

Transgenic Core Facility
Institute of Molecular Biology, Academia Sinica
2789-9312, 2652-1438

TCF PROTOCOL

DNA Quality and Quantity Required for TCF Gene Targeting Service

1. The quantity of DNA required for gene targeting service is at least 150µg dissolved in less than 400µl TE buffer (pH8.0).
2. Circular DNA should be reasonably fresh (no more than two months).
3. The quality of the DNA preparation should be Qiagen grade or better. (DNA preparation from Commercial Mini Kits is unqualified.)
4. DNA preparation need to be free of contaminant (mainly avoid of any possible toxicity). (CsCl₂ banding, commercial Midi and Maxi Kits are preferred. Never vacuum drying your DNA.)
5. DNA digestion should be fresh and complete.
6. Digested DNA should be kept at – 20°C and referred to TCF right away. The total volume of stocking restriction enzymes adding in DNA solution should be less than 10%. (Heat inactivation and DNA purification are strictly prohibited.)
7. TCF perform DNA extraction according TCF standard procedure to maintain consistency.
8. TCF will test DNA quality and quantity before scheduled electroporation. Should the submitted DNA be differ from TCF's specification, TCF will not proceed the scheduled service.