

Protocol for electroporation

EITHER

When you're doing a topo-isomerase ligation, dilute the salt solution with 1 part salt : 4 parts water

OR

After the ligation is complete, add 4 parts water to 1 part ligation product.

THEN

Pre-chill cuvet(s) on ice.

Make sure to have SOC ready for immediately after electroporation.

Turn on the electroporation machine. (The switch is on the right side of the machine.)

Add 2 uL of ligation to a 50 uL aliquot of cells. Flick the sides of the tube gently. (Cells are kept in the -80 in the basement.)

Pipette the entire cell + ligation mixture into the trough of the cuvette.

Wipe the sides of the cuvette with a kimwipe. Place the cuvette in the receptacle of the electroporation machine.

Make sure you're on the "Home Screen" of the machine. Choose the "Pre-set protocol" option (#4).

Choose bacterial.

Choose the first option -- 1 mm, E. coli.

Press the red button for a pulse.

Immediately after the pulse has completed (when the graph appears on the screen), add 250 uL of SOC broth to the cuvette. Pipette up and down. Move the cells and SOC from the cuvette back into the microcentrifuge tube. Incubate in the shaker, while shaking (200 RPM), at 37 degrees for an hour. Proceed with transformation as normal.