

Estimation of DNA by spectrophotometric method

Dr. Mahesha H B., Yuvaraja's College, Mysore.

Aim: To estimate the DNA using spectrophotometer.

Principle: The DNA absorbs radiation strongly in the UV region of spectrum due to the conjugated bonding system of the constituent purine and pyrimidines. They show characteristic maxima at 260 nm.

Requirments:

DNA Sample, spectrophotometer *etc.*,

Procedure:

1. Pipette out 3 ml of the given sample in cuvette
2. Measure the absorbance at 260 nm against distilled water/saline.
Theoretically one OD at 260 nm corresponds to 50 $\mu\text{g/ml}$ of dsDNA and it is 33 $\mu\text{g/ml}$ for ssDNA
3. Then from the following formula calculate the concentration of DNA in the given sample.

Result: The given unknown sample contains ---- μg DNA/ml.

Observations and Calculations

Optical density of the given DNA sample is ----- OD

The DNA concentration ($\mu\text{g/ml}$)= Absorbance at 260 X 50
