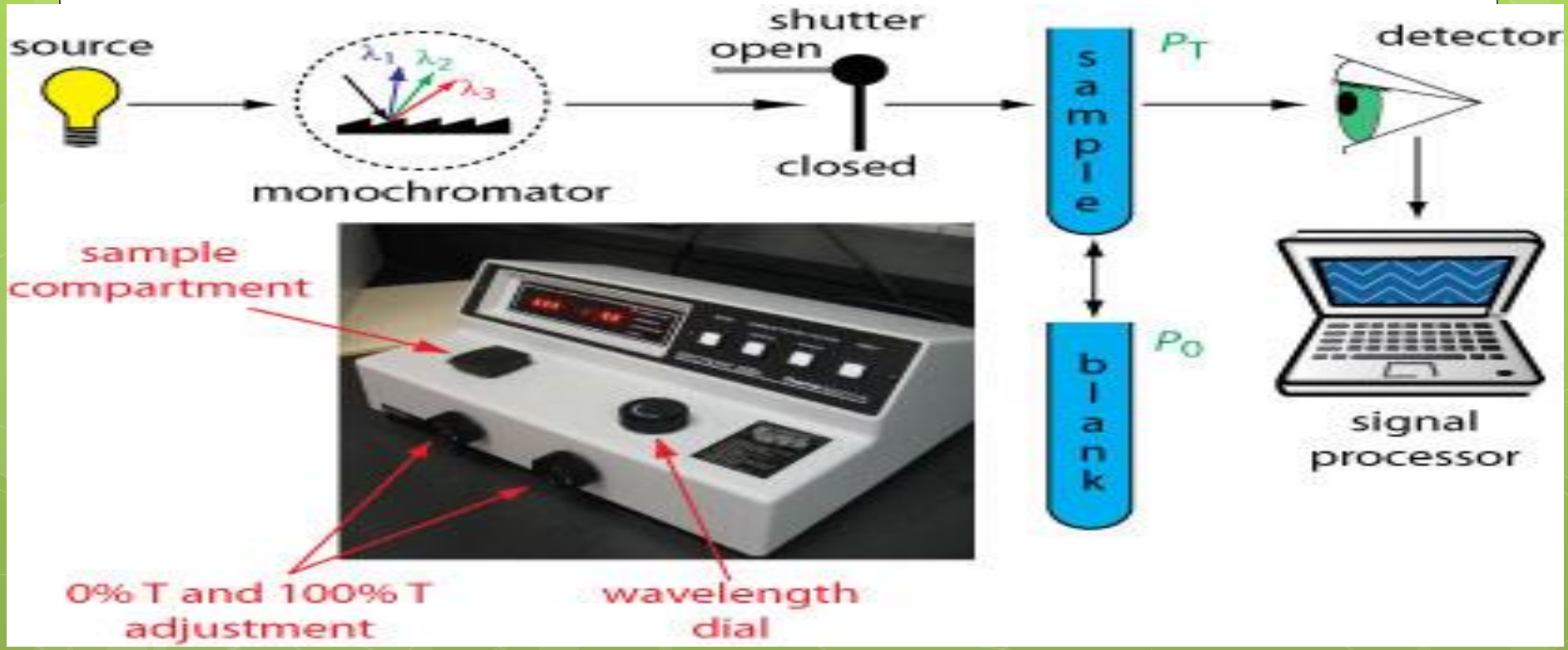
LAB 1 PART(2)practical part

preparation of Calibration curve of salicylic acid with the application of statistics



Wear gloves

Wear laboratory coats

Wear safety glasses

Wash hands

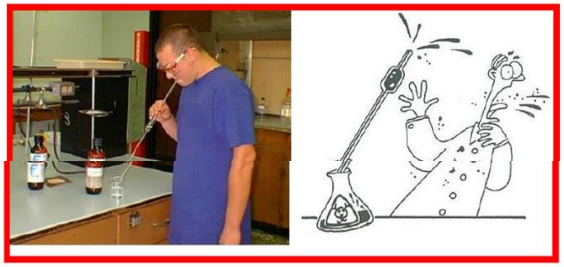
Tie back long hair

Wear closed-toe shoes
that cover the top of the foot

Do not wear dangling jewelry and long sleeves

Do not handle personal items cell phones, calculators, pens, pencils, etc.)

