F212: Nodule 1: Enzymes June 2009-January 2013 Mark schemes

- (a) state that enzymes are globular proteins, with a specific tertiary structure, which catalyse metabolic reactions in living organisms;
- (b) state that enzyme action may be intracellular or extracellular;
- (c) describe, with the aid of diagrams, the mechanism of action of enzyme molecules, with reference to specificity, active site, lock and key hypothesis, induced-fit hypothesis, enzymesubstrate complex, enzyme-product

complex and lowering of activation energy;

- (d) describe and explain the effects of pH, temperature, enzyme concentration and substrate concentration on enzyme activity;
- (e) describe how the effects of pH, temperature, enzyme concentration and substrate concentration on enzyme activity can be investigated experimentally,
- (f) explain the effects of competitive and noncompetitive inhibitors on the rate of enzyme-controlled reactions, with reference
- to both reversible and non-reversible inhibitors;
- (g) explain the importance of cofactors and coenzymes in enzyme-controlled reactions;
- (h) state that metabolic poisons may be enzyme inhibitors, and describe the action of one named
- (i) state that some medicinal drugs work by inhibiting the activity of enzymes

Question	tion Answer		Marks Guidance
3 (a)		ڻ.	5 Mark the first answer on each prompt line. If the answer is correct and an additional answer is given that is incorrect or
			contradicts the correct answer then = 0 marks
	globular; catalysts;		
	intracellular; extracellular / hydrolytic;		DO NOT CREDIT metabolic (as given in Q) DO NOT CREDIT digestive (as given in Q)
	inhibitors;		

Question		Answei	Mark	Caracitor
3 (b)		carry out with and without, Ca ²⁺ / cofactor;	ĊΊ	1 ACCEPT 'use a control with no calcium' 1 ACCEPT calcium as AW for Ca ²⁺ as the question is testing
				AO3 1 IGNORE increase / decrease , concentration
	N	idea of using at least three <u>concentration</u> s (of Ca ²⁺) (other than zero);		2 ACCEPT implication of 3 or more concentrations, e.g. 'use several concentrations'
	ω	keep , concentration / volume of , enzyme / rennin, constant ;		3 IGNORE amount
	4	keep , concentration / volume of , caseinogen / substrate / milk, constant ;		4 IGNORE amount
	ψı	keep , temperature / pH , constant ;		5 IGNORE 'use a water bath' unqualified
	o	measure / AW , appearance of , product / casein or		6 This mp is for measurement of the dependent variable 6 ACCEPT observe as AW for measure appearance
		measure disappearance of , substrate / caseinogens or assess cloudiness (of solution);		disappearance 6 ACCEPT filter and weigh 6 ACCEPT 'assess degree of solubility / insolubility'
	7	over time intervals / after fixed time / end point time ;		7 'measure how much substrate is left after 30 min' = 2 marks (mp 6 and 7)
	o	replicates / repeats ;		8 IGNORE repeat / replicate on its own – must imply minimum of 3 in total, i.e. original plus two

	_				3 (c)	Question
	5 some enzymes don't need cofactors;	4 proteins are not stored in the body but vitamins and minerals are;	3 idea that proteins are used for purposes other than enzymes;	2 idea that in enzyme action total mass of, cofactor / coenzyme, very small compared to total mass of protein;	1 idea of cofactors / minerals, being, recycled / used again;	Answer
Total 11						Marks
			3 CREDIT stated example, e.g. muscle / hormones / antibodies.3 IGNORE growth / repair / replace	2 AWARD only if the enzyme context is clearly stated		Guidance

	Allswer	
(a) (i)	2	Mark the first answer on each prompt line. If the answer is correct and an additional answer is given that is incorrect or
		contradicts the correct answer then = 0 marks.
A substrate;		
B active site;		
(ii) idea of simple representation of the , process / structure	of the , process / structure 1	Examples of acceptable responses 'to make the process easy to understand'
idea of showing people how it works;	works;	it is a visual representation'
		IGNORE ' because that's the way it works'
(iii) supported by , more evidence / new research / more v		IGNORE 'because it is still unproven'
idea of fitting evidence more closely (than lock & key);	/ new research / 1 max more work ;	ACCEPT example, e.g. X-ray crystallography
	vork;	ACCEPT example, e.g. X-ray crystallography ACCEPT e.g. 'it has now been found that the enzyme shape changes during the reaction'

Otion	\dashv	Answer	Marks	Guidance
(b) (i)			3 max	Mark the first answer on each prompt line. If the answer is correct and an additional answer is given that is incorrect or contradicts the correct answer then = 0 marks.
		enzyme / LDH,concentration / volume;		1 IGNORE 'amount / number'
	N	substrate / lactate, concentration / volume ;		2 IGNORE 'amount / number' 2 IGNORE 'reactants'
				1 or 2 CREDIT 'volume / concentration , of solution' once if no reference to enzyme or substrate
, to , til	رن د	time;		
		idea that fish should be as closely <u>related</u> as		4 ACCEPT e.g. 'same type of fish' 4 IGNORE size / age / sex
1 1200 1	رن ن	рН;		
(ii)			_	Do not award mark if more than one letter given.
		L;		ACCEPT lactate and water at all temperatures

