1. One flower pigment pathway in roses is shown below:

\[ \text{White} \quad \xrightarrow{E1} \quad \text{orange} \quad \xrightarrow{E2} \quad \text{red} \]

A true breeding white was crossed to a true breeding orange and all the F1 progeny were red.

A. How many genes must function to make red flowers? 2

B) Give a legend that shows how flower color is inherited in this case.

\[ \begin{align*}
\text{A}_, \text{B}_ & \quad \text{red} \\
\text{A}_, \text{bb} & \quad \text{orange} \\
\text{aa}, \text{B} & \quad \text{white} \\
\text{aa}, \text{bb} & \quad \text{white}
\end{align*} \]

C) Use your legend to show the genotype of the white and orange parents and the red F1:

\[ \begin{align*}
\text{white} & \quad \text{aa, BB} \\
\text{orange} & \quad \text{AA, bb} \\
\text{red} & \quad \text{Aa, Bb}
\end{align*} \]

D) What ratio of flower colors would be expected in the F2 from selfing the F1?

9 red : 3 orange : 4 white

2) Spores from 4 different Neurospora mutants that would not grow on minimal medium (Min) but would grow if the amino acid arginine (ARG) was added to minimal medium were tested for growth on 3 potential ARG precursors; ASC, ORN and CIT with the following results (+ growth, -, no growth).

<table>
<thead>
<tr>
<th>Mutant</th>
<th>Min</th>
<th>ASC</th>
<th>CIT</th>
<th>ORN</th>
<th>ARG</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>2</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>3</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>4</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

Show the pathway for arginine synthesis and show which enzyme is missing in each mutant strain.

\[ \begin{align*}
\text{M1} & \quad \text{ORN} \\
\text{M2} & \quad \text{CIT} \\
\text{M3} & \quad \text{ASC} \\
\text{M4} & \quad \text{ARG}
\end{align*} \]
3. With regard to phenylketonuria in humans:

a) How is it inherited? **Homozygous recessive, single gene**

b) What is the most noted effect of failure to treat PKU? **Very low IQ**

c) How is it treated? **Special diet with limiting phenylalanine**

4. The following 15 base sequence is from the **middle** on one strand of the DNA in a gene that codes for a 200 amino acid protein:

\[\begin{align*}
5' &- \text{TGGCAGCTAATGCAG} - 3' \\
3' &- \text{ACCGTCGATTACGTC} _5' \\
\end{align*}\]

a) Write the sequence of the complementary DNA strand just below the given strand.

b) Recalling that mRNA is always made in the 5’ to 3’ direction, which strand is the template strand? **Given strand X my strand** (the other would give a stop which can’t be in the middle of the coding sequence)

c) Draw a labeled picture of translation, including the product, when the last codon of the mRNA from this gene segment has just come into position.
5. Although mutations occur in DNA, the effects are also reflected in mRNA and any protein encoded. (check each correct response in the questions below.)

A) A **missense** point mutation involving the UUG (Leu) codon could give the following change:

\[ \underline{\text{Leu}} \underline{\text{X}} \text{Ser} \underline{\text{Ile}} \underline{\text{Stop}} \underline{\text{X}} \text{Val} \underline{\text{X}} \text{Met} \underline{\text{X}} \text{Trp} \underline{\text{X}} \text{Arg} \underline{\text{X}} \text{Tyr} \]

B) Now check those missense changes from above that represent a single **transition** mutation:

\[ \underline{\text{Leu}} \underline{\text{X}} \text{Ser} \underline{\text{Ile}} \underline{\text{Stop}} \underline{\text{Val}} \underline{\text{X}} \text{Met} \underline{\text{X}} \text{Trp} \underline{\text{X}} \text{Arg} \underline{\text{X}} \text{Tyr} \]

C) Now check those changes that represent a nonsense mutation:

\[ \underline{\text{Leu}} \underline{\text{X}} \text{Ser} \underline{\text{Ile}} \underline{\text{Stop}} \underline{\text{Val}} \underline{\text{X}} \text{Met} \underline{\text{X}} \text{Trp} \underline{\text{X}} \text{Arg} \underline{\text{X}} \text{Tyr} \]

D) **Chemicals** likely to cause transition mutations include:

\[ \underline{\text{X}} \text{EMS} \underline{\text{X}} \text{HNO}_2 \underline{\text{X}} \text{5BU} \underline{\text{X}} \text{acridine dye} \underline{\text{X}} \text{UV light} \underline{\text{X}} \text{Dioxin} \underline{\text{ICR170}} \]

