DNA Quantification at 260 nm Wavelength

MATERIAL

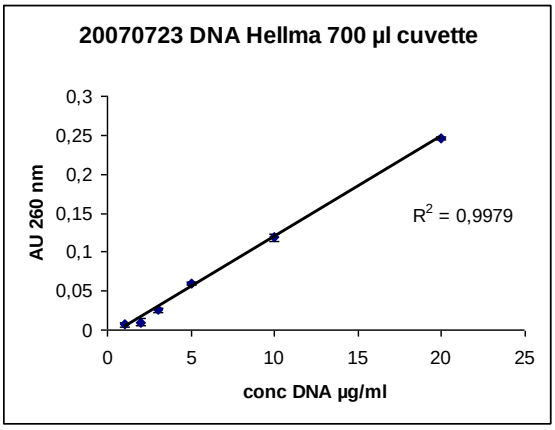
- DNA 1kb EZ ladder 50µg/250 µl (EZ labs, Taipei, Taiwan) diluted in ddH_20.
- Standard curve with concentrations 1 µg/ml, 2 µg/ml, 3 µg/ml, 5 µg/ml, 10 µg/ml, 20 µg/ml. Diluted in ddH20.
- SpectroArt 200, Protein Function, customised wavelength 260 nm, 6 standard curve points, read blank 2 times and samples 5 times.
- Hellma™ cuvettes: 5 µl TrayCell with 1 mm PL, 100 µl black wall cuvette 10 mm PL, 700 quartz cuvette 10 mm PL.
- ddH_20 for washing.
- Cotton buds for cleaning of TrayCell.

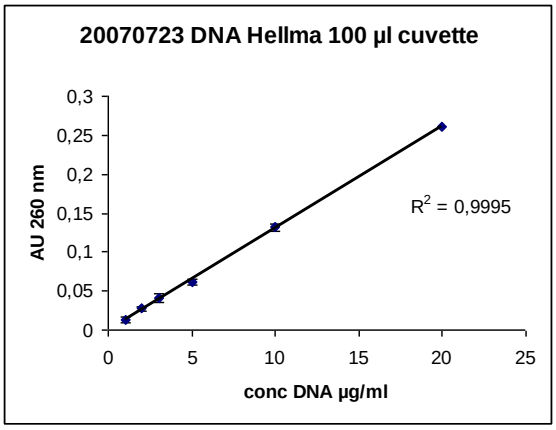
PROCEDURE

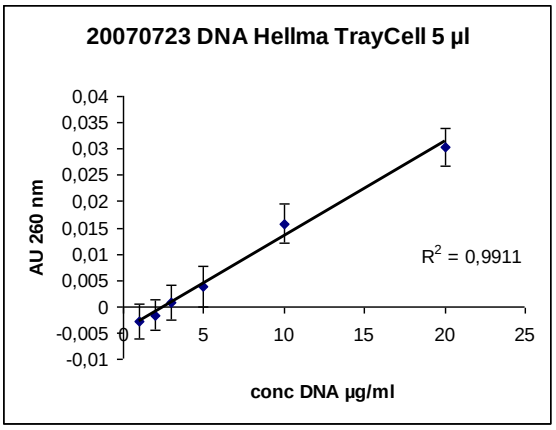
- **700 µl cuvette:** default flash and average settings were applied. Between runs and concentrations, the cuvette was removed from the spectrophotometer to be washed before new concentration liquid was filled into it. Washing was performed by first squirting ddH_20 into the cell using a flask with a long tube to build up enough pressure. The cell was then dabbed against dry tissues to remove excess fluid and then wiped clean using lint free Kimwipes. 600 µl was filled into the cuvette for measurements.
- **100 µl cuvette**: same procedure as for 700 µl cuvette. The 100 µl cuvette was filled with 150 µl for measurements.
- **TrayCell:** the cell was fitted properly into the spectrophotometer and flash/average settings were adjusted to maximum (8 flashes, 50 averages). The best obtainable position of the TrayCell was tested by using scanning function with air in the cuvette. The position that generated least noise was chosen and the TrayCell was kept in that position throughout the measurement series. After a measurement, the cap was removed and a cotton bud was used to collect the drop from the window and residue from the mirror. Then, a cotton bud was soaked in

ddH$_2$0, and used to wipe the mirror and the window. Another cotton bud was used to dry window/mirror clean and the procedure was then repeated with new water and a new bud. Finally, a new bud was used to wipe window/mirror dry and clean. Measurements were performed on 5 µl samples.

RESULTS







REMARKS

- Cleaning and position important for all measurements, but especially for TrayCell measurements.
- Position important for 100µl cuvette and TrayCell measurements
- No lint or fingerprints must be on the windows that are exposed to the light beam.
- Because of the virtual dilution factor of 10 for measurements with the TrayCell, the absorbance values obtained are approx. 10 times lower when measuring with the TrayCell. Very low concentration samples can therefore not be measured accurately with the TrayCell. A higher starting concentration is recommended for the absorbance values to reach above noise levels.
- Slowly empty the pipette into the 100 µl cuvette in order to avoid build-up of bubbles in cuvette window.

Tove Sivik
Application Manager
Email: info@wealtec.com

Wealtec BioScience Co. Ltd.
27Fl. No. 29-1 Sec.2, Jungjeng E. Rd., Danshuei Jen, Taipei, Taiwan 25170
TEL: +886-2-8809-8587 FAX: +886-2-8809-8589; http://www.wealtec.com