_			(iv)				(11)	Question
the induced fit, will be easier / AW;	easier for active site to change shape (as part of induced fit);	more_bonds can form / greater surface area for contact (between active site and substrate);	easier for / increased chance of , substrate, entering <u>active</u> site;	4 idea of activation energy harder to reach;	3 less chance of substrate entering active site / less chance of ESC formation / fewer collisions between substrate and active site;	2 (at 1°C) low kinetic energy / KE , of , enzyme / substrate;	1 (1°C is) below the <u>optim</u> um temperature / <u>optim</u> um temperature is higher , for this enzyme ;	Answer
							2 max	Marks
	DO NOT CREDIT if a candidate thinks that flexibility increases kinetic energy	ACCEPT 'different bonds can form' ACCEPT '(named) bonds form more easily'	Answers must imply 'easier' or 'quicker' ACCEPT 'fitting into' 'joining' 'binding' IGNORE refs to 'binding to a larger range of substrates' IGNORE refs to ESC	4 ACCEPT 'activation energy is greater'	3 ACCEPT 'fewer ESC formed' 3 ACCEPT 'slower ESC formation' 3 IGNORE denatured	2 ACCEPT 'molecules' / 'particles'	1 ACCEPT 'optimum is 10°C' 1 IGNORE '1°C is not the <u>optim</u> um temperature' 1 ACCEPT '1°C is further away from the <u>optim</u> um (than 10°C)'	Guidance

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Question	Answer	Warks	Galdance	
(c) (i)	different, amino acids / amino acid sequence / primary	2	ACCEPT different it groups present	
	structure;			

	(d)					(c)	Question
	(E)			(iii)		3	on
any example of potential application;	enzyme could have potential / future , application ;	different , allele / gene (would code for the polypeptide);	different , proportion / ratio , of bases / nucleotides ;	different , base / nucleotide , sequence ;	different, (named feature of) secondary / (named feature of) tertiary / quaternary, structure ;	different, amino acids / amino acid sequence / primary structure;	Answer
	1 max	, , , , , , , , , , , , , , , , , , ,		. 2		N	Marks
eg medical use, low temperature washing powder, scientific research	IGNORE refs to enzyme being useful to the Antarctic fish IGNORE genetic resource or any ref to biodiversity ACCEPT 'could be of use to humans'	ACCEPT 'mRNA will be different'	ACCEPT 'number of bases / nucleotides' ACCEPT 'different numbers of A or T / C or G' ACCEPT 'more adenines' etc	IGNORE 'different gene sequence' IGNORE mutation ACCEPT different triplet / codon	ACCEPT e.g. more α-helices / different or fewer (named) bonds / (different) prosthetic group / co-factor / ion / co-enzyme / R-groups in different orientation / polypeptide OR chain will fold differently IGNORE 3D IGNORE protein / enzyme , will fold differently	ACCEPT 'different R groups present'	Guidance

	18	Total	
6 IGNORE education unqualified		6 educating people in the fishing industry;	
		5 idea of promoting other species (for eating);	
4 ACCEPT 'in captivity' / AW 4 ACCEPT 'fish farming' 4 ACCEPT ref to sperm / egg, banks		4 ex situ (conservation) / captive breeding;	
 3 CREDIT in terms of maintaining fish's food source 3 IGNORE 'feeding fish' 3 IGNORE refs to 'in National Parks' unqualified 3 e.g. 'protect habitat by banning fishing' = 2 marks (mp1 and mp 3) 		3 idea of protecting (this) habitat (from drilling etc);	
2 ACCEPT refs to net / mesh size2 ACCEPT idea of patrolling / enforcing		2 idea of quotas / limits on numbers caught;	
1 Answers must refer to banning or legislating (and tishing) 1 IGNORE 'legislation' unqualified, 1 IGNORE less fishing unqualified 1 IGNORE 'ban hunting' unqualified	2 max	1 ban fishing (in this area / Antarctic);	(II)
Guidance	Marks	Answer	Question

Quest	Question		Expected Answers	Mark
2 (a)	a)	(enz	(enzymes are) proteins / used in metabolism / used in named metabolic pathway;	
		alte	alter rate of (chemical) reaction / lowers activation energy / provides alternative route for reaction / is not changed / is not used up;	<u> </u>
2 (b)	b) (i)			
		time;		
2 (b)	(ii)	P1	idea of different samples have different concentrations of, catalase / enzyme;	
	<u> </u>			
	<u> </u>	3	One of source the extract for the whole experiment from a single source;	ā
		M3 M4	thorough, mixing, required before use; filter / purify, extract; idea of using, known / standard, concentration of enzyme;	
		3 7	commercial source of catalase;	2

