Purification of DNA Oligos from Gel

- Resuspend oligos in H₂O or TE buffer.
- 2. Pour Native-PAGE at appropriate concentration for size of oligos.
 - e.g. 20% Native PAGE for 15mer-30mer

Make PAGE gel with 0.5X TBE buffer \rightarrow ddH₂O, acrylamide, TBE buffer.

- *Does not need stacking gel.
- 3. Load samples with loading buffer without dye. Add one lane with dye at far right or left lane.
- 4. Run Gel in 0.5X TBE buffer at 150-400 V COLD. Gel heats up. Make sure buffer is enough to keep gel cold. Use cooling chamber or cold room.
- 5. Run to approximately 3/4 of the way to the bottom of gel. You can use minigel if available.
- 6. Remove gel. Place on Saran Wrap. Place on TLC Fluorescence Plate. UV shadow with short wavelength light.
- 7. Mark dark shadows. DNA shadows with UV light.
- 8. Cut out band (as small of a piece as possible). Cut gel slices into many small pieces. Place in 1.5 ml eppendorf tube.
- 9. Add 500μl 0.3 M NaAcetate (pH 4.8-5.2). Shake O/N at 37 °C in a vortexer.
- 10. Spin out gel slice (Save and redo if necessary). Phenol/CHCl₃ extract. Precipitate with ethanol and acetate. Wash 2X with 70% ethanol to remove salt.
- 11. Resuspend in sterile H₂O or TE buffer. (Use approx. 105µl)
- 12. Spec for concentration (use $5\mu l/500\mu l$ dilution 1:100)
- 13. Use primer in experiment.