# ThermoFisher ND-1000 NanoDrop

1. **Overview:**
   a. The Thermo Scientific NanoDrop™ 1000 Spectrophotometer measures 1 ul samples with high accuracy and reproducibility. The full spectrum (220nm-750nm) spectrophotometer utilizes a patented sample retention technology that employs surface tension alone to hold the sample in place.
   b. The small sample requirement and ease of use make the NanoDrop 1000 Spectrophotometer ideally suited for measuring:
      - Nucleic acid concentration and purity of nucleic acid samples up to 3700 ng/ul (dsDNA) without dilution
      - Fluorescent dye labeling density of nucleic acid microarray samples
      - Purified protein analysis (A280) up to 100 mg/ml (BSA)
      - Expanded spectrum measurement and quantitation of fluorescent dye labeled proteins, conjugates, and metalloproteins
      - Bradford Assay analysis of protein
      - BCA Assay analysis of protein
      - Lowry Assay analysis of protein
      - Pierce Protein 660 nm Protein Assay
      - Cell density measurements
      - General UV-Vis spectrophotometry

2. **Training and Use:**
   a. Before using the NanoDrop, new users must be trained. Contact admin.tbep@utoronto.ca to arrange a training session.
   b. Book your timeslot in Google calendar under the account bookhere.equipment@gmail.com, and indicate your name and the lab to which you belong.

3. **Responsibility:**
   a. Labs are responsible for providing their own consumables (gloves, Kimwipes, etc.) for use with the NanoDrop.
   b. Users must properly clean the NanoDrop after use.

4. **Precautions:**
   a. **ALWAYS** wear gloves when touching the instrument.
   b. Keep stainless steel arm closed whenever possible; avoid dust contamination.
   c. Do **NOT** place a kimwipe between the pedestals when not using the Nanodrop.

**5. Procedure:**

a. Preparation:
- Quickly wipe the upper and lower pedestal with a Kimwipe soaked in RNAseZAP.
- Promptly rinse off the RNAseZap from the pedestals with a Kimwipe that has been soaked in water from the "Cleaning" bottle (Ultrapure or RNase/DNase-free dH2O).

b. Initialization:
- Choose "Nucleic Acid". Initialize the instrument by following the on-screen prompt to load a water sample (1.5-2 uL) from the "Blanking" water bottle (UltraPure or RNase/DNase-free dH2O). Click "OK".
- Once the initialization is complete, vigorously wipe the upper and lower pedestal dry with a clean Kimwipe. Between all subsequent measurements, repeat this cleaning.

c. Quantification:
- For RNA quantification, choose "RNA-40".
- Load a 1.5-2 uL buffer blank (typically water, you can use the "Blanking" water bottle). Click "Blank".
- Load a 1.5-2 uL buffer blank, click "Measure".
- Confirm that readings near 0 ng/uL are measured.

d. Measuring:
- Load 1.5-2 uL of your first sample. Enter a sample ID. Click "Measure".
- Repeat for all your samples, wiping the pedestal clean every time.

e. Data Export:
- Export your data by clicking "Show Report">"Reports" (top toolbar)>"Save Report…">"Export Report & Standards Tables". Save the report in the following directory: C:/nanodrop data/Reports/your username folder.

f. Cleaning and Maintenance:
- After you have measured your final sample, wipe the upper and lower pedestal dry with a clean Kimwipe. Apply 5 ul of "Cleaning" water onto each bottom pedestal.
- Lower the upper pedestal arm to form a liquid column; let it sit for approximately 2-3 minutes.
- Wipe both pedestals dry one more time with a new Kimwipe.
- NOTE: Typically dH20 is sufficient for removal of samples that have dried on the optical pedestals. There are a few cases (i.e. dried proteins) that may require a more rigorous cleaning protocol. For these cases, we recommend that 0.5M HCl be substituted for the 5 ul of dH20. Please follow an HCl application with 5 ul of dH20 to ensure any residual HCl is removed.

- **REMINDER:** Do **NOT** place a kimwipe between the pedestals when not using the Nanodrop. Leave the pedestal **CLOSED** after use.

## 6. References:

a. Refer to the ND-1000 User Manual for additional information.