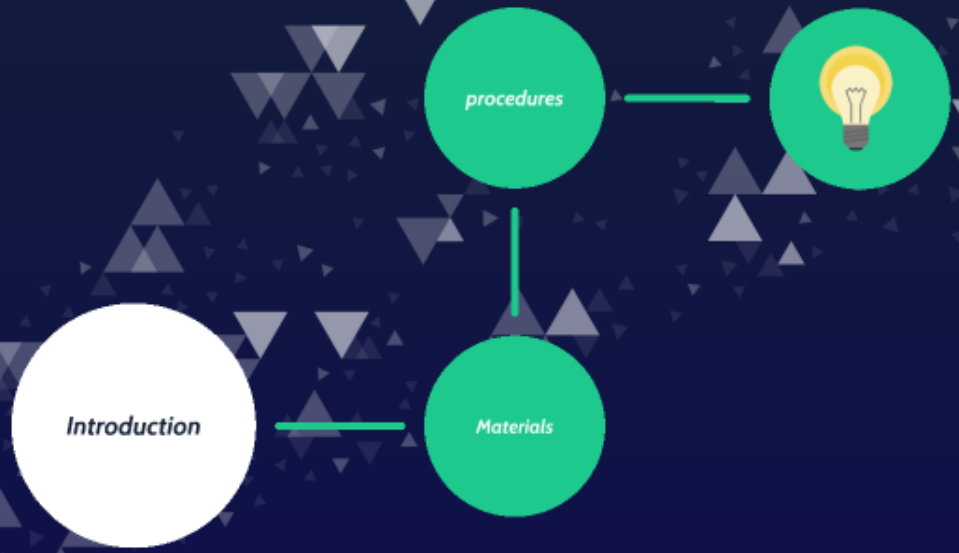
DNA Quantification

Prepared by:Rana Alqusumi



DNA Quantification

OBJECTIVE:

To quantify the amount of DNA in a genomic DNA sample.

Background:

1-The most commonly used method for nucleic acid measurement is **UV spectrophotometry**.

2-The nitrogenous bases in the nucleic acids have a maximum absorption at **260nm.**

3-Proteins have a maximum absorption at **280nm**

4-optical density(OD)=the amount of light that DNA has absorbed at 260nm

Equations

DNA concentration:

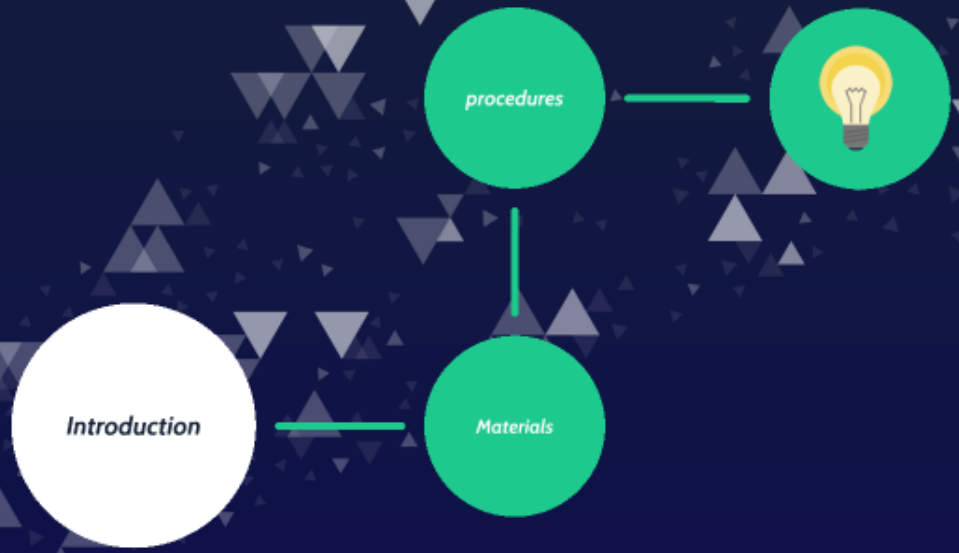
- DNA concentration (µg /µl) = OD at 260 nm × standard value.
- If OD260 is 1.00, it is equivalent to 50 µg DNA per ml (standard).