Do not mouth pipette



Do not bring food and drinks

Do not handle broken glass with fingers; use a dustpan and broom.

SPECTROPHOTOMETRIC METHOD

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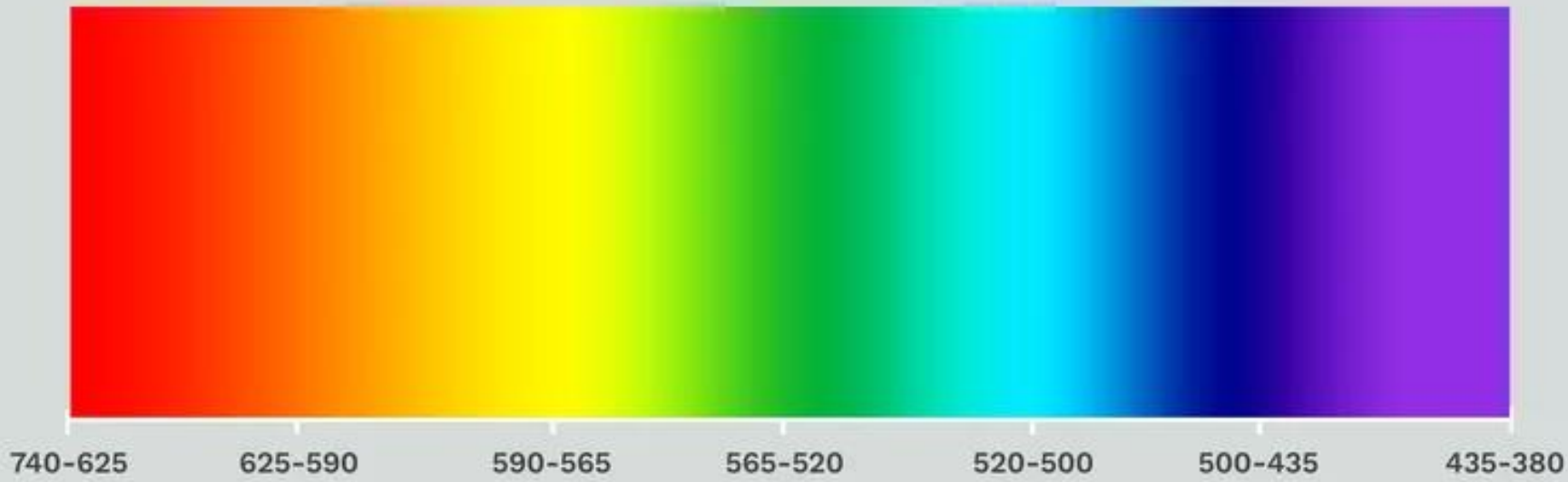
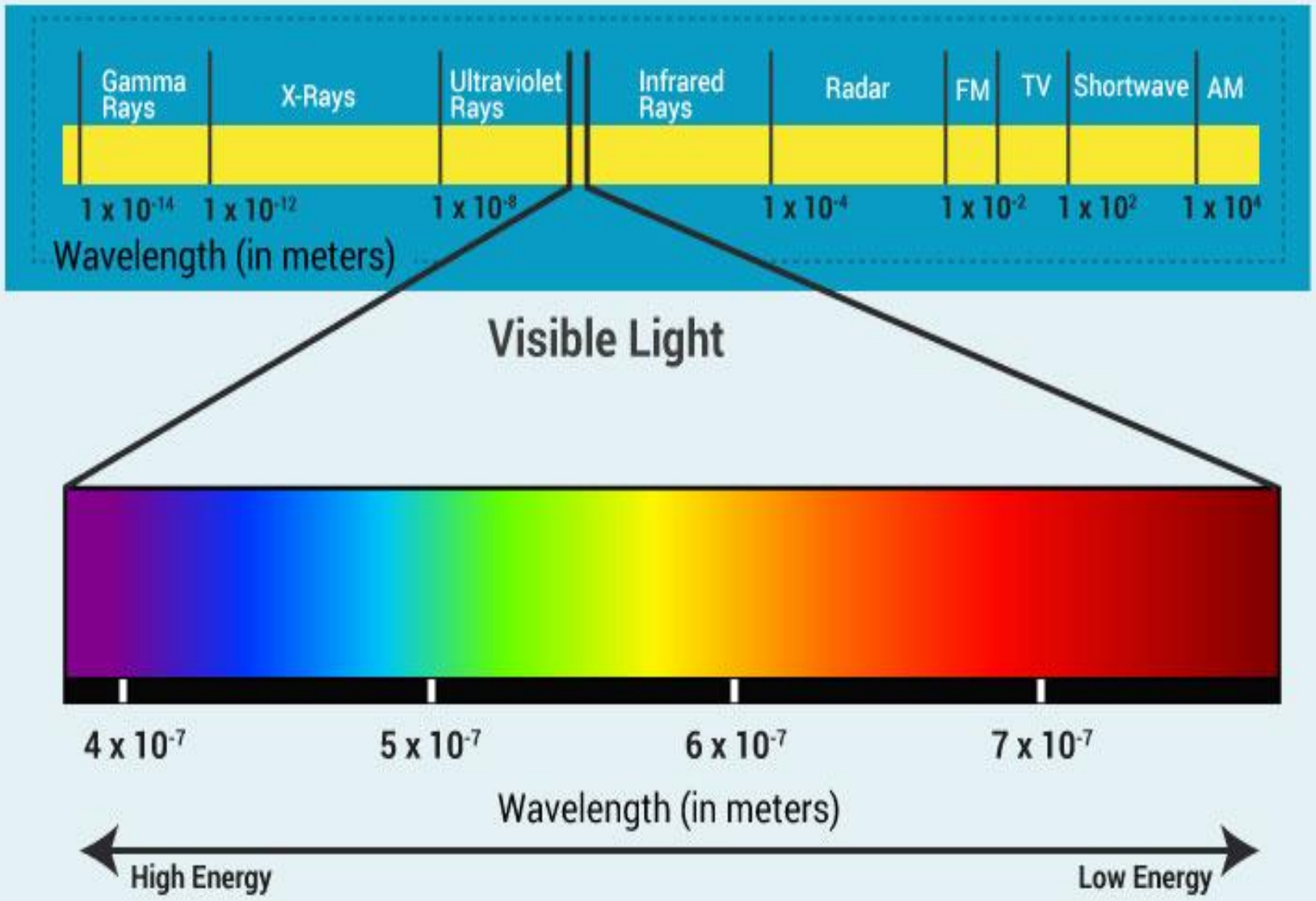
The Visible Light Spectrum

