PRACTICAL DATA INTERPRETATION IN BIOCHEMISTRY AND CELL BIOLOGY

These questions are designed for final-year undergraduate students (400 level) in Biological Sciences or related disciplines within the Nigerian university system, with an emphasis on laboratory data interpretation

Authored by

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Disclaimer

These questions are provided solely for practice, self-assessment, and academic skill development. They are not intended to replace official university examinations, continuous assessment tests, or instructor-designed course materials.

General Instructions
Answer all questions. Base your responses strictly on the data and information provided. Show relevant reasoning and calculations where required.

Mark Distribution

- Problem 1 (Inhibition of COX-2 by Plant Extracts): 25 marks
- Problem 2 (Effect of a Mutation on Enzyme Activity): 25 marks
- Problem 3 (Gene Expression Response to Environmental Stress**):** 25 mark

Total Marks: 75
Time Allowed: 2 hours 15 mins

Problem 1: Inhibition of COX-2 by Plant Extracts

Cyclooxygenase-2 (COX-2) is an inducible enzyme that catalyzes the conversion of arachidonic acid into prostaglandin precursors involved in inflammatory responses. A researcher investigates the effect of three plant extracts, A, B, and C, on the activity of COX-2 in vitro. The activity of the enzyme is measured at three different concentrations of each plant extract. The results are shown in the table below.

Extract	Concentration (µg/mL)	% COX-2 Inhibition
A	100	12
A	500	45
A	1000	72
B	100	8
B	500	28
B	1000	56
C	100	15
C	500	50
C	1000	55

Questions

1**(**a) Plot % inhibition vs. concentration for each extract.
(b) Estimate the IC_{50} for each extract.

2(a) Explain why Extract C shows a plateau in inhibition at higher concentrations.
(b) Compare the potency of Extracts A and B and justify your reasoning.

3**(**a) Suggest an appropriate negative control for this experiment.
(b) Propose an additional experiment to determine whether the inhibition is competitive or non-competitive.

4**.** Predict how changing the pH from 7.4 to 8.0 might affect the activity of Extract B and explain your reasoning.

Problem 2: Effect of a Mutation on Enzyme Activity

Lactate dehydrogenase (LDH) catalyzes the reversible conversion of pyruvate to lactate during cellular metabolism. A point mutation is introduced into the active site of LDH from *Plasmodium falciparum*. The activities of the wild-type (WT) and mutant (Mut) enzymes are measured at different pyruvate concentrations, and the enzyme kinetics data are shown below.

Enzyme	Substrate (mM)	Reaction Rate (µmol/min)
WT	1	12
WT	5	50
WT	10	85
Mut	1	5
Mut	5	20
MuT	10	30

Questions:

1**(**a) Plot reaction rate vs substrate concentration for WT and Mut.
(b) Estimate Vmax and Km for both enzymes.

2(a) How does the mutation affect substrate affinity?
(b) Suggest why Vmax is reduced in the mutant.

(3) Design an experiment to determine whether the mutation affects enzyme stability or catalytic efficiency.

(4) Predict how the mutant enzyme would respond to a competitive inhibitor compared to the WT enzyme and justify your reasoning.

Problem 3: Gene Expression Response to Environmental Stress

A researcher studies the expression of the heat shock protein gene Hsp70 in *Arabidopsis thaliana* under normal conditions (22°C) and heat stress (42°C) for 2 hours. Relative mRNA levels are measured by qPCR:

Condition	Relative Hsp 70 mRNA Level
22°C	1
42°C	12

Questions:

1**(**a) Calculate the fold change in Hsp70 expression under heat stress.
(b) Explain why this change occurs at the molecular level.

2(a) Which transcription factors are likely involved in heat stress response?
(b) How does Hsp70 help the cell survive heat stress?