Purity of DNA:

- purity of DNA=OD at 260/OD at 280
- Pure DNA ratio between 1.6-2.0
- If the ratio less than 1.6 that indicates the present of protein contamination
- If the ration more than 2.0 that indicates the present of RNA contamination

DNA Quantification

Prepared by:Rana Alqusumi

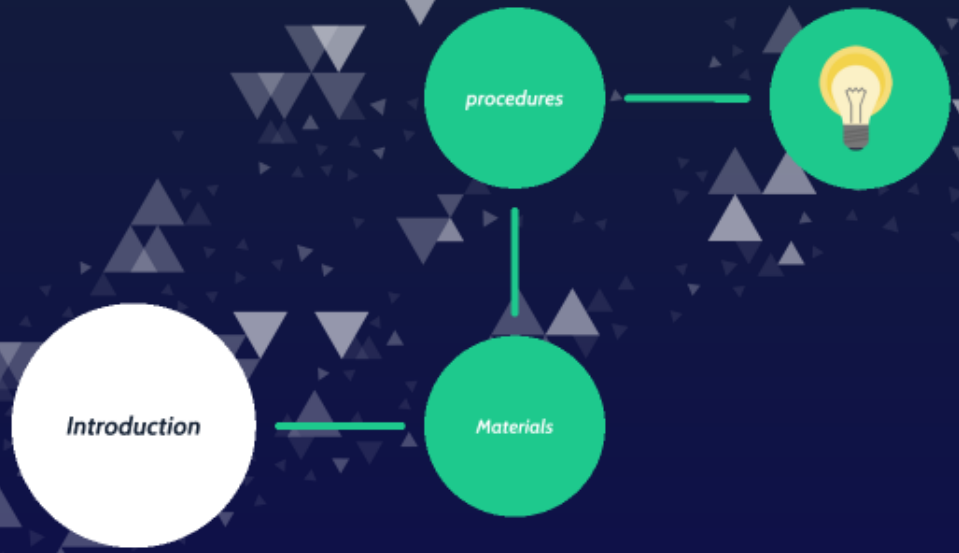


Materials

1-Sample to be measured.
2-Nano Drop
.3-P2 or P10 micropipettor with tips.
4-free lab wips
5-Purified water
6-Blanking solution (H2O)

DNA Quantification

Prepared by:Rana Alqusumi



Porceduers

1

2

3

4

5

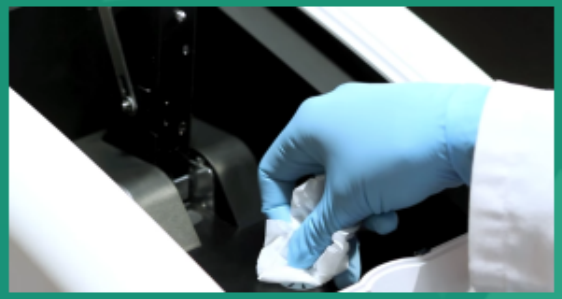
6

7

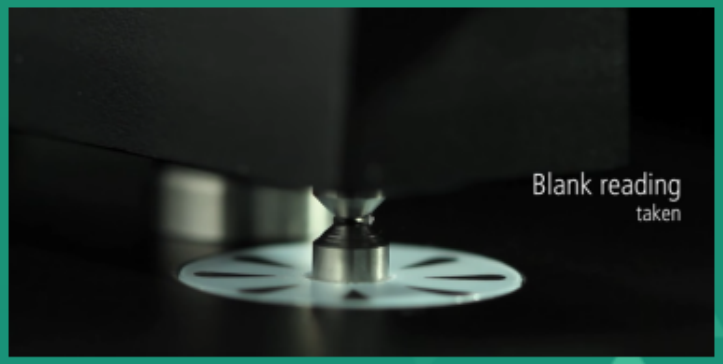
1.Select the appropriate application from the Home screen(DNAorRNA). For DNA measurements,select either the dsDNA or ssDNA assay.

