DNA Sample and primer Preparation for DNA sequencing

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1. DNA sample preparation:

   - Take 50µl of your Wizard miniprep DNA (***see Protocol for Wizard Plus minipreps***) and re-precipitate by ethanol and 7.5 M NH4OAc (***see Protocol for ethanol precipitation of DNA sample***).
   - Wash twice with 70% ethanol and dissolve the pallet into 10µl ddH2O (make sure you wash the pallets very well because even 10-20 mM salt can inhibit the sequencing reaction! The washing process can also remove some inhibitors). You need to check 1ul on an agarose gel.

2. Sequencing Primer preparation:

   - Provide at least 10µl sequencing primer (you will need 1-2ul per sequencing reaction) at the concentration of 30 ng/µl.
   - Too high concentration of primer might increase the background from non-specific priming.

3. Label each tube well and send for sequencing (along with a DNA Sequencing Request From; ask T.-C. for this form). The sample drop place for University of Chicago Cancer Research Center DNA Sequencing facility is located at Cummings 10th floor hallway. Put into the freezer and sign up the check-in sheet.

4. The turnaround time is usually 24hrs. Follow Protocol A38 to download and analyze the sequencing data.

Reference: http:// cancer-seqbase.uchicago.edu