AS 3 ENZYMES

ANSWERS & MARK SCHEMES

QUESTIONSHEET 1

(a) hydrogen peroxide; 1 (b) (i) to release catalase from within liver cells; 1 Any 2 of: same weight of liver/same species/animal for liver/same volume of buffer at same pH/keep cool or at 4°C 2 /homogenise in blender for same time;; (c) vary temperature of water bath/incubator; 10° C intervals over suitable range (eg. $0 - 70^{\circ}$ C); same standard homogenate of liver in each case; measure volume of oxygen produced in a standard time/in 1 minute; repeat experiment at each temperature and calculate means; allow acclimatisation time at each temperature before adding peroxide; max 5 (d) Any two of: volume of oxygen is affected by changes in temperature/pressure/ concentration of catalase in different liver extracts may vary/ some oxygen may be retained in conical flask by frothing;; 2 TOTAL 11 **QUESTIONSHEET 2** (a) amylase; reject 'pancreatic amylase' 1 (b) (i) acts as cofactor/allosteric effector; binds to the enzyme changing its molecular shape; so that the active site becomes operative/can bind with substrate; max 2 (ii) pH too low/acid; 2 enzyme is denatured; (c) Enzyme = maltose; Juice = intestinal/succus entericus; Product = glucose; 3 TOTAL 8 **QUESTIONSHEET 3** substrate concentration is the limiting factor; (a) (i) some active sites free; (thus) increase in substrate concentration can increase rate; 3 number of active sites is limiting factor/all available active sites occupied with substrate; number of substrate molecules exceed number of active sites; (thus) increase in substrate concentration cannot increase rate; 3 (b) enzymes have an optimum pH (at which they work at maximum rate); pH changes from optimum cause changes in shape/charges/ionisation state of active site; therefore reduces number of enzyme-substrate complexes; (and thus) reduce the rate; 4 TOTAL 10

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QUESTIONSHEET 4

(a) a specific series of linked reactions; each step of which is catalysed by a specific enzyme;	
named example/glycolysis/Krebs/any other example;	max 2
(b) removes CO ₂ from (carboxylic) acid group/decarboxylation;	1
(c) modifies/changes enzyme so that it can function/acts as a cofactor; (reject 'gives energy')	1
(d) once a certain amount of adrenaline/nor-adrenalin has been produced; nor-adrenaline inhibits conversion of tyrosine to dopa/feedback inhibition/end product inhibition;	2
(e) adrenal medulla/sympathetic nerve synapses;	1
	TOTAL 7
QUESTIONSHEET 5	
(a) substance/chemical which is structurally similar to normal substrate;	
and competes with it for active site of enzyme;	2
(b) number of substrate molecules exceeds number of molecules of inhibitor/substrate molecules occupy mo	st
/many of the active sites; therefore little inhibition/inhibition reversed by high substrate concentration;	2
(c) competitive inhibitors (CIs) structurally similar to substrate, non-competitive inhibitors (NCIs) not similar;	,
CIs bind to active site, NCIs bind elsewhere on enzyme; effect of NCI is permanent/cannot be reversed by increasing substrate concentration whereas Cl effect is	s temporary/can be
reversed by high substrate concentration;	3
	TOTAL 7
QUESTIONSHEET 6	
(a) (i) X is a protease/peptidase/splits peptide bonds/hydrolyses peptide bonds;	
causes inactive enzyme precursor to change shape/conformation; produces active/catalytic form/exposes active sites;	3
	Č
(ii) precursor fits into complementary active site of enzyme X;credit reference to specificity;	2
(iii) metabolic control/control of reaction rates/prevent cell lysis/protection of gut wall;	1
(b) either: pepsinogen; HCl;	
or: trypsinogen; enterokinase; (accept any correct example)	max 2
(accept any correct example)	
	TOTAL 8

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QUESTIONSHEET 7

~	
(a) raise temperature of water in waterbath to 65°C (checking with thermometer); maintain by adding hot water/thermostatic control; keep sucrase and sucrose in separate tubes until both solutions are 65°C/equilibrate for at least 5 minutes; add sucrase to sucrose and mix gently;	; 4
(b) forms a brick-red precipitate with reducing sugars/glucose/fructose; sucrose is a non-reducing sugar/does not react with Benedicts reagent; but if sucrose is digested (by sucrase) will give a positive Benedicts test;	max 2
(c) (i) reaction fastest at 40°C; because higher kinetic energy/more collisions between enzyme and substrate/ref to optimum temper enzyme substrate complexes formed quicker;	rature/
 (ii) initial colour change may indicate early enzyme activity; but kinetic energy/molecular excitation/high temperature; caused denaturation/disruption of active site/breakage of hydrogen bonds/disulphide bridges/loss 	of tertiary structure;
	TOTAL 11
QUESTIONSHEET 8	
(a) Any four of: use equal masses of each tissue/ ground up equally/same time and speed of grinding/ equipment washed between samples/ equal volumes of hydrogen peroxide used/ tissues completely immersed/	
effervescence measured at same time interval;;;;	4
(b) (i) liver;	1
(ii) most metabolically active/highest or fastest respiratory rate;	1
(c) more enzyme = more active sites; more collisions between substrate and active site/faster formation of enzyme-substrate complexes;	2
	TOTAL 8
QUESTIONSHEET 9	
(a) changes shape of active site;	1
(b) (i) decreases/stops it;	1
(ii) substrate no longer able to bind to active site so that enzyme-substrate complex cannot form; allosteric inhibitor attaches to enzyme at a site other than the active site; but changes the molecular shape of the enzyme;	max 2
(c) Y; normal substrate of glucose phosphorylase is glucose; Y is similar shape/structure;	3
	TOTAL -