		_	
temperature; maximum / peak / V _{max} ; <u>denatured</u> ; <u>active</u> ;			
3)	(iii)	(C)	72
) 2;	(3)	<u>(c)</u>	73
no , reaction / oxygen produced , at 60(°C) ;	<u> </u>		
comparative figures to support any point;	(J)		
idea that before peak / after peak , temperature increase has	4		
(overall) fall more rapid than rise;	ω		
rate, rises / increases, initially; peak at / maximum at / highest at / decrease after, 40(°C);	2 1 (3)	(C)	73
compare results with , others / book / internet values			
mean / range / standard deviation / error bars / % error;		<u></u>	
repeat / replicate; compare replicate values / identify anomalous results;	(iii)	(b)	2
Expected Answers	ion	uest	
Expect: e values / i te values / i tandard de with , othe with , init eases , init			tion (iii)

	***	2 (6			2 (a		2 (a	Que
		(a) (iii)		,-,-	(a) (ii)		(a) (i)	Question
3 iodine, released / no longer in complex / AW;	helix, lost / unravels / AW;	1 hydrogen / H, bonds break;	2 (between) electronegative / δ^- , and electropositive / δ^+ ;	1 between oxygen and hydrogen (atoms);			blue-black / black / dark blue ;	Expected Answer
2 max		, CHA	N					Mark
3 ACCEPT no longer contained in helix	2 ACCEPT spiral / coil	IGNORE refs to denaturation	2 DO NOT CREDIT ions / + and - 2 ACCEPT slight / partial (negative / positive), charge	1 DO NOT CREDIT molecules / ions	CREDIT marking points from clearly labelled diagram max 1 if incorrect charges are on atoms	ACCEPT acceptable colour change	DO NOT CREDIT blue or purple unqualified by	Additional Guidance

I		_		12 using, standard		greater mass pp	41 more transmissi OR	mass of precipitate	T10 reading of, trans	9 use (red / orange) filter;	8 calibrate / zero,	C7 colorimeter;	C6 remove precipita	B5 changes to, gree		B3 heat with, Benec	B2 same <u>volume</u> s (2 (b) 1 take samples at	Question	
	use graph to read off concentration of maltose / AW;	plot, transmission / absorbance / mass of ppt, against	on curve;	using, standard / known, concentrations (of maltose);		greater mass ppt, = more maltose present ; ora	more transmission / less absorbance, of filtrate, OR	ale;	reading of, transmission / absorbance OR		calibrate / zero, using, a blank / water / (unreacted)		remove precipitate / obtain filtrate ;	changes to, green / yellow / orange / brown / (brick) red ;	Benedict's;	heat with, Benedict's (solution) / CuSO4 and NaOH;	same <u>volume</u> s (of solutions) added / removed (each time) ;	take samples at a range of times / AW;	Expected Answer	
	6 max									•									Mark	
				12 CREDIT 'serial dilutions'	11 DO NOT CREDIT if precipitate is added to	transmission / more absorbance, = more maltose present'	11 if unfiltered Benedict's / precipitate is clearly indicated as being present in sample, ACCEPT 'less		not, pass through'	9 DO NOT CREDIT if colour of filter is incorrect	8 IGNORE 'control'		C6 CREDIT description of method e.g. filtering / centrifuging / decanting		mixture at the beginning	B3 DO NOT CREDIT boil / warm B3 DO NOT CREDIT if Benedict's added to the	B2 must be in context of Benedict's test rather than reaction mixture		Additional Guidance	

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		<u>c</u>								**	(c)	Question												
ω	2	(iii)								ယ	(i) 2	on												
enzyme substrate complex / ESC, forms more, easily /	(Cl') binds to, enzyme / amylase / amylose / substrate;	(acts as a) cofactor;	Allow a + /- 1% for any concentration of maltose a	Difference in maltose concentration When chloride ions are either present or absent	Chloride ions absent	Chloride ions present	Presence or absence of chloride ions			figures with units to support mp 1;	increases / greater / faster ; reaction completed in / plateaus after / concentration is	Expected Answer												
ıre, easily q	substrate		/ concentr	0	0	0	0.0 min	The per		8	er / concentration is													
ly / quickly ;	u i				į		ation of	12	12	24	0.5 min	centage			es.									
			maltose	34	20	54	1.0 mìn	The percentage concent	2 max															
3 ACCI	2 ACCI	1 IGNORE 'coenzyme'	and a +/-	41	29	70	nin	itration o	maltose 3 ACCE 3 DO No 3 ACCE within ±	3 two ma	1 ACCE													
3 ACCEPT description	2 ACCEPT binds to, active site		