Qiagen Miniprep

miniprep
scan for the full protocol

PROTOCOL

1. For each clone, **top-label a 1.5 mL tube** with the clone ID from the labsheet. This is the tube you will elute into.
2. **Pellet 2 mL** of the 4 mL saturated culture — fill a 2 mL tube. Spin 1 min, pour off the supernatant — **bleach it** before it goes down the drain. The buffer volumes are the same for anything from 1 to 5 mL.
3. **Resuspend** in **250 µL P1**. No clumps. The RNase A must already be in the P1.
4. **Lyse** with **250 µL P2**. Mix gently — **do not vortex**. It goes clear and viscous.
5. **Neutralise** with **350 µL N3**. Invert thoroughly. A white precipitate forms.
6. **Spin 5 min** at max speed to pellet the debris.
7. Transfer the **supernatant** to a blue QIAprep column. Spin **15 s**.
8. Add **500 µL PB**, spin. Add **750 µL PE**, spin.
9. Discard the flow-through and **spin 90 s to dry**. PE is 70% ethanol and carryover ruins everything downstream.
10. Move the column to your labelled tube. Add **50 µL EB** to the **centre of the membrane** and spin **45 s**.

– Label **the top and the side** of each tube — a tube marked only on the lid goes anonymous the moment someone puts the lids down.
– **A clone that yields no DNA was probably a satellite colony.** Note it and exclude it.