The visible light spectrum is the section of the electromagnetic radiation spectrum that is visible to the human eye.



ThoughtCo.

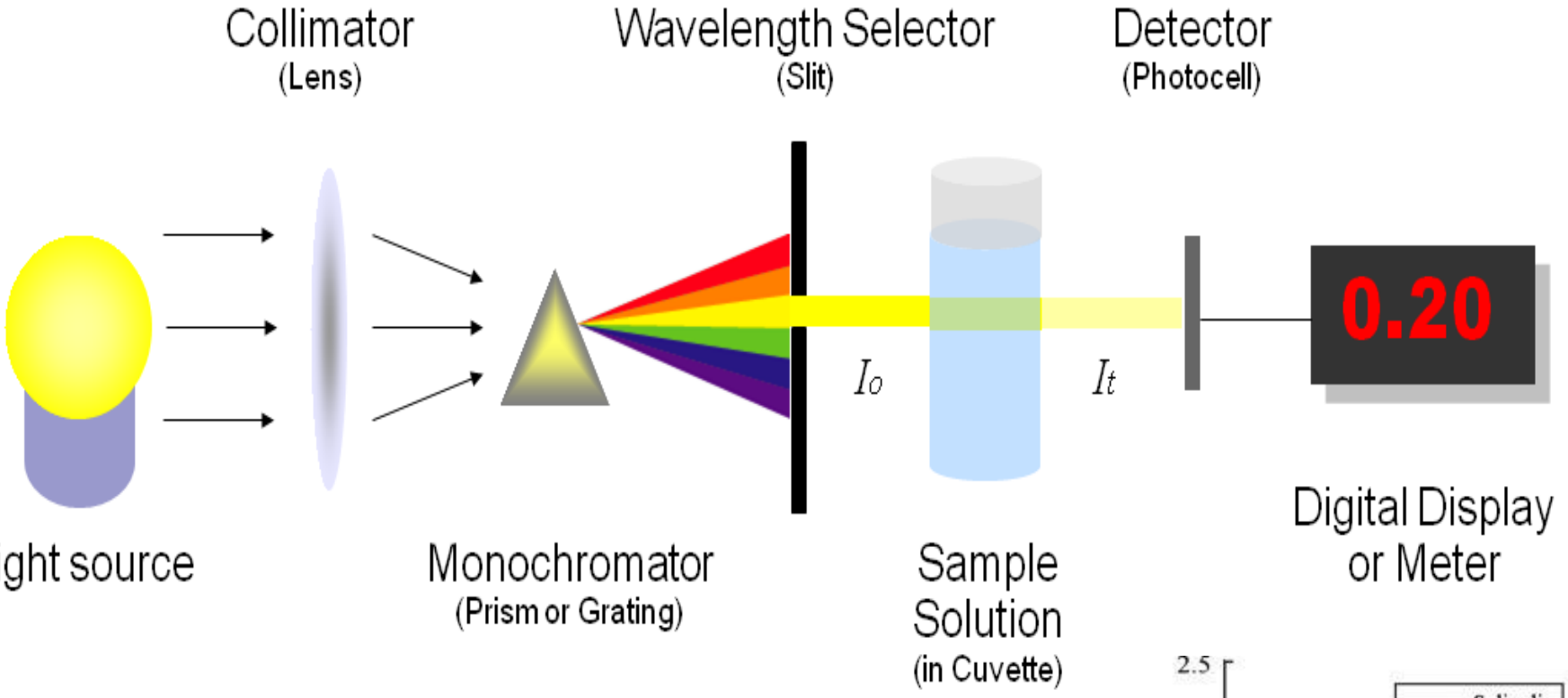

BYJU'S
The Learning App
Gamma Rays
X-Rays
Ultraviolet Rays
Infrared Rays
Radar
FM
TV
Shortwave
AM
1 x 10-14
1 x 10-12
1 x 10-8
1 x 10-4
1 x 10-2
1 x 102
1 x 104
Wavelength (in meters)
Visible Light
4 x 10-7
5 x 10-7
6 x 10-7
7 x 10-7
Wavelength (in meters)
High Energy
Low Energy

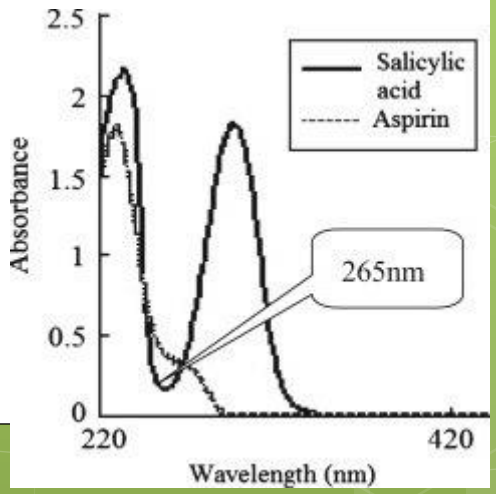
- In pharmaceutical spectrophotometric analysis

