**Created by Yongtao Zhu (Spring 2021)**

**Protocol for PCR Using the Phusion High-Fidelity DNA Polymerase**

1. Label the PCR tube with the primer names
2. Assemble reagents in the following order:

* 25 µl Phusion 2X Master Mix
* 1.5 µl 10 µM Forward Primer
* 1.5 µl 10 µM Reverse Primer
* 1 µl genomic DNA (~100-200 ng/µl, lower concentrations should also work)
* 21 µl autoclaved ddH2O

Total volume in each tube should be 50 µl. Gently mix the reaction by flicking the tube (do not vortex). Collect all liquid at the bottom of the tube with a quick spin

1. Transfer PCR tubes to a thermocycler and begin the cycling program (Saved Protocols 🡪 ZHU LAB🡪 PHUSION). Change the annealing temperature and extension time if needed. Phusion works at 2 kb/min.