) Indicate whether the following statements are true or false. If false, correct the statement or provide a brief explanation for why the statement is false.

i) DNA replication is initiated at promoter sequences in the DNA.

ii) RNA polymerase requires primers to initiate RNA synthesis.

iii) Okazaki fragments are the short fragments of DNA that are produced on the leading strand at the DNA replication fork.

iv) The 5' to 3' direction of DNA synthesis implies that deoxyribonucleotides are added to the 5' OH group on the growing strand.

v) Transcription terminates at stop codons in the mRNA sequence.

b) Shown below is the DNA sequence of a gene from a virus that encodes a short viral peptide, and also the sequence of the mRNA synthesized from this gene.

**genomic DNA sequence:**

```
5' - AGCTCATGTGCGAGTCCTGACGCTGACGTAGG - 3' 
3' - TCGAGTACACGCTCAGGACTGCGACTGCATCC - 5'
```

**mature mRNA sequence:**

```
5' - UCAUGUGCGAACGCUGACGUAGG - 3'
```

i) In the genomic DNA sequence shown above, draw boxes around the exons.

ii) Write the sequence of the peptide encoded by this gene. Indicate the NH₃⁺ and the COO⁻ ends of the peptide.
### Question 7

a) Indicate whether the following statements are true or false. And if false, correct the statement or provide a brief explanation for why the statement is false.

i) Plasma B cells secrete antibody into the bloodstream.

   ii) T cells produce antibodies that neutralize antigen.

   iii) Each B cell can make many different kinds of antibodies.

b) When a rabbit protein is injected into rabbits, no antibodies against this protein are generated. If, however, the same rabbit protein is injected into guinea pigs, the guinea pigs generate antibodies against the rabbit protein. Briefly (in one or two sentences) explain this observation.

c) The genomes contained in almost all of the somatic cells in an adult human are identical. Name one (diploid) cell type that is an exception to this and name the primary mechanism by which this cell type arose.
**Question 8**

You are studying a common genetic condition. The mutant allele differs from the wild-type allele by a single base-pair (bp) substitution. This substitution eliminates a NheI restriction site that is present in the wild-type allele. (The mutant allele is not cut by NheI.) A pedigree of a family exhibiting this condition is shown below:

You isolate DNA from four individuals in the pedigree. Using PCR techniques, you amplify a 1000 bp portion of their DNA which includes the site affected by the mutation. You digest the PCR products with NheI and analyze the resulting DNA fragments on a gel:

![DNA gel image]

a) Based on these data, is this gene located on an autosome or the X-chromosome? Briefly justify your reasoning.

b) Based on these data, is the mutant phenotype dominant or recessive to wild-type and why?

c) If individuals 3 and 4 have a daughter, what is the probability that she will be affected? Justify your reasoning.
d) You sequence the region around the NheI site in the wild-type PCR product. You then sequence the corresponding region in the mutant PCR product and discover that not only did the mutation eliminate the NheI site in the mutant allele but it has created a new PvuII restriction site. The recognition sites for the two enzymes are indicated below.

NheI cuts: 5' GCTAGC 3'  PvuII cuts: 5' CAGCTG 3'  3' CGATCG 5'  3' GTCGAC 5'

A portion of one strand of the wild-type DNA sequence is shown below:
5'....GCTAGCTG...3'

What is the sequence of this same region in the mutant allele? Indicate the 5' and the 3' ends of the DNA sequence.

e) Individuals 1 and 2 have another child, 9, who is affected by the genetic condition.

You PCR amplify the 1000 bp region affected by the mutation from individuals 1, 2, and 9, digest the PCR products with NheI or PvuII, and analyze the restriction fragments on a gel:

What event occurred and how does this explain the data shown above?
Question 9

Consider the following hypothetical chromosomal region containing two genes, \textit{fadA} and \textit{fadB}, necessary for the breakdown of oleic acid in a bacterium.

\[ \text{Px} \quad \text{fadX} \quad \text{P} / \text{O} \quad \text{fadA} \quad \text{fadB} \]

\text{P = Promoter}
\text{O = Operator}

The FadX protein, which is continuously produced, binds to the operator in the presence of oleic acid.

a) Is the FadX protein a repressor or an activator of the \textit{fadA} and the \textit{fadB} genes? Briefly justify your reasoning.

b) For each of the following mutants (m1 - m4), predict the level of FadA in the presence of oleic acid. Circle either "Low" or "High".

\begin{center}
\begin{tabular}{l l l}
\textbf{Level of FadA with oleic acid present} & \\
m1 & O is deleted & Low & High \\
m2 & Loss-of-function mutation in \textit{fadX} & Low & High \\
m3 & P is deleted & Low & High \\
m4 & FadX is always bound to O & Low & High \\
\end{tabular}
\end{center}

Question 10

The Philadelphia chromosome results from a translocation event where pieces of chromosomes 9 and 22 switch. No DNA is lost, it is just rearranged.
Question 10, continued

If cells that have a translocation like that described above were to undergo gamete production, some of the gametes would have a complete genome and others would not.

a) The process of gamete production is called _________________.

b) If cells with the combination of chromosomes shown in the picture above form gametes,
   i. What combinations of chromosomes will be present in gametes that have a full set of genetic information?

   ii. What combinations of chromosomes will be present in gametes that are missing genetic information?

Chronic Myeloid Leukemia (CML) is the cancer associated with the Philadelphia chromosome. The Philadelphia chromosome translocation creates a novel gene by attaching a portion of the coding region of the ABL gene (normally found on chromosome 9) to a portion of the BCR gene found on chromosome 22. This fusion gene can be transcribed and translated to create a novel protein, the Bcr-Abl* protein.