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QUESTIONSHEET 10

- (a) (i) B: pepsin works in stomach/acid environment;
 - alkaline solution would disrupt charges on/shape of active sites/tertiary structure/might denature enzyme;
- 2

2

1

C+D: boiling denatures enzyme; (ii)

E: no enzyme present;

active site/tertiary structure lost;

- 1
- (b) add more egg white to tube A/repeat experiment in tube A;

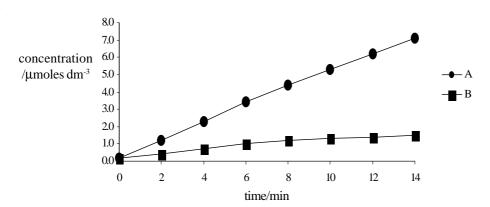
1

(c) to show that boiling inactivated the enzyme in both pH's;

TOTAL 7

QUESTIONSHEET 11

(a)



time on X axis and concentration on Y axis with units;

suitable scale which is easy to use (at least ½ the graph paper sheet);

accurate clear plotting;

points joined cleanly using a ruler (according to Institute of Biology Guidelines for A level Biology);

(lines of best fit/curves will not be accepted by Examining Boards in this type of question)

curves labelled clearly/suitable key;

(b) readings from graph at 11 and 3 minutes are 5.75 and 1.75

$$\frac{5.75 - 1.75}{8} = ; \quad 0.50 \text{ micromoles dm}^{-3} \text{ min}^{-1};$$
(correct no of significant fix

(correct no of significant figures needed.

units needed.

allow consequential errors and range ± 0.05 .)

(c) (i) A, because the rate of substrate digestion was the highest overall; 1

5

2

(ii) temperature;

use a water bath/incubator;

pH;

use a buffer;

4

(d) Any two of:

tenderising of meat/

predigesting baby foods/

rennet in cheese manufacture/biological washing powders/making leather pliable/dissolving blood clots /lowering protein content of flour for biscuit manufacture/brewing;;.

2

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QUESTIONSHEET 12

(a) (i) C;

(ii) D;

(iii) B;

(iv) A;

(b) malonic acid/malic acid;

inhibits succinic dehydrogenase;

TOTAL 6

4

2

QUESTIONSHEET 13

Feature	Amylase	Lactic dehydrogenase
Will breakdown lactose	×	× ;
Found only in animals	×	× ;
Requires NAD	×	✓ ;
Is a Hydrolase	✓	× ;
Can be made by GE	✓	✓ ;
Can be used to make yoghurt	×	× ;

(Lactobacilli used to make yoghurt)

TOTAL 6

QUESTIONSHEET 14

(a) 43%;

(b) (i) pancreas;

(ii) to provide the correct pH for the enzyme/may be a enzyme cofactor;

(c) amylase in seed hydrolyses starch to reducing sugars/maltose; to act as respiratory substrate to provide energy for germination;

amylase will only operate efficiently near optimum temperature/increase in temperature increases amylase activity;

(d) heat energy disrupts hydrogen bonds holding 3-D structure (of protein) together; ref denaturation;

TOTAL 8

3

2

QUESTIONSHEET 15

ATP/adenosine triphosphate;

glycosidic;

hydrolases;

water;

oxido-reductases;

NAD/FAD;

