GenoSensor

EduPrimer™ DNA Profiling Kit

Brief Protocol

EduPrimer™ DNA Profiling Kit is specifically designed for exposing novice students to PCR principles and technique. The kit is simple to use. The whole lab can be done within 3 hours.

DNA Preparation ~ 7 min

1. Add 200µL of **Solution A** to a 1.5mL microcentrifuge tube.
2. Thoroughly swab inside cheek with provided swab and put it into **Solution A**.
3. Vortex the sample for 10 seconds, then heat in pre-heated 95ºC heat block for 5 minutes
4. Briefly spin sample down in microcentrifuge. Remove the swab with tweezers.
5. Add 20µl **Solution B** to the sample tube. Vortex or invert to mix for at least 10 seconds.
6. Spin sample for 1 minute at 12,000rpm
7. Use 10 µl of supernatant as DNA template for PCR.

PCR Reaction Mixture ~ 5 min

Mix the following reagents into a standard PCR tube:

2X PCR Master Mix	10µl
Genomic DNA Template	10µl
Volume total =	20µl

PCR Parameters ~ 78 min

1. 94ºC – 2 minutes
2. 94ºC denaturing – 20 seconds}
3. 58ºC annealing – 20 seconds} **repeat steps 2, 3, & 4 for 40 cycles**
4. 72ºC extension – 20 seconds}
5. 72ºC – 5 minutes
6. 4ºC – finished / hold

Agarose Gel Electrophoresis ~30 min

- Pour 1% agarose gel, using your preferred staining method*.
- Use 10 µL of PCR product in each well to visualize on gel.
- Run at ~100V for 10-20 minutes and stop before loading dye has run off gel
- Visualize and record the results manually or by photography

  Larger expected band – **400bp** (Alu element inserted)

  Smaller expected band – **100bp** (no Alu insert)

*It's recommended that an in-gel stain visualized by UV light is used as they provide the sensitivity necessary for the smaller fragments amplified in this experiment.

Additional Required Materials

Thermal cycler, Heat block, Microcentrifuge, Micropipettes, Pipette tips, PCR tubes, Gel electrophoresis apparatus

(Full protocol for students also available on our website)