Quick Sheet:
Generate a Consensus Sequence using SnapGene Viewer and NCBI

Edit the Forward Trace File

1. Open SnapGene Viewer.
2. Select Open >> Open Files >> YourFile_forward.ab1.
3. Select File >> Save As >> YourFile_forward.scf to create a copy of your raw data file.
4. Select "Show quality values" in the lower right hand corner.
5. Trim the 5'-end by identifying the contiguous sequence with ≥ 40 quality scores and highlighting ALL bases prior to this sequence. Hit 'Delete.'
6. Repeat for the 3'-end.
7. Scroll through the sequence. Are all quality scores ≥ 40? Adjust nucleotides to 'N' as needed.
8. Select File >> Export >> FASTA Format.

Edit the Reverse Trace File

9. Repeat Steps 1-8 with the reverse sequence.

Generate a Consensus Sequence

10. Open NCBI in your web browser: https://www.ncbi.nlm.nih.gov/
11. Select "BLAST" from the right-hand 'Popular Resources' menu
12. Select "Nucleotide BLAST."
13. *(optional)* Enter a Job Title.
14. Click " Align two or more sequences" at the bottom of the first box.
15. Load your forward FASTA file in the top box and the reverse FASTA file in the second box. Hit BLAST.
16. Check the % Identity. It should be 100%. If not, refer back to the trace files and investigate the discrepancy.
17. If your identity is 100%, select the Arrow next to "Download" and download FASTA (aligned sequences). Save. You have now generated a **Consensus Sequence**.