

Making Standards for Q-PCR (Qiagen)

1. Excise DNA from 1% TBE gel.
2. Weigh the gel.
3. Add 3X vol of QG buffer to gel weight
4. Incubate at 50° C for 10 min (until gel is completely dissolved)
5. Check the pH of DNA gel solution by color
6. Add 1 gel volume of isopropanol to sample & mix
7. Place a Q/A quick spin column in a 2 ml collection tube
8. Place DNA solution in QIA quick spin column, centrifuge for 1 min.
9. Discard flow thru
10. Add 0.5 ml of buffer QG to spin column, centrifuge 1 min.
11. Add 0.75 ml buffer PE, spin 1 min
12. Wait 1 min, then spin again at ~17,900xg to remove all residual EtOH
13. Place spin column in 1.5 ml tube
14. Add 50 µl EB buffer to center of spin column, spin for 1 min.
15. Let stand 1 min., spin again

Q-PCR

1:10 dilutions

A → B → C → D → E → F → G