Lab sheet #8

Determination of plasma amylase

Objective:

• To estimate the concentration of amylase in serum.

Method:

1. In a clean test tube add:

Chemical	Sample	
Amylase substrate	1.0 ml	
Pre-warm at 37°C for 5 minutes then add:		
Sample1	0.025 ml / 25µl	

- 2. Mix and incubate at **37°C for 90 seconds** and read the absorbance at **405 nm** against distilled water.
- 3. Continue readings every **30 seconds for 2 minutes** and determine $\Delta A/min$.

2) Applications → 2) Simple Kinetics → wave-length (405 nm) → 1) Seconds → Duration (120 sec = 2 min) → Intervals (30 sec) → Print Data Table (off) → Press start (2 times)

 \rightarrow The rate of **increase** in Ab is measured at 405nm and is <u>proportional to the amylase activity</u> in the sample.

-Results:

Time (Seconds)	Absorbance at 405 nm
0	
30	
60	
90	
120	

Calculations:

Amylase Activity in TEST (U/L) = $\Delta A/\min x \ 4824$

- 1- Δ **A/Min** = (Δ A1+ Δ A2) / 2 :
- $\Delta A1 = (A60s A30s) + (A30s A0s) = \dots$

 $\Delta A2 = (A120s - A90s) + (A90s - A60s) = \dots$

- $\Rightarrow \Delta A/Min = \dots$
 - 2- Amylase Activity in TEST (U/L)=