NADP;

activation energy; TOTAL 8

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QUESTIONSHEET 16

(a) plant cell wall/middle lamella (of cell wall) contains pectin/calcium pectate/magnesium pectate;

this is degraded/broken down by pectinase;

thus releasing cell contents/juice/oils/pigments from fruit;

fewer bits of fruit/cell wall left suspended (in juice);

max 3

(b) use same mass of apple tissue in each sample;

tissue from same apples/batch of apples/variety of apples;

at same stage of ripeness;

homogenise (in blender/food mixer) for a standard time/speed;

one sample with pectinase solution added and one sample with an equivalent volume of water added;

allow to stand for a suitable time/at least 30 minutes/to enable enzyme to work;

both samples at same temperature/room temperature/37°C in water bath/incubator;

filter/low power centrifugation in a standard way/for a standard time;

measure volume of juices collected;

compare clarity by eye/measure turbidity with photometer;

do replications;

max8

TOTAL 11

QUESTIONSHEET 17

(a) pH/hydrogen ion concentration influences the ionisation of R-groups/side chain groups;

which influence the conformation/shape of the protein/enzyme/active sites;

so that active site can join to the substrate/ref induced fit hypothesis/lock and key hypothesis;

best fit/most efficient enzyme action occurs at optimum pH/around pH 2.2;

shape of active site less suitable at pHs either side of the optimum so that rate of activity falls;

pH may also affect ionisation of substrate which could influence rate of reaction;

ref to extremes of pH cause denaturation;

max 5

(b) shape of active site must adjust slightly to accommodate different shapes/ionic states of albumin and haemoglobin; slight changes in pH will slightly alter active site shape/ionisation;

thus trypsin is able to digest more than one type of protein;

ref to induced fit hypothesis/mechanism;

max 3

(c) acetylcholine esterase is important in regulating nervous/synaptic transmission;

operates efficiently over a wide pH range/pH 6.9 – 10, so not affected by body pH changes in this range;

also operates reasonably effectively from pH 6.0 - 6.9/in dilute acidic conditions;

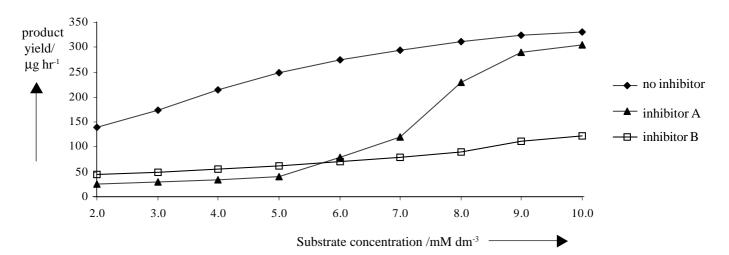
max 2

(d) operates effectively over a wide range of pH values whereas pepsin would only be effective in low pH range/high acidity; pH 2 would affect flavour of meat/make it unpalatable/papain could be used at pH 7 which would not affect the meat flavour;

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QUESTIONSHEET 18

(a)(i)



axes correct and labelled;

suitable scale;

accurate plotting;

points joined with a ruler (IOB recommendations)

curves labelled/suitable key;

5

- (ii) same concentration of enzyme in each;
 - above limiting concentration;
 - same volumes for each reagent;
 - suitable pH maintained by a buffer;

suitable temperature/37°C maintained by waterbath;

same incubation time for each;

max 4

(b) 1. competitive;

because when substrate concentration is in excess/rises above inhibitor concentration/above 5.0 mM dm⁻³; product yield rises to a high level;

3

2. non-competitive;

because yield of product remains low;

even when substrate concentration is high/in excess of inhibitor;

3

(c) succinic dehydrogenase; succinic acid; fumaric acid; malonic/malic acid; (accept any other correct example)

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QUESTIONSHEET 19

(a) enzymes are attached to/entrapped by insoluble materials/matrix which gives support to the enzyme; enzyme is then held in place during the reaction; whole cells/yeasts/bacteria with specific enzymes can also be immobilised;

3

(b) product will not be contaminated by the enzyme; since enzyme cannot escape from the matrix;

enzyme can be recovered and used again many times; useful if enzyme is costly/hard to extract/produce;

enzyme is more stable at extremes of temperature/pH; since protected by the matrix;

very useful for continuous fermentations; since enzymes/cells remain trapped in matrix;

immobilised whole cells mean that enzyme sequences/several enzymes can operate simultaneously; reducing number of steps in the process/reducing cost; (any three pairs of marks) **max 6**

(c) use of glucose isomerase to convert glucose to fructose (when making fructose syrups); continuous production of ethanol using yeast/zymase; production of vinegar using Acetobacter;

max 2

TOTAL 11

QUESTIONSHEET 20

(a) hundreds of enzymes in cell must work in relationship together/in integrated fashion;

some will need to work quicker/slower than others so that products are formed in an orderly way/at correct rate/at a rate that does not cause inbalance;

having different pH-activity profiles/different optimum pHs means that they are not all working at peak rate; thus intracellular pH has a controlling influence on (intracellular) enzyme actions;

max 2

(b) carbonic anhydrase has a large number of active sites per molecule whereas succinic dehydrogenase only has one or a few; carbonic anhydrase has high activity to convert all carbon dioxide to carbonic acid/hydrogen carbonate ions in red cells (and converse reaction);

must have reserve capacity so that (blood) CO₂ tension does not build up during strenuous activity; succinic dehydrogenase limited by capacity of mitochondrion to absorb pyruvate, so will not need a very high turnover number;

max 2