## SANGER SEQUENCING



## Quick Guide







AGRF offers a full range of Sanger sequencing throughout Australia, with easy to use and reliable services with four points of entry depending on your specific project needs. All Sanger sequencing services are NATA accredited. You can submit your samples online and drop off at a local collection point, or send to AGRF via post.

## **PURIFIED TEMPLATE & PRIMER**

Prepare your samples by combining the following in a 1.5 mL Eppendorf tube with a snap-lock lid:

- Template DNA
  - Amount depends on template size
  - We recommend gel quantification to determine template concentration

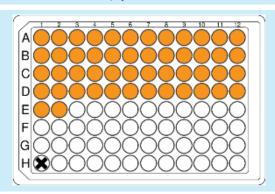
Template Size	Recommended Quantity
PCR product 100 - 200 bp	3 - 8 ng
PCR product 200 - 400 bp	6 - 12 ng
PCR product 400 - 600 bp	12- 18 ng
PCR product 600 - 800 bp	18 - 30 ng
PCR product > 800 bp	30 - 75 ng
Plasmid, single-stranded	150 - 300 ng
Plasmid, double-stranded	600 - 1500 ng

- Primer
  - 1μL from 10μM stock or 2μL from 5μM stock
  - One primer only per sample
  - Tm of 55 60°C
  - AGRF can add a primer for you refer to our Sanger Service Guide for available primers.
- Milli-Q water
  - Bring to final volume of 12 μL
  - $\circ$  If AGRF is to add a primer, bring to 9  $\mu$ L

Lid label:
ABC 001

Template DNA
+ primer +
Milli-Q water

- Labelling
  - Label on the tube lids and on side for long names
  - Where possible use simple naming on the tube lid, such as your initials and a sample number
  - If submitting > 12 tubes, number the tubes in sequential order for easy sorting
- Plates
  - Plate (95 samples): discount applies
  - Partial plate (47 to 94 samples): Partial plate pricing applies
  - Plate format (<47 samples): Tube pricing applies
  - Seal the plate using a heat seal and label on the side with your submission name
  - Fill every well of each row/column without skipping
  - Leave well H1 empty



- Submission
  - Log in to your account by visiting the AGRF website
  - > Log-in > Submit Your Samples
  - Complete the submission form
  - Print out submission receipt, include with your samples and send to AGRF

Contact us: 1300 247 301 CustomerCare@agrf.org.au www.agrf.org.au