

## BCH 447- Lab sheet

### Acidic hydrolysis of glycogen

#### Method:

- 1- Label tubes (1 -9)
- 2- Pipette 0.4 ml PS buffer in a tube and use it as blank
- 3- Pipette 0.4 ml glycogen solution into each of the remaining tubes (1-9)
- 4- Add 0.6 ml 2 M HCl to all tubes
- 5- Incubate in boiling water bath in intervals of 4 min.
- 6- After the desired time, stop the reaction by adding 1 ml 1.2 M NaOH to neutralize HCl
- 7- Add 0.5 ml PS buffer to all tubes
- 8- Add 2 ml DNS reagent, cover with aluminum foil and heat all the tubes for 10 min. in a boiling water bath, the cool by immersing in cold water.
- 9- Add 5.5 ml water to each tube.
- 10- Read absorbance at 540 nm against the blank.

#### Result:

Tubes	Time (min)	Abs. at 540 nm	Hydrolysis %
Blank			
1	0		
2	4		
3	8		
4	12		
5	16		
6	20		
7	24		
8	28		
9	32		