Pico Green Quantification
This will quantify how much DNA is present in the sample so we can create equal molarities to send for sequencing.

This procedure ***HEAVILY*** relies on the two excel sheets, the procedure is broadly in there as well :

1. In the MATH sheet: Put in number of samples and how many repetitions of standards. It will tell you exact amount of TE needed and total volumes used
2. In the PicoGreen sheet: Where you will record the readings and who is where, it will create the standard curve and determine how much sample and DepC to add to get 5 nm of DNA

Method:
Bench Portion

- Fill in the # samples and how many sets of standards you are doing
- Make the total TE in RNase free conical tubes (dark green lid), invert to mix
- Subset the amount the amount of TE needed to dilute the PicoGreen in another RNase free conical tube
- Make the 3 mom standards in autoclaved 2/1.5 mL tubes (2, 0.2, and 0.02) LOOK AT EXCEL FOR AMOUNTS
    - Add TE first, then 2 uL lambda DNA to the 2, invert then centrifuge for quick pulse
    - Do serial dilution, invert then quick pulse between each (10 uL of 2 into 0.2, 10 uL of 0.2 into 0.02)
- Add TE to wells
    - Use multichannel for samples (they all get 49 uL)
    - Then do standards
- Create the baby standards in the wells (look at sheet, use mom standards to make 7 baby standards; 500, 250,100, 10, 4, 1, blank)
- Add 1 uL of sample the well (to ones that are getting sample)
- Add the PicoGreen to the TE we subset earlier, FOIL IT, invert to mix
- Use multichannel to add 50 uL of diluted PicoGreen to ALL wells
  ** these steps need to be done FAST as the PG is light sensitive
- Place foil lid, gentle vortex. Centrifuge for 1 minuet at 4400 rmp
  ** while this is happening, you can go turn on machine to heat bulb

Reading Plate

- Open plate reading program
- Select PicoGreen assay
- Select all then clear
- Remove foil and place plate in
- Run and record readings in the table on the excel (save to folder)
- Remove the plate, close machine, and turn off (plate in biohazard waste)