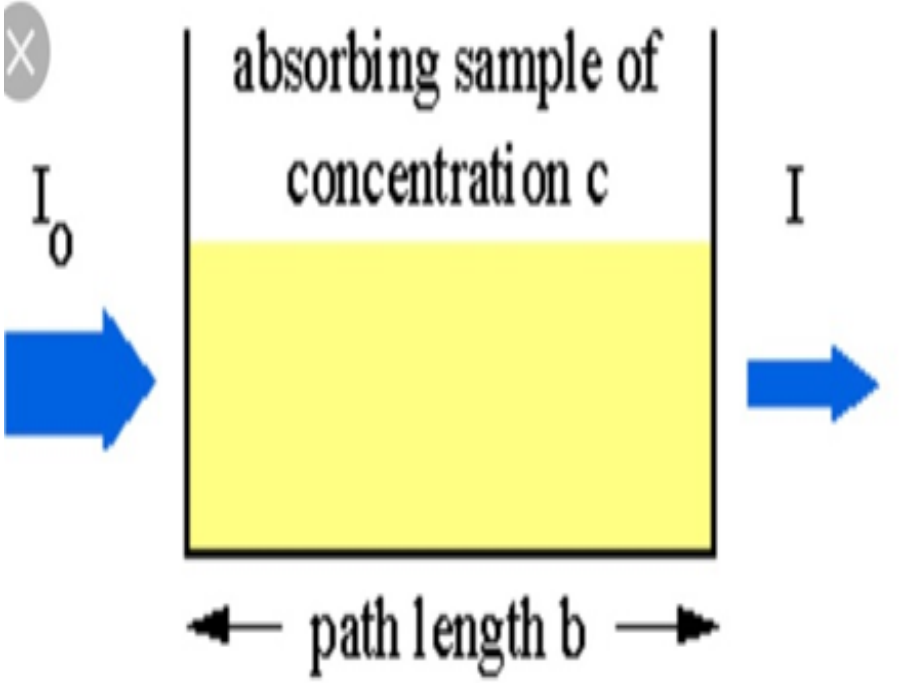
Visible light , of wave length 400-760 nm	Ultraviolet , between 200-400 nm

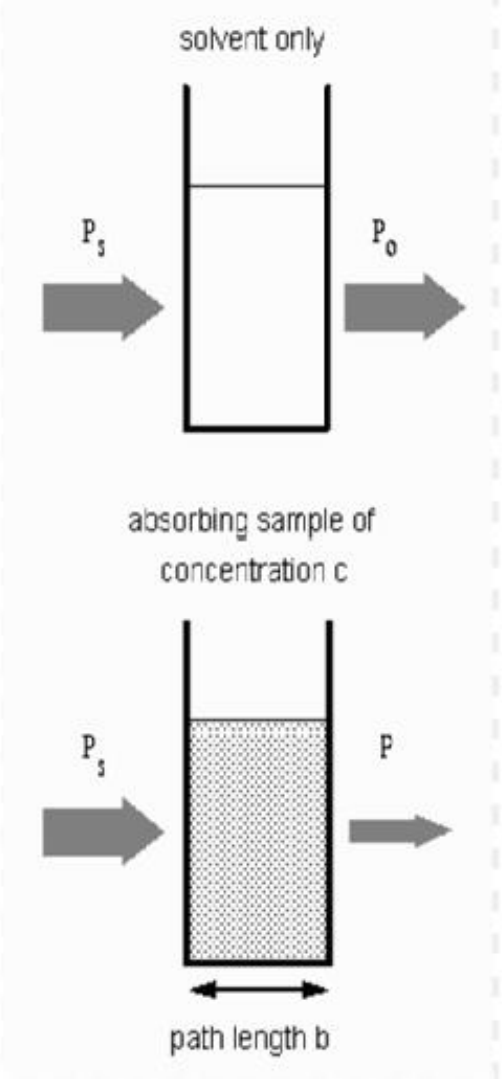
The measurement of the amount of visible or ultraviolet light of a definite wave length which is absorbed by a solution involve the use of spectrophotometer

SPECTROPHOTOMETER






absorbing sample of
concentration c
I_0
I
path length b


solvent only
P_s
P_o
absorbing sample of
concentration c
P_s
P
path length b

The fundamental relationship

- Used in spectrophotometry is **Beer** and **lambert** laws in combination:

$$\log \frac{I_o}{I_T} = Kct$$

where :

I_o : Incident light.

