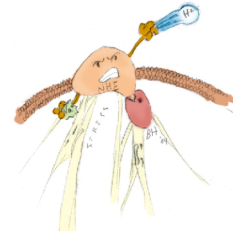


Quick and Dirty Plasmid Prep Protocol



Introduction: Only good for dirty quick preps of plasmids for storage or transfection.

1) Culture of bacteria for plasmid DNA isolation

- Expand a colony overnight hours in 5-7 ml of media with the appropriate antibiotic.

2) Prepare a modified alkaline lysis for storage.

- Spin down 5-7 ml of overnight culture for 10 minutes on high in 12 ml falcon tube or large test tube.
- Aspirate (remove) supernatant and resuspend cell pellet in 750 μ l Buffer P1 with RnaseA
- Add 750 μ l Buffer P2. Mix by inversion 4-6 times and incubate at room temp for 5 min.
- Add 750 μ l chilled Buffer P3, mix by inversion and incubate on ice for 15 min.
- Centrifuge for 30 min.
- Transfer and save supernatant into a clean tube. If particulate material remains, re-centrifuge or filter.
- Transfer 1.0 ml into two centrifuge tubes, and add 700 μ l isopropyl alcohol (IPA) to each tube.
- Mix by inversion and centrifuge at max speed for 30 min.
- Rinse pellet with 70 EtOH and re-centrifuge as above
- Let pellet dry for 10 - 20 min and resuspend pellet 50 - 75 μ l TE.

Subject 5 - 10 μ l of sample to 0.8% agarose gel electrophoresis for analysis.