A FEW REVIEW QUESTIONS DISCUSSED IN CLASS

The *putP* gene product transports proline and the toxic proline analog dehydroproline (DHP) into the cell. Thus, *putP* mutants are unable to use proline as a sole carbon source and are resistant to DHP on minimal medium with succinate as a carbon source. [Draw a cartoon showing what you would do and indicate the approximate number of bacteria or colonies expected at each step.]

- How could you select for *putP* mutants?
- How could you screen for *putP* mutants?

Although a suppressor mutation that restores functional interactions with a mutant protein may suppress some alleles of the protein much better that others, very few interaction suppressors are absolutely allele-specific.

- What is an allele specific suppressor?
- How would you test for an allele specific suppressor?
- Why are so few interaction suppressors absolutely allele specific?

Phage P1 efficiently infects and lyses both *galE* and *galU* mutants of *Salmonella typhimurium*, but not *gal*$^+$ strains. When a culture of a *galE62* mutant is infected with P1, most of the cells are killed but about 1 in 10^7 of the *S. typhimurium* cells is a Gal$^+$ revertant that is resistant to P1. Similarly, when a culture of a *galU14* mutant is infected with P1, about 1 in 10^7 of the *S. typhimurium* cells is a Gal$^+$ revertant that is resistant to P1. When a culture of a *galE62 galU14* double mutant is infected with P1, the reversion frequency is somewhat less — about 1 in 10^8 of the *S. typhimurium* cells is a Gal$^+$ revertant that is resistant to P1.

- If the reversion of the *galE62* and *galU14* mutations were independent events, what would be the predicted reversion frequency of the double mutant?
- The frequency of reversion of both the *galE62* and *galU14* mutants is greatly increased by the mutagen ICR-191 but not alkylating agents. What does this suggest about the nature of these mutations? Explain your answer.
- Given the above results, what is a likely reason that the frequency of double revertants is much higher than expected for two independent events?

Resistance to the toxic proline analog Azetidine-2-carboxylic acid can occur in two ways: (i) specific missense mutations in the *proB* gene (the first step in proline biosynthesis) which make it insensitive to feedback inhibition; and (ii) mutations that inactivate the *putP* gene (the permease which transports proline into the cell).

- Which class of mutants would you expect to be more common?
- Indicate whether each of the two types of mutation is dominant or recessive to the wild-type allele of that gene.

Describe a specific use for 3 of the following 5 types of mutations: missense mutations, nonsense mutations, frameshift mutations, deletion mutations, or insertion mutations.

Mutagenesis with UV light causes a variety of types of mutations, but the mutagen ICR181 specifically causes frameshift mutations. A large number of *putP* mutations were obtained with both mutagens. About 5% of the *putP* mutants obtained following UV mutagenesis had detectable CRM, while none of the mutants obtained following ICR181 mutagenesis had detectable CRM.

- What does the presence of CRM indicate?
- Suggest a likely explanation for these results.