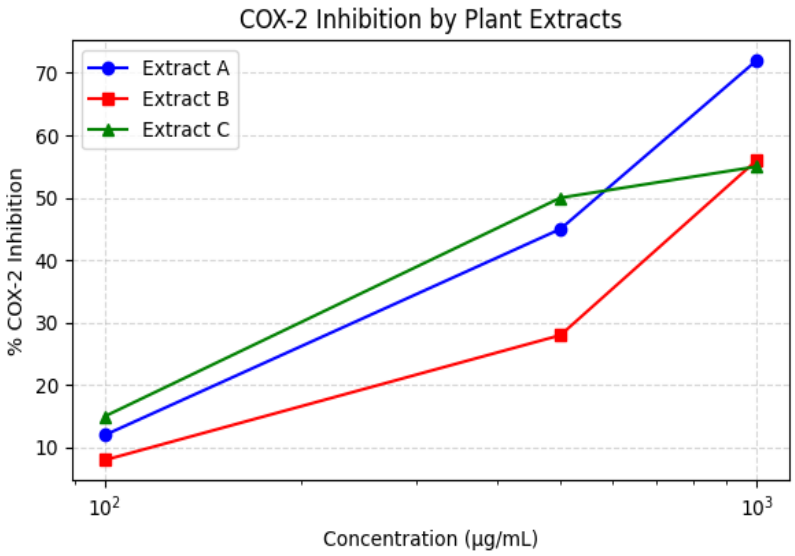
3(a) Suggest a method to test whether Hsp70 induction is directly due to heat shock transcription factors.
(b) Propose a control experiment for your method.

(4) Predict the effect of a small molecule inhibitor that blocks Hsp70 function on the plant's survival under heat stress.

ANSWER KEYS

Problem 1: Inhibition of COX-2 by Plant Extracts
Total: 25 marks

1(a) Plot % inhibition vs concentration for each extract
[6 marks]



Criterion	Marks
Correct axes (concentration on x-axis, % inhibition on y-axis)	2
Correct plotting of data points for Extract A	1
Correct plotting of data points for Extract B	1
Correct plotting of data points for Extract C	1
Clear legend / differentiation of extracts	1

Partial credit awarded if axes are correct but some points are misplaced

1(b) Estimate the IC50 for each extract
[4 marks]

Extract	Expected IC50 (approx.)	Marks
A	~450–500 µg/mL	1.5
B	~750–850 µg/mL	1.5
C	~450–500 µg/mL (lower efficacy)	1

Accept reasonable estimates based on graphical interpolation.

2(a) Explain why Extract C shows a plateau at higher concentrations
[4 marks]
Award marks for any two well-explained reasons:

Point	Marks
Saturation of enzyme binding sites	1
Presence of partial inhibitors	1
Antagonistic compounds in crude extract	1
Solubility or aggregation effects	1

2(b) Compare the potency of Extracts A and B and justify
[3 marks]

Criterion	Marks
Correct identification that Extract A is more potent	1
Justification using lower IC50 for A	1
Reference to higher inhibition at same concentrations	1

3(a) Suggest an appropriate negative control
[2 marks]

Answer	Marks
COX-2 + solvent only (no extract)	2
Vague or incomplete control	1

3(b) Propose an experiment to determine inhibition type
[4 marks]

Criterion	Marks
Use of multiple substrate concentrations	1
Comparison with and without inhibitor	1
Reference to kinetic analysis (Km and Vmax)	1
Correct interpretation (competitive vs non-competitive)	1

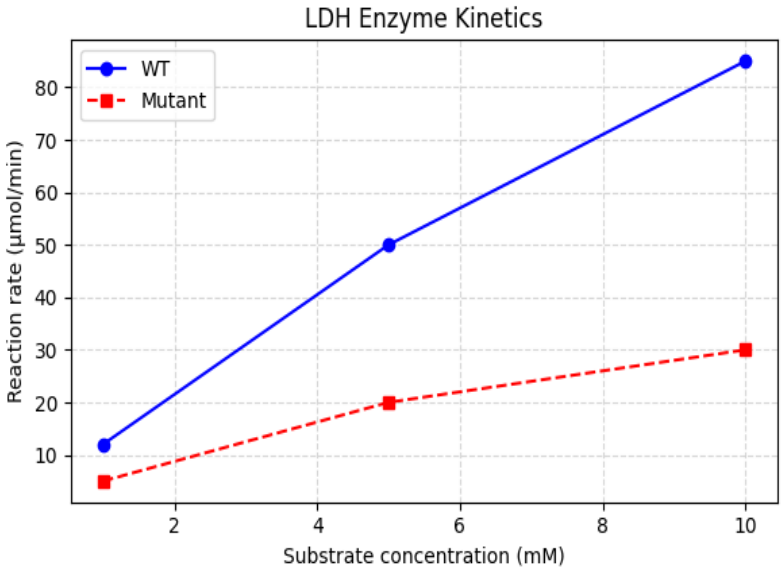
4. Effect of pH change on Extract B activity
[4 marks]