![Diagram of chromosome 22 with genes ABL and BCR and primers](image)

You want to design PCR primers to quickly determine if a cell is carrying the Philadelphia chromosome.

c) Which pair of primers could you use to amplify the BCR-ABL fusion gene?

- 5’ GGAATTCC 3’  and  5’ TAACTTA 3’
- 5’ CCTTAAGG 3’  and  5’ ATTCAAT 3’
- 5’ TAAGTTA 3’  and  5’ GGAATTCC 3’
- 5’ ATTGAAT 3’  and  5’ CCTTAAGG 3’
Solutions:

Question 1:

a) You would need to exclude the squid from an area of the ocean. If squid are the keystone species, you would expect to see a decrease in the species diversity of the small fish and invertebrates.

b) i) The interaction between *V. fischeri* and the other bacterial species initially colonizing the light organ can best be described as competition.

ii) The interaction between the squid and *V. fischeri* can best be described as symbiotic or mutualism or mutualistic. The squid are less visible to predators, and the bacteria are provided a safe, nutrient rich environment.

iii) No, The fundamental niche is anywhere that the *V. fischeri* can exist. You know that *V. fischeri* can also live in the ambient water, and it is likely that they could exist in a wide variety of places.

iv) No, The realized niche is where you actually find the *V. fischeri*. You know that *V. fischeri* can also live in the ambient water, so the light organ can not be the realized niche.

Question 2:

a) X-linked dominant

b) \((12+13)/250=10\%\)

c) Yes, we would expect some of these non-parental females. The father, and F1 male, would give a wild type X chromosome to this F2 daughter. If the mother gave a recombinant X chromosome (which we know are produced because there are recombinant sons), then a blue-eyed, normal-antennae daughter can be produced.

Question 3

a) In order, they are DNA, RNA, and protein

b) DNA \(\rightarrow\) RNA \(\rightarrow\) protein

c) DNA \(\rightarrow\) RNA \(\rightarrow\) protein

This process is **transcription**. In **eukaryotes**, this process occurs in the **nucleus**.

This process is **translation**. In **prokaryotes**, this process occurs in the **cytoplasm**.

Question 4

a) The m1 and m4 mutations are in the same gene. This is a complementation test. The diploid strain has the same growth phenotype as the haploid single mutants; the two mutations fail to complement.
(fail to produce the wild-type phenotype) in the double heterozygote. The m1 and m4 mutations must both inactivate the same gene (which codes for an enzyme essential for serine biosynthesis) so that the diploid double mutant has two mutant alleles of the same gene.

b)  

\[ C \xrightarrow{m1, m4} B \xrightarrow{m3} A \xrightarrow{m2} \text{serine} \]

c) i) This haploid mutant will grow on media supplemented with intermediate A.

ii) Intermediate C will accumulate when this haploid mutant is grown on minimal medium.

Question 5

a) i) van der Waals

ii) hydrogen

iii) ionic

iv) van der Waals

b) strongest interaction with substrate 2 is van der Waals

strongest interaction with substrate 3 is ionic

c) substrate 3 would bind the most tightly. In substrate 3, you have two ionic bonds and two van der Waals. In substrates 1 and 2, you have one ionic bond, one hydrogen bond and two van der Waals.

Question 6

a) i) FALSE. DNA replication is initiated at the origin of replication. RNA polymerases bind to promoter sequences to initiate transcription.

ii) FALSE. DNA polymerase requires primers to initiate DNA replication.

iii) FALSE. Okazaki fragments are made on the lagging strand of the replication fork.

iv) FALSE. DNA synthesis occurs by addition of dNTPs to the 3' OH growing strand.

v) FALSE. Transcription terminates at the termination stop sites in the DNA. Translation stops at stop codons in the mRNA.

Question 6, continued

b)  

\[ \begin{align*}
5' &-\text{AGCTCATGTGCGAGTCCTGACGCTGACGTAGG} - 3 \\
3' &-\text{TCGAGTACACGCTCAGGACTGCGACTGCATCC} - 5
\end{align*} \]

ii) NH$_3^+$-met-cys-glu-arg-COO$^-$

Question 7

a) i) True.

ii) False. B cells produce antibodies.

iii) False. Each B cell makes one distinct kind of antibody.
b) The rabbit protein is recognized as foreign (non-self) by the guinea pig.

c) B cells, by gene rearrangement of Ab genes (VDJ rearrangement). Also, T cells (by rearrangement of T cell receptor genes).

**Question 8**

a) An autosome, because individual 6, a male, has 2 alleles.

b) The mutant phenotype is recessive, because individuals 5 and 6 each have one copy of the mutant allele, m, and are both phenotypically normal.

c) 1/4. Since individuals 3 and 4 already have an affected child, then they must both be heterozygotes.

d) 5’...GCCAGCTG...3’

e) A mutation occurred which led to the production of a new mutant allele, m*. This mutant allele has a recessive phenotype and its PCR product is cut by neither Nhel nor PvuII. Individual 9 has the genotype m/m*.

**Question 9**

a) The FadX protein is an activator, since FadA and FadB need to be synthesized in the presence of oleic acid.

b)

m1: If the activator FadX cannot bind to the operator, transcription of fadA will be low.

m2: Without a functional FadX activator transcription of fadA will be low.

m3: Without the promoter, RNA polymerase cannot transcribe the fadA gene, leading to a low level of FadA.

m4: If FadX is always bound, it will lead to the constitutive expression of fadA, leading to a high level of FadA.

**Question 10**

a) Meiosis

b) (i) (9; 22) and (9:22; 22:9)

   (ii) (9; 22:9) and (9:22; 22)

c)

5’ TAAGTTA 3’ and 5’ GGAATTCC 3’