**Week 10: Determine v2 Phenotype, Start Genotyping**

Learning Objectives for Promoter Discovery

*Skills*

* Convert DNA concentrations into volumes to pipet for desired amount of DNA

*Cognitive*

* Explain how dideoxy DNA sequencing is performed and analyzed

**Pre-Lab**

1) Watch 2 videos from list for week 10 lab
<<https://www.bio.davidson.edu/people/macampbell/113/2iterationsGGAstudent_S2024.html>>

2) Answer each of these four questions in two sentences or less.

A) How can you convert a DNA concentration into a volume needed to deliver 320 ng of DNA into a sequencing tube?

B) What is a dideoxy nucleotide? How does it differ from RNA and DNA nucleotides?

C) How many primers are used for DNA sequencing? Contrast your answer to the number of primers used in PCR.

D) What is a chromatogram that is produced during automated DNA sequencing?

**Information: Prepare Plasmids for Genotyping**

In Lab

1) Calculate how to set up the sequencing reactions using the Nanodrop Excel data and the Excel template file. The dilution calculation PDF file helps you focus on the task at hand, and asks you one bonus question. Your task is to set up 4 separate sequencing reactions for v2 X1, v2 X2, v2 X3 and v1 Xn. Record the pre-printed number on your 4 sequencing reaction tubes so next week you will know which sequencing results are yours.

2) Discuss how you can determine if the promoter you actually cloned matches the desired sequence that you sent off to the company to synthesize. What does it mean to “aligning multiple DNA sequences”?