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• prepare 500 ml each of cathode & anode buffers

- Anode: 50 mM Bis Tris

$$X (\text{1000 mM}) = (500 \text{ mL})(50 \text{ mM})$$

$$X = 25 \text{ mL}^{\text{Bis Tris}} + 475 \text{ mL H}_2\text{O}$$

- Cathode: 15 mM Bis Tris, 50 mM Tricine

$$X (1000 \text{ mM}) = (500 \text{ mL})(15 \text{ mM})$$

$$X = 7.5 \text{ mL Bis Tris}$$

$$X (1000 \text{ mM}) = (500 \text{ mL})(50 \text{ mM})$$

$$X = 25 \text{ mL Tricine}$$

$$+ 467.5 \text{ mL H}_2\text{O}$$

• need 3X dilution of Hao's "Blue Native Gel Dye" in all samples except ladder

↳ 6.6 μL each sample, 3.3 μL dye

• prep 15 μL of 1 mg/mL A2, then use 2-fold dilutions to decrease to 0.5 mg/mL and 0.25 mg/mL

$$\text{↳ } X (9.62 \text{ mg/mL}) = (15 \mu\text{L})(1 \text{ mg/mL})$$

$$X = 1.56 \mu\text{L} + 13.44 \mu\text{L H}_2\text{O PBS}$$

• start w/ 0.75 mg/mL B4, then dilute to 0.5, 0.25

$$\text{↳ } X (0.75 \text{ mg/mL}) = (15 \mu\text{L})(0.5 \text{ mg/mL})$$

$$X = 10 \mu\text{L} + 5 \mu\text{L H}_2\text{O PBS}$$

• load 8 μL / sample

• lanes:

1. ladder

2. A2 1 mg/mL

3. A2 0.5 mg/mL

4. A2 0.25 mg/mL

5. ladder

6. B4 0.75 mg/mL

7. B4 0.5 mg/mL

8. B4 0.25 mg/mL

9. ladder

• start gel 11 AM, 150V

B4 5/19/17
0.75 mg/mL
19.65 mM

A2 7/2/18
9.62 mg/mL
209 μM

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• after ~10 min, no sample is visually detectable in any wells

↳ perhaps the gel was oriented backwards

↳ repeat in opposite orientation

• this time load 9.5 μ l each

• also used newer "Native PAGE" gel, instead of "NuPAGE"

• new gel start 11:45 AM

↳ added 25 min to run