Checking RNA-content of samples with the NANODROP

Principle

Once RNA was extracted from plant samples, it can be checked with the Nanodrop.

The Nanodrop is quantitative (expressing results as ng RNA / μ l sample) as well as pseudo-qualitative with the 260/280 and 260/230 ratios. The Nanodrop works following the same principle as classic spectrometers. Check the Nanodrop Manual on Nanaodrop.com for more information

(http://www.nanodrop.com/library/nd-1000-v3.7-users-manual-8.5x11.pdf).

Equipment

Reference (Company, Type,)
Thermo Scientific
Life Technologies AM9780

Procedure



Always wear gloves and work on clean surfaces.

Preparation

- Remove the RNA samples from the -20°C freezer and thaw them on ice.
- Flick & spin down
- Start up the ND1000 software (icon on desktop)
- Choose "Sample Type" -> "RNA-40"
- Clean the nanodrop electroptics with RNaseZAP

Calibration

- Pipette 1.5 µl RNase-free water on the electroptic
- Carefully close and click OK in the software.

Measurement



Wipe the electroptics clean after each sample

- Pipette 1.5 µl RNase-free water on the electroptic
- Carefully close and click "Blank" in the software.
- Wipe the electroptics clean
- Pipette 1.5 µl sample on the electroptic
- Carefully close, enter the sample ID and click "Measure" in the software.
- Repeat the process for all samples

Interpretation of the results

- Ratio 260/280 should be between 1.8 and 2.1 (<1.8 could be a problem)
- Ratio 260/230 should be between 1.8 and 2.2 (<1.8 could indicate a guanidine contamination)
- The results in RNA $ng.\mu l^{-1}$ are used to calculate the dilution to use when starting the cDNA production.