2-Following the on-screen instructions,establish a blank by pipetting1-2µl of the blanking buffer onto the bottom pedestal, lower arm and press Blank.

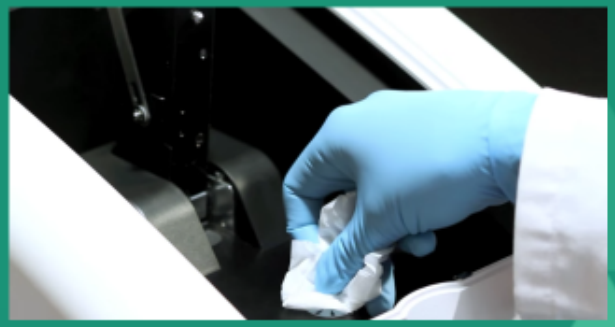
3.When measurement is complete,raise the arm and wipe the buffer from both the upper and lower pedestals using a dry laboratory wipe.



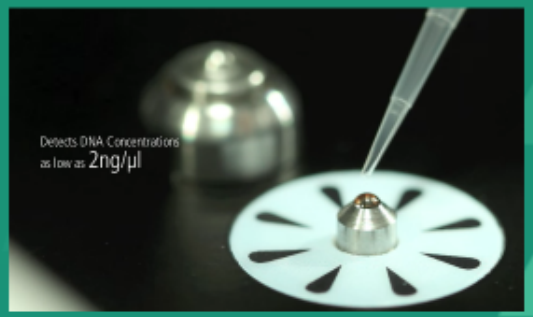
4-Confirm Blank by pipetting a fresh aliquot of blanking buffer onto the bottom pedestal, lower the arm and press Blank.



4-When measurement is complete,raise the arm and wipe the buffer from both the upper and lower pedestals using a dry laboratory wipe.



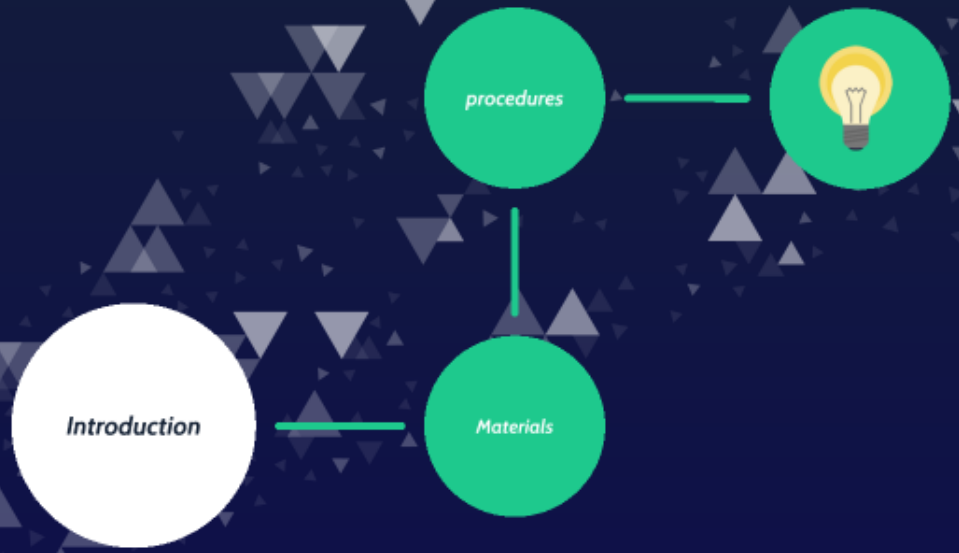
6.Measure sample by pipetting1-2µl of sample onto the bottom pedestal,lower arm and press Measure.



