Forensic Biology & Bioinformatics II

1

General Lab Safety



- ABSOLUTELY NO FOOD OR DRINK OR PHONE. Water bottles INSIDE the bag.
- All **BAGS** need to be **HOOKED** or placed under the lab bench.
- **STOOLS** should be **TUCKED** in when you're standing- **TRIPPING HAZARD**.
- LONG HAIR needs to be TIED BACK.
- **CLOSED SHOES, GLOVES** and **GOGGLES** must e worn at all times.
- Discard the tips into the tip waste bin on your bench. Tip discard containers are for **TIPS ONLY!**
- REMEMBER TO PUT THE LID BACK ON WHEN YOU ARE DONE
- WASH HANDS with soap and water when finished!



Learning Objectives

- Forensics II
 - Explain the principles of DNA electrophoresis in STR analysis
 - Compare DNA profiles

Bioinformatics II

- Construct a phylogenetic tree using nucleotide sequences
- Use MEGA to align protein sequences

Part 1: Forensic Biology II

Gel Electrophoresis

A technique used to separate **DNA fragments** according to their size.



Influencing factors

1. Charge

DNA positively charged or negatively charged?





Influencing factors

- **2. Size:** Smaller molecules/fragments will travel faster and further
- **3. Density of gel:** The denser the gel is, the slower the particles will move. You want a gel that is dense enough to separate particles but that won't take too long.



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Before you start...

Do's:

- Wear gloves
- Wear safety goggles
- Tip discard containers are for TIPS and PCR tubes ONLY!
- Be careful working with the reagents!!

Don'ts:

• Do not use the same pipette tip for different solutions

DNA profile





5.6

5.6

5.6

5.6

16.0

5.4

5.4 5.4

5.4

6.0

6.0

6.0

6.0

16.0

What is DNA ladder?

Thermo Scientific[™] GeneRuler[™] 100 bp Plus DNA Ladder, ready-to-use, is designed for sizing and approximate quantification of wide range doubleagarose stranded DNA and on polyacrylamide gels. The ladder is composed of fourteen chromatographypurified individual DNA fragments (in base pairs): 3000, 2000, 1500, 1200, 1000, 900, 800, 700, 600, 500, 400, 300, 200, 100. It contains two reference bands (1000 and 500 bp) for easy orientation.

0.5 µg/lane, 8 cm length gel, 1X TBE, 5 V/cm, 1 h



Calculate size of bands.

Select the band Look at the bands on the ladder closer from top and bottom to the selected band.

Ex:

Band 1

500 bp – 2.3 cm

300 bp – 3.5 cm

Band 1 – 2.6 cm

"Rate of Movement Across Gel" between Ladder Bands

Rate = (Differences in band size) / (Differences in distance)

500bp -300 bp = 200 bp 3.5 - 2.3 = 1.2 cm

200bp/1.2cm = 166.7bp/cm





Rate of Movement Across Gel =

166.7bp/cm For BAND 1 (2.6 cm): Difference between upper ladder band and Sample band 1. 2.6-2.3 = 0.3 cm

Multiply this by the calculated rate 0.3 cm * 166.7 bp/cm = 50.1 bp 50.1 bp Difference between ladder band and Sample band 1.

Subtract difference from the ladder band size (bp) 500 - 50.1 bp = 449.99 bp



Sample Data for your assignment, in case your electrophoresis didn't work properly



Please note that the height of the gel that we used in the lab was 8 cm

TPA (100 and/or 400 bp) TPOX1 (216-264 bp) CSF1PO (287-331 bp)

Disposal



GELS – BIOHAZARD WASTE

TBE Buffer – Sink



Discard container on bench for TIPS ONLY!

Part 2: Bioinformatics II

Phylogenetic trees

A diagram that represents evolutionary relationships among organisms



The pattern of branching in a phylogenetic tree reflects how species or other groups evolved from a series of common ancestors.

Assignment

- Assignments must be typed and not hand written.
- 2 points are automatically deducted for late assignments, and 1 additional point is deducted for each additional day overdue.
- Both lab assignment 6 and 7 (under post lab on canvas) due before the beginning of lab 8.
- Additionally, pre-lab quiz for lab 8 is due by Monday 7:59am.



Biofuel Video Project

- Due date for video—Sunday, October 29 @ 11:59 pm
- Double check that video uploaded to YouTube actually is visible to others and plays!
- Don't wait until the last minute—uploading takes TIME
- Submit link via Peerceptiv (through Canvas)
- Due date for reviews—Sunday, November 5@ 11:59 pm
 - You **must** submit a video to be eligible to review.
 - You are graded on your review—don't just give all 5's for everyone—it WILL hurt your grade.
 - You **must** do three reviews. As you finish one, the next will be assigned.