1A  Research has shown that histone acetylation is triggered by neuronal activity in rat brains during fear conditioning and memory formation.

a)  What enzymes are involved in the histone acetylation status of the cells involved? HATs (histone ecetyl transferase) add acetyl, HDACs (Histone deacetylases) remove them.

b)  What does the increase in acetylation infer about expression of the underlying genes?

Increasing ‘loosens’ the binding of DNA by masking positive charged lysine or arginines in the exposed tails of histones; thus increasing transcription and gene expression.

c)  What might be expected if acetylation or deacetylation could be inhibited?

Inhibition of acetylation should prevent expression and make fear conditioning/memories less while inhibiting deacetylation might increase or maintain expression levels beyond normal levels

1B  In the yeast *S. pombe*, the maintenance of centromeric heterochromatin is associated with H3K9 methylation. Maintenance requires intact functioning of the RNAi mechanism,

a)  What is ‘heteochromatin’?  highly condensed, inactive chromatin

b)  What is meant by H3K9 methylation?  Adding a methyl group to the amino acid K (lysine) at position 9 of histone 3

c)  What is RNAi?  Briefly explain how it works.

RNA inhibition; it works when a double stranded RNA is cleaved into fragments (around 21 nucleotides) by dicer. The fragments are carried by Argonaut proteins to the RISC complex where one of the strands can be used to find a complementing sequence in any mRNA and cause it to be ‘sliced’ or to prevent its translation. The dsRNA may come from a virus, from siRNA or microRNAs transcribed as a means of rapidly shutting down expression of a specific gene.

d)  Suggest a model for how RNAi could be involved in maintaining H3K9 methylation.

A microRNA could be made that would function as a guide destroy the messages for a gene that makes a histone demethylase (note- soemof these are being investigated for anti cancer potential)
2. Genes A and B are on the long and short arms of the same chromosome. An individual is heterozygous for both genes with both dominant alleles inherited from one parent and both recessives from the other. 2.1 Use labeled (show alleles) diagrams to show this chromosome pair in metaphase of a) mitosis and b) meiosis I.

![Diagram of chromosomes in metaphase](image)

a)                        

2.2. Give the genotypes of both daughter cells of each division (assume no crossovers occur).

a)_________2 Aa;Bb_________   b) _________ A B & a b _________

3. It has been shown in Arabidopsis that the normal CDC20.1 gene product is involved in a checkpoint between metaphase and anaphase.

a) What is a likely criterion for proceeding at this checkpoint?

Verification that all kinetochores are connected to centromeres and to spindle fibers (and others OK)

b) Predict two “opposing” potential outcomes of a defective CDC20.1 gene.

1) Division could begin before it should resulting in aneuploid cells with extra or missing chromosomes

2) No cell division could occur,

4. You would like to clone a gene from an organism such as *Eragrostis tef* (a cereal grass) for which very, very limited DNA sequence information is available. However, you know the gene has been cloned from some other cereal species and you have evidence that the protein it encodes is found in high levels in leaves exposed to UV light. You also are able to compare amino acid sequences of the protein from other species.

a) Very briefly, list 4 significantly different approaches you might take to clone the gene from *Eragrostis tef*. 
1) shotgun clones, likely detecting fragments with portions of the gene via Southern blots using sequences from the closest relative.

2) cDNA cloning using UV treated leaves as a source of enhanced levels of mRNA

3) Potentially generating fragments with degenerate (if necessary) PCR primers made based on regions of high sequence identity from relatives

4) Using the amino acid sequence to predict degenerate PCR primers to amplify fragments of the gene which could be assembled or used to identify clones with that sequence.

b) For one of the approaches, describe the steps, enzymes, etc. you would use to clone, verify, and express the gene in *E. coli* or another microbe.

This varies with the method most chose to use cDNA cloning and that makes a lot of sense if the goal is to get an expressible copy of the gene. Start with mRNA capture and Reverse transcription based on oligo-dT and likely end with an attempt to express a full length clone in Pichia, but no points lost if stopped at *E. coli*.

5. Crosses between two red eyed flies gave progeny in the ratio of 250 red and 70 purple. Different researchers came up with one gene (simple dominance) and 2 gene (13:3 epistasis) models. First provide a legend showing the genotypes and associated phenotypes for each model, then determine if either model is incompatible with the observed data? (show your work). Also suggest a genetic method for determining which model is correct.

One gene legend  
R_ red  
rr  pruple

Two gene:  
A_ B_ Red  
A_ bb  purple  
aa_ _ red

(a red to purple to red pathway will work)

<table>
<thead>
<tr>
<th>One gene model, 320 progeny phenotype observed</th>
<th>expected</th>
<th>ob-exp</th>
<th>(ob-exp)^2/Ex</th>
</tr>
</thead>
<tbody>
<tr>
<td>red</td>
<td>250</td>
<td>240</td>
<td>10</td>
</tr>
<tr>
<td>purple</td>
<td>70</td>
<td>80</td>
<td>-10</td>
</tr>
</tbody>
</table>

Chi-squared with 1df = 1.667. A deviation this large would occur by chance between 10 and 25 % of the time (actually 19.7%), so the Ho cannot be rejected.

