Electroporation of *E. coli* cells

- Thaw cells on ice (~5 min)
- For each transformation:
    - Chill one 0.5 ml tube and a clean electroporation cuvette on ice.
    - Add 30 µl of nanopure water and an appropriate amount (*) of DNA to each tube.
    - Add 25 µl of competent cells to each tube, mix by pipetting.
    - Transfer content to cuvette.
    - Before pulsing cells, tap cuvette to bring all liquid down, wipe metal sides with Kimwipe to remove water drops. Pulse with 1800 Volts. Time constant should be superior to 3.
    - Add 500 µl of LB to each cuvette, mix, aspirate out and transfer to a 14 ml Falcon tube. Let recover for 30 min at 37°C with shaking.
    - Plate on LB agar plate supplemented with appropriate antibiotic. Incubate overnight at 37°C.

Testing competent cells.

Use pUC18 as a standard. Keep a stock at 10 ng/µl, dilute down by one thousand-fold to 10 pg/µl before transformation and use 1 µl (10 picograms). Plate only one tenth of the final 556 µl. Count the next day.

(*) For regular plasmid transformation, one nanogram of DNA should give rise to about 100,000 transformants assuming an efficiency of 10^8 transformants per µg.

Note about ligations. Ligations contain salt. Salt will induce your electroporation to arc which will obliterate all your cells. At max, use 0.5 µl of your ligation mixture per transformation. If more is required, perform an ethanol precipitation followed by a rinse with 70% ethanol prior to transformation.

Washing cuvettes after use.
Cuvettes can almost be reused indefinitely provided they're well washed.
After use, flush hard with distilled water in the sink for at least 30 secs per cuvette.
Add 0.2 M HCl to each cuvette, let stand for 10 min.
Flush hard with distilled water again.
Add 100% ethanol, let stand for 10 min. Empty, dry out inverted on your bench, store.