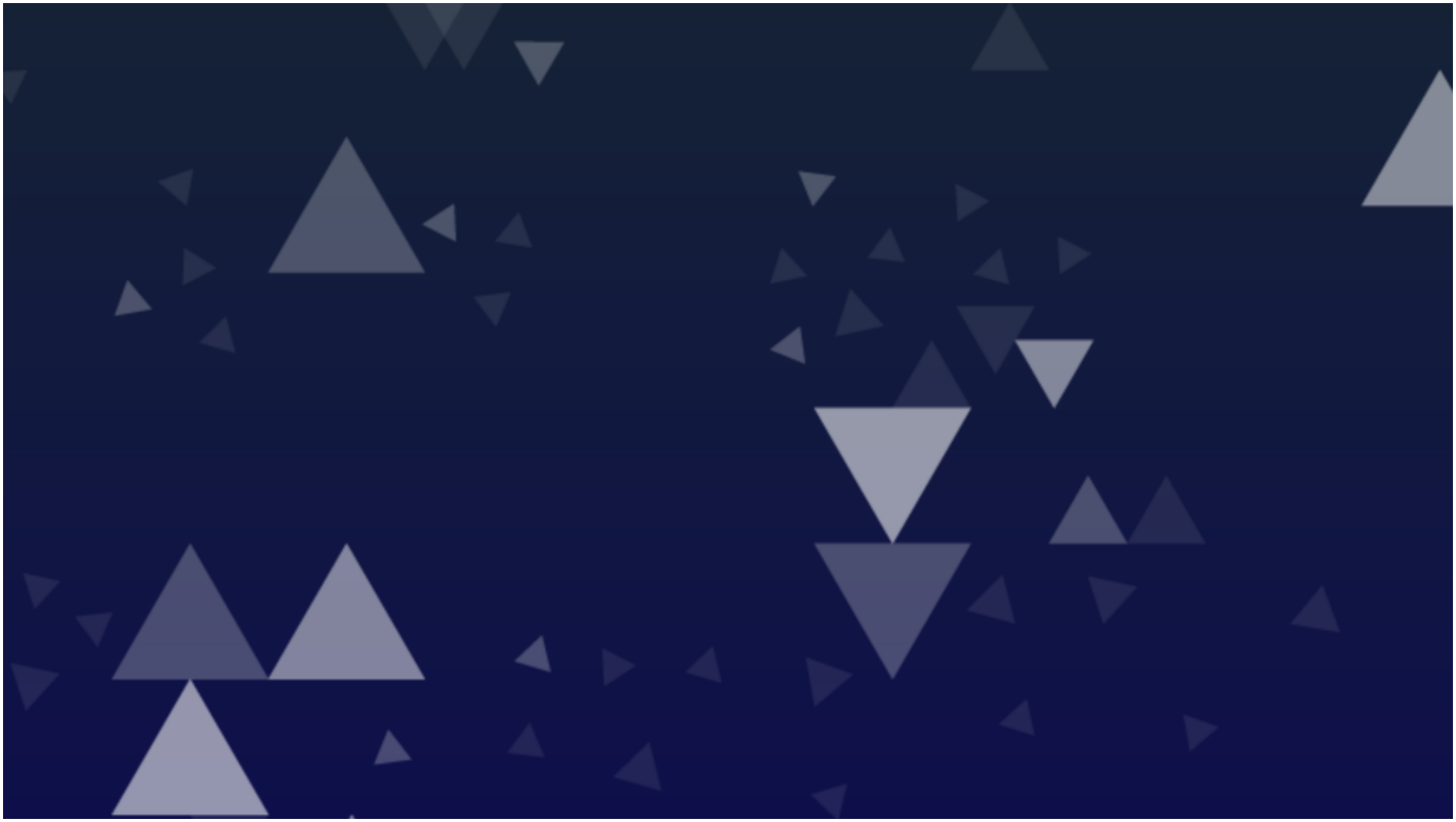

DNA
concentration
DNA purity
10.797
1.587
0.433

DNA Quantification

Prepared by:Rana Alqusumi




Losers quit when they fail.
Winners fail until they succeed.
Robert T. Kiyosaki



DNA Quantification

Prepared by:Rana Alqusumi