Criterion	Marks
Recognition that pH affects enzyme and inhibitor	1
Explanation involving ionization state / binding	1
Prediction of altered inhibition (increase or decrease)	1
Logical, biologically sound reasoning	1

Problem 2: Effect of a Mutation on Enzyme Activity

Total: 25 marks

1(a) Plot reaction rate vs substrate concentration for WT and Mut
[6 marks]



Criterion	Marks
Correct axes (substrate concentration on x-axis; reaction rate on y-axis)	2
Correct plotting of WT data points	2
Correct plotting of Mut data points	2

Partial credit awarded if axes are correct but some points are inaccurately plotted.

1(b) Estimate Vmax and Km for both enzymes
[6 marks]

Enzyme	Expected Vmax (approx.)	Expected Km (approx..)	Marks
WT	~90–100 µmol/min	~4–5 mM	3
Mut	~30–35 µmol/min	~6–8 mM	3

Allow ±10–15% variation. Partial credit if only one parameter is correctly estimated

2(a) Effect of mutation on substrate affinity
[3 marks]

Criterion	Marks
Correct inference that Km is increased in mutant	1
Interpretation that substrate affinity is reduced	1
Logical linkage to active-site mutation	1

2(b) Reason for reduced Vmax in the mutant
[3 marks}

Explanation	**Marks**
Reduced catalytic turnover (kcat)	1
Disruption of active-site geometry	1
Impaired transition-state stabilization	1
Partial enzyme misfolding	1

3. Experiment to test stability vs catalytic efficiency
[4 marks]

Criterion	**Marks**
Clear experimental design (e.g. temperature or time-course assay)	1
Comparison of WT and Mut under identical conditions	1
Measurement of activity decay or thermal denaturation	1
Clear distinction between stability and efficiency	1

4. Response of mutant to competitive inhibitor
[3 marks]

Criterion	**Marks**
Correct prediction (greater sensitivity or altered inhibition)	1
Explanation involving altered active site	1
Reference to Km or substrate competition	1

Problem 3: Gene Expression Response to Environmental Stress

Total: 25 marks

1(a) Calculate the fold change in Hsp70 expression under heat stress
[3 marks]

Criterion	**Marks**
Correct calculation (42°C / 22°C = 12 ÷ 1)	2
Correct expression of result as a fold change (12-fold increase)	1

1(b) Explain why this change occurs at the molecular level
[4 mark]
Award marks for any four relevant points:

Explanation	**Marks**
Heat causes protein denaturation/misfolding	1
Activation of heat shock transcription factors (HSFs)	1
Binding of HSFs to heat shock elements (HSEs)	1
Increased transcription of Hsp70 gene	1
Increased mRNA stability (if explained correctly)	1

2(a) Transcription factors involved in heat stress response
[3 marks]

Criterion	Marks
Identification of heat shock transcription factors (HSFs)	2
Correct biological context (stress-induced activation)	1

2(b) Role of Hsp70 in cell survival during heat stress
[4 marks]
Award marks for any four valid functions:

Function	Marks
Acts as a molecular chaperone	1
Prevents protein aggregation	1
Assists refolding of denatured proteins	1
Facilitates degradation of irreversibly damaged proteins	1
Maintains proteostasis under stress	1

3(a) Method to test whether Hsp70 induction is directly due to HSFs
[4 marks]

Criterion	Marks
Appropriate method suggested (e.g. ChIP-qPCR, reporter assay)	1
Direct assessment of HSF binding or activity	1
Application under heat stress conditions	1
Measurement of Hsp70 expression or promoter activity	1

3(b) Control experiment for the proposed method
[3 marks]

Control	Marks
Use of non-heat-stressed samples	1
Use of mutant or inactive transcription factor	1
Negative control promoter/antibody	1

4. Effect of an Hsp70 inhibitor on plant survival under heat stress
[4 marks]

Criterion	Marks
Prediction of reduced survival or increased damage	1
Explanation involving protein misfolding	1
Reference to loss of chaperone activity	1
Logical, biologically consistent reasoning	1