Gel Extraction

By Penn iGEM 2013 (Using QIAGEN information)

Goal: Run digested DNA on a gel to confirm and extract the DNA from that gel.

Protocol:

- 1) Create 0.8% Gel
 - a. Weigh out 0.4g agarose and add to beaker
 - b. Measure 50mL TAE and add to beaker
 - c. Microwave until solution clear
 - d. Add 5uL Syber Safe to solution
 - e. Pour into casting mold. Use large comb for gel extraction
- 2) Add 10uL loading dye to 50mL digest, mix well and load.
- 3) Run gel on 120 volts for 30 min
- 4) Extract gel slice with clean knife, be sure to cut close to avoid excess agarose
- 5) Weigh gel in colorless tube
 - a. Calculate volume (100uL=100mg)
- 6) Add 3 volumes of Buffer QG to 1 volume of gel
- 7) Incubate at 50 degrees C for 10 min, vortex every 3 min
- 8) Be sure solution is yellow before proceeding.
- 9) Add 1 gel volume of isopropanol, pipet up and down
- 10) Apply sample to QIAquick spin column
- 11) Add 0.75 mL PE, centrifuge for 1 min
- 12) Discard flow through, centrifuge again
- 13) Place column into clean 1.5mL microcentrifuge tube
- 14) Elute with 35uL of Eb
- 15) Nanodrop for yield