

Protocol G: Preparation of reporter DNA

David Booth

Abstract

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Protocol

Use standard plasmid isolation kits to prepare >5 µg of reporter plasmids

Step 1.

- Qiagen Spin Miniprep Kit Cat No. 27104
- I usually do two minipreps for one nucleofection

Determine DNA concentration using UV-Vis spectrophotometry

Step 2.

- Quantify the DNA by UV-Vis spectrophotometry using the approximation of 1 OD at 260 nm = 50 ng/µl of DNA

Concentrate DNA to a final concentration of 5 µg/µl

Step 3.

- Use a standard ethanol precipitation with sodium acetate or speed-vac to concentrate DNA
- Be careful not to overdry DNA or else it will become insoluble. DNA from ethanol precipitation pellets tends to be more soluble.

Store at -20°C

Step 4.