I_T: Transmitted light.

K: Proportionality constant.

c: Concentration.

t: thickness of substance.

$\log \frac{I_o}{I_T}$: E = Extinction or Absorbance.

$E = -\log_{10} T$.

$T = \text{Transmission} = \frac{I_T}{I_o}$.

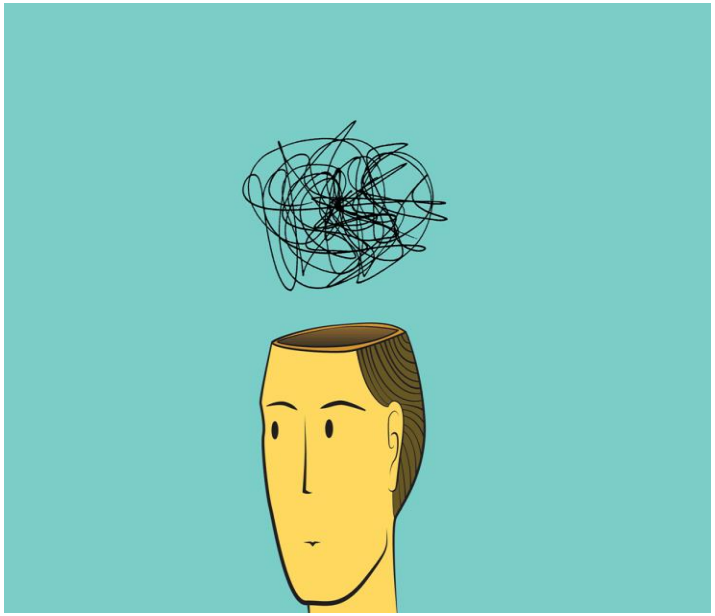
What is the aim of experiment ?

- The aim is to prepare a calibration curve of salicylic acid from a series of standard solutions **why?**
- to use it as a reference curve to obtain the concentration of unknown sample of the same drug .

Procedure and calculations:

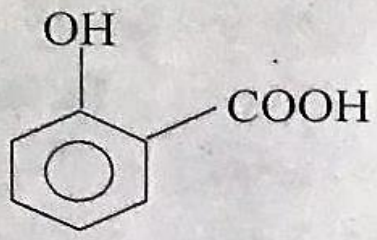
- 1. **prepare 250 ml of stock solution of sodium salicylate containing equivalent of 200 mg salicylic acid /100 ml**

- **Why sodium salicylate ?**

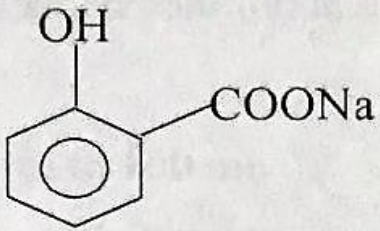


Why sodium salicylate ??

- **Note : sodium salicylate is readily soluble in cold water , so it is used to prepare the stock solution of sodium salicylate after calculating its equivalent content of the acid .**
- **while salicylic acid is sparingly soluble in cold water (1 part in 550 parts of water), but more soluble in hot water (1part acid in 15 parts of boiling water), from which it can be recrystallized**



Salicylic acid



Sodium salicylate

I- To calculate the number of (mgs) of (S.A) salicylic acid needed to prepare the stock solution:

mg of S.A	mls
200	100
x	250

x = 500 S.A.
needed = 0.5 g

II- To calculate the equivalent of sodium salicylate to 500 mg salicylic acid.

