Questions Lab 10: PCR and electrophoresis

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BIO 1120 – Section 03

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Important Questions to Consider

WHAT WERE the unknown dyes you separated by electrophoresis?

The unknown dye I used was Dye #17. After electrophoresis, the dye migrated from 7 cm to 12 cm on the gel, meaning it traveled 3 cm. Since Amido Black moved 4 cm, the Rf value for the unknown dye is:

Rf=3cm / 4cm =0.75

After electrophoresis, the unknown dye appeared yellow. Comparing the Rf value and color to the known dyes, the most likely match is Congo Red, which has an Rf of 0.79 and is orange, though the final color appeared more yellow. It's also possible that the dye could be Rose Bengal (Rf = 0.72), depending on how the pH or gel conditions affected the observed color. Although Bismark Brown Y is yellow, its Rf value is –0.20, which does not match the observed Rf of 0.75. Therefore, Congo Red is the most likely identity for the unknown dye.

2. WHAT CAN YOU conclude about the charges and molecular weights of the dyes in your unknown relative to each other and to amido-black?

In electrophoresis, dyes that move farther are usually **smaller** or carry a **stronger negative charge**. In my case, I measured the distance my unknown dye (#17) traveled from the center well at 10 cm to where the bands were visible from 7 to 12. That means it spread 3 cm to the right and 2 cm to the left. The farthest distance was **3 cm**.

Amido Black traveled about **4 cm**, so:

**Rf = 3 ÷ 4 = 0.75**

Since my unknown dye moved **less distance** than Amido Black, we can conclude that it likely has a **slightly higher molecular weight** or a **weaker negative charge** compared to Amido Black. Its **yellow color** and **medium mobility** could also suggest a dye like **Bismark Brown Y**, although the Rf value doesn’t align perfectly with it.

In summary, the dye in my unknown (#17) is likely **larger or less negatively charged** than Amido Black, leading to its slightly **shorter migration distance**.

3. HOW MIGHT THE Rf values for the dyes you electrophoresed be different if the gel and buffer you used were run at pH 11 instead of 7? What if they were run at pH 3?

If pH was 11 (more basic), dyes might get more negative charge and move faster to positive side. If pH was 3 (more acidic), then more H⁺ ions would neutralize charges so dyes move less or slower. So pH change affect how far they go. At higher pH = more movement, at lower pH = less.

4. IF THERE WERE just 100 copies of each of your alleles at the D1S80 locus at the start of your PCR amplification, how many copies of each of those alleles would there be after 28 rounds of PCR amplification?

So it double every cycle. That’s 2²⁸ × 100 = 26,843,545,600 copies. Kinda crazy how it goes from 100 to like 26 billion!

A plastic box with colorful dots

AI-generated content may be incorrect.