E) Chemicals likely to cause frameshift mutations include:

\[ \underline{\text{EMS}} \underline{\text{X}} \text{HNO}_2 \underline{\text{X}} \text{5BU} \underline{\text{X}} \text{acridine dye} \underline{\text{X}} \text{UV light} \underline{\text{X}} \text{Dioxin} \underline{\text{ICR170}} \]

F. Chemicals or agents that would show a positive Ames test include:

\[ \underline{\text{X}} \text{EMS} \underline{\text{X}} \text{HNO}_2 \underline{\text{X}} \text{5BU} \underline{\text{X}} \text{acridine dye} \underline{\text{X}} \text{UV light} \underline{\text{X}} \text{Dioxin} \underline{\text{ICR170}} \]

6. In the blank before each characteristic, tell whether it is a feature of eukaryotes only (E), prokaryotes only (P) both (B) or neither (N).

\[ \text{N} \text{ The nuclear membrane remains intact through mitosis} \]

\[ \text{B} \text{ AUG is the usual start codon} \]

\[ \text{E} \text{ Mitochondria are present in the cytoplasm} \]

\[ \text{P} \text{ The DNA is in a closed circle} \]

\[ \text{B} \text{ All 3 stop codons are used} \]

\[ \text{P} \text{ F-met is inserted at the beginning of mRNA translation} \]

\[ \text{E} \text{ Introns must be removed before mRNA can be correctly translated} \]

\[ \text{N} \text{ Messages are translated 3’ to 5’} \]

\[ \text{P} \text{ Messages often encode several enzymes for a specific pathway} \]

\[ \text{B} \text{ The genetic code is degenerate} \]

\[ \text{B} \text{ Multiple copies of the genes for rRNA are present} \]

\[ \text{B} \text{ TATA boxes are present in promoters of genes that make mRNA} \]

\[ \text{P} \text{ Translation is inhibited by ‘mycin’ antibiotics} \]

\[ \text{E} \text{ Messages have a polyA tail} \]

\[ \text{N} \text{ rRNA codes for ribosomal proteins} \]
7. a) Which of these tools could be used in making a plasmid-based shotgun clone library?

X EcoRI  _____ Reverse transcriptase  _____ Oligo dT primer
_____ pUC19  _____ phage λ  _____ RNAse H  X DNA ligase

b) Which of these tools are required for making a plasmid-based cDNA clone library?

_____ EcoRI  X Reverse transcriptase  X Oligo dT primer
X pUC19  _____ phage λ  X RNAse H  X DNA ligase

c) Which of the following would be used for PCR amplification of a DNA sequence.

_____ EcoRI  _____ Reverse transcriptase  _____ Oligo dT primer
X TAQ polymerase  _____ RNAse H  _____ DNA ligase
_____ forward primer  X _____ all 4 dNTPS  X _____ reverse primer

8. Tell whether β-gal’ase, permease and transacetylase would be absent (A) constitutive (C) or regulated (R) in the following E. coli strains. Assume a minus component is inactive and that FS indicates a frameshift. Note that 4 strains have 2 copies of the Lac Operon per cell.

<table>
<thead>
<tr>
<th>Strain</th>
<th>β-gal’ase</th>
<th>permease</th>
<th>TA’ase</th>
</tr>
</thead>
<tbody>
<tr>
<td>P I P O Z Y A</td>
<td>C</td>
<td>C</td>
<td>A</td>
</tr>
<tr>
<td>P I P O Z Y A</td>
<td>C</td>
<td>A</td>
<td>C</td>
</tr>
<tr>
<td>P I P O Z Y A / P I P O Z Y A</td>
<td>R</td>
<td>R</td>
<td>R</td>
</tr>
<tr>
<td>P I P O Z Y A / P I P O Z Y A</td>
<td>R</td>
<td>R</td>
<td>R</td>
</tr>
<tr>
<td>P I P O Z Y A / P I P O Z Y A</td>
<td>R</td>
<td>A</td>
<td>R</td>
</tr>
<tr>
<td>P I P O Z Y A / P I P O Z Y A</td>
<td>C</td>
<td>C</td>
<td>C</td>
</tr>
</tbody>
</table>

9. The normal allele of a gene has 2,000 nucleotides (2Kb) between two EcoR1 cut sites, but a defective allele has an extra internal site that gives two 1Kb fragments. DNA from a man, his wife, and fetal cells were tested using a DNA probe that was inside the first half of the gene with the following results: Man and wife; both 2 Kb and 1 Kb fragments. Fetus: 1 Kb only.

a) What is the procedure called that allows detecting the fragments with a probe?

Southern Blotting

b) What are the fragments called? RFLPs

c) is the fetus normal or affected? yes

d) How do you know? It only had 1KB fragments, it did not have the normal 2KB allele

10. Finish this 6 base restriction endonuclease target site: 5’ G T C G A C