For 13:3 phenotype observed expected ob-exp (ob-exp)^2/Ex

<table>
<thead>
<tr>
<th>progeny phenotype</th>
<th>observed</th>
<th>expected</th>
<th>ob-exp</th>
<th>(ob-exp)^2/Ex</th>
</tr>
</thead>
<tbody>
<tr>
<td>red</td>
<td>250</td>
<td>240</td>
<td>10</td>
<td>0.385</td>
</tr>
<tr>
<td>purple</td>
<td>70</td>
<td>80</td>
<td>-10</td>
<td>1.67</td>
</tr>
</tbody>
</table>

Chi-squared with 1df = 2.05. A deviation this large would occur by chance between 10 and 25 % of the time (actually 15.2%), so this Ho cannot be rejected either.
It is not OK to just say the one is better based on a slightly greater probability of chance occurrence; either model is compatible with the observations. You might be able to make a relative prediction using Bayes’ Theorem.

6. Mendel counted 10 F3 progeny of plants from selfing the F2 progeny of a TT X tt cross. Suppose that one tall F2 plant produced only 8 seeds and these all grew into tall progeny. What are the odds Mendel would have been wrong if he counted this as a TT plant? (You can show you answer as a formula without working it out if you prefer.)

\[
\frac{\binom{8}{1} \left(\frac{3}{4}\right)^8}{\frac{1}{3} \left(\frac{3}{4}\right)^8 + \frac{2}{3} \left(\frac{1}{4}\right)^8} = 16.66\%
\]

7. Using the legend R"R" red, R"R, roan, RR white, if a roan bull is mated to 8 roan cows:
   a) what is the probability that the progeny will include 3 red, 3 roan and 2 white animals?
   \[
   \frac{8!}{3!3!2!} \left(\frac{1}{4}\right)^3 \left(\frac{1}{2}\right)^3 \left(\frac{1}{4}\right)^2
   \]
   b) What is the probability that 4 calves will be solid color and 4 roan?
   \[
   \frac{8!}{4!4!} \left(\frac{1}{2}\right)^4 \left(\frac{1}{2}\right)^4
   \]
   c) What is the probability at least one calf will be white?
   \[
   1 - \left(\frac{3}{4}\right)^8
   \]

8. In maize, homozygous recessive ‘viviparous or vp’ mutants germinate while the seed is still on the cob. This is the result of their inability to make the plant hormone ABA. One vp mutant, vp5/vp5 occurs in a step in the pathway before carotene is made, so these plants
cannot protect chlorophyll and are albino after exposure to sunlight. (They are normally lethal, but can be grown to maturity in culture with sugar added.) The herbicide fluridone blocks the same desaturase step as the vp5 mutation. Another mutant, vp-6 catalyzes the last step for the conversion of carotene to ABA.

a) Which of the ‘extensions” to Mendel’s simple inheritance patterns are involved in this example? Explain how.

 Phenocopy; fluridone mimics vp5/vp5 effect
 Epistasis/ genetic heterogeneity the two genes in the same path, one can mask effect of the other.
 Peliotrophy- albinsim and vivipary cause more than one phenotype
 Lethal (recessive) in that vp5/vp5 will die in standard conditions

b) If a plant of the genotype Vp5/vp5, Vp6/vp6 was self pollinated, predict the ratio of:

i) Viviparous progeny 7/16

ii) progeny phenotypes considering both genes assuming random assortment

9 normal: 3 viviparous only : 3 albino + vivaperous

C) Now predict the ratios in testcross progeny of the double heterozygote if the genes are linked at 20 map units in:

 coupling  40 normal: 60 vivarous
 40 normal ;50 albino, vivarous : 10 vivparous only

 repulsion 10 normal : 90 vivparous
 10 normal : 40 vivparous only : 50 albino vivparous

9. Both humans and Drosophila have the XX/XY sex chromosome sex difference, yet they differ significantly as to how sex is determined and how gene dosage is compensated. Describe the kinds of observations that lead to this conclusion.

Sex chromosome abnormality differences:

<table>
<thead>
<tr>
<th>Drosophila (with two sets of autosomes)</th>
<th>Homo sapiens</th>
</tr>
</thead>
<tbody>
<tr>
<td>XXX</td>
<td>sterile female</td>
</tr>
<tr>
<td>XXY</td>
<td>fertile female</td>
</tr>
<tr>
<td>XO</td>
<td>male</td>
</tr>
</tbody>
</table>

Dosage: X chromosome inactivation in humans does not occur in Drosophila

10. a) The RBMX gene is on the X chromosome of both mammals and marsupials. Defects in the gene lead to abnormal splicing and a high risk of cancer if homozygous or
hemizygous. Predict the outcome of crosses of a heterozygous female to a normal male for both species.

Humans + XFY all daughters normal, though XFXr will vary in the amount of gene product and half of the sons will have a high risk of cancer

In marsupials where the male X is always the one inactivated, half of sons and daughters will be affected.

b) Genes da and abo are closely linked genes (2 cM) on chromosome II in Drosophila. Each is recessive to the wild-type normal allele. A male heterozygous for both genes in coupling is test-crossed to a homozygous recessive female.

1) Show the genotypes of each parent

\[
\begin{array}{c|c|c|c}
& da^+ & abo^+ & da & abo \\
\hline
\text{male} & da & abo & da & abo \\
\text{female} & & & & \\
\end{array}
\]

2) Predict the ratio in the progeny.

50 % like each parent (no XOs in male Drosohila)