M. wt of sodium salicylate	M. wt of S.A
160	138
x	0.5

- **579.7 mg** of sodium salicylate needed to prepare 200 mg of salicylic acid /100ml

- 2- dissolve 579.7 mg of sod. Salicylate in 180 ml of water then complete the volume to 250 ml by using volumetric flask **(PRIMARY STOCK)**

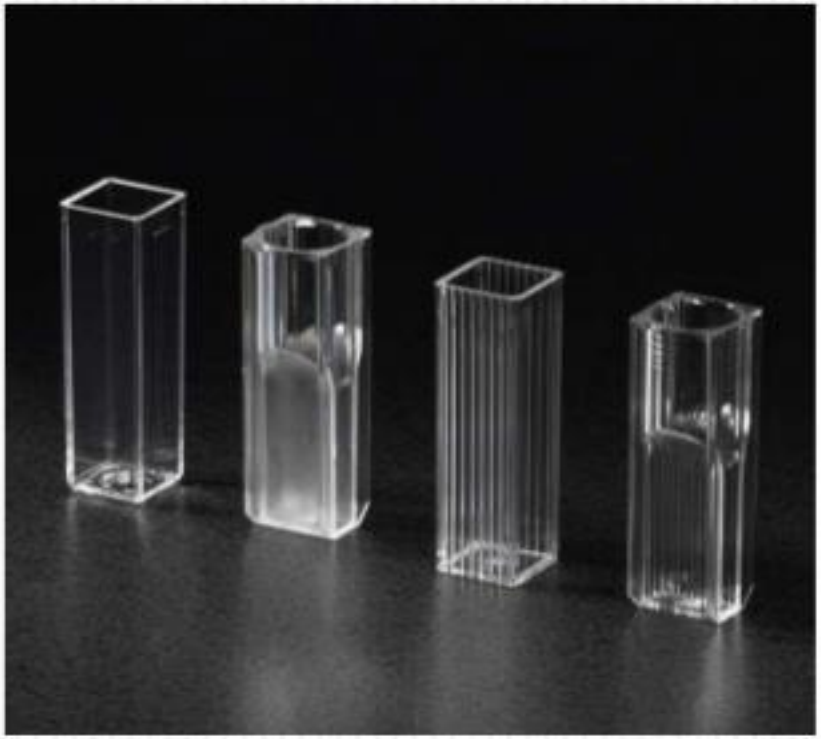
- 3- the **PRIMARY** stock solution of sodi salicylate containing equivalent of 200 mg\100 ml of salicylic acid

- From this primary stock prepare other stock solution 100 mg \100 ml by using
- CIVI=C2V2
- 200%X V1= 100%X100
- V1=50 ml of primary stock and complete the volume up to 100 ml d,w **(SECONDARY STOCK)**

- from this secondary stock Prepare serial dilutions (use volumetric procedures) accurately prepare solutions containing equivalent of 50, 40 ,30 , 20 , 10mg/ **10 ml**
- For example to prepare 50 mg\10 ml
- C1V1=C2 V2
- 100 mg x V1= 50 mgx10 ml
- V1 =5 ml of stock solution to be taken and complete the volume by d.w up to 10 ml
- Same procedure for other dilutions

- For 40 mg → 4 ml of stock complete up to 10 ml d.w
- For 30 mg → 3 ml of stock complete up to 10 ml d.w
- For 20 mg → 2 ml of stock complete up to 10 ml d.w
- For 10 mg → 1 ml of stock complete up to 10 ml d.w

- 4- set the spectrophotometer wave length at 265 nm(lamda max for salicylic acid)
- 5- the blank in this experiment is d.w
- 6- auto zero the spectrophotometer by using blank
- 7-put the sample in the cuvette and read absorbance



- 8 –record your results concentration versus absorbance
- 9- draw your results by using graph paper (make calculation using excel program)
- 9 – obtain straight line equation by using least square method
- 10- calculate y prime for each concentration
- 11 – draw y prime versus concentration by using excel Microsoft program
- 12- add trend line


I
have
a
Question!