Update 1 October 2018 HJAM

Nanodrop Protocol

Purpose: Determine DNA quality and quantity.

Note: Always wear gloves when using the Nanodrop PC.

Protocol

I. Measure Samples:
   a. Open the ND-1000 program and select nucleic acid.
   b. Open arm and pipette 2ul of MilliQ water onto the spec.
      i. Pipette carefully into center of plate.
      ii. MilliQ water can be found on the shelf above the computer.
   c. Carefully lower arm and ensure drop of liquid in caught between top and bottom sensors.
      i. Do not drop the arm as this can damage the sensor.
   d. Click "OK" on the computer screen pop-up that instructs you to measure water.
   e. Wipe both sensors with kim-wipe.
   f. Measure a blank. This should be the buffer into which your sample was eluted.
      i. Pipette 2ul buffer onto spec.
      ii. Carefully close arm.
      iii. Visualize drop to ensure the drop was caught between the arm and the plate.
      iv. Click "Blank" on the screen (located in top ribbon).
   g. Wipe both sensors with kim-wipe.
   h. Measure your sample(s).
      i. Pipette 2ul sample onto spec.
      ii. Carefully close arm.
      iii. Visualize drop to ensure the drop was caught between the arm and the plate.
      iv. Type sample ID in right window.
      v. Click "Measure" on the screen (located in top ribbon).
   i. Wipe both sensors with kim-wipe.
   j. Repeat step 8-9 for remaining samples.
   k. When finished, pipetted 2uL MilliQ water onto the plate, wipe clean, and fold and place clean kim-wipe between sensors for next user.

II. Export Data:
   a. Click "Show Report" in top ribbon.
   b. Click Reports/ "Save Report."
      i. Select "Full Report."
   c. Save file in Dropbox/OGL Shared Files/General Lab/Common Lab Space/Equipment/Nanodrop
      i. Select OGL lots and create new lot folder.
      ii. *OR* if it is not an OGL lot, save in your personal folder in same location.
   d. Naming Protocol: Lot Number_tubenumbers_date_initials
      i. Ex: L00234_1-8_2-12-18_HJAM

III. Data Analysis:
   a. View your data output in Excel.
      i. Open Excel first, then open file.
      ii. Make sure to 'save as' your file, otherwise it will overwrite your .xls formulas or modifications and save it as a .ndv file.
   b. DNA is absorbed at A260, while any contaminates are absorbed at A280 (protein) and A230 (carbohydrate), which alter the 260/280 ratio and 260/230 ratio.

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    i. You want both of these ratios to be 1.8-2.2
c. Concentration is given and can be used to calculate normalized yield (using the weight of tissue extracted).
    i. (Concentration * elution volume)/extracted tissue weight
    ii. Make sure units are the same (i.e., ng/uL vs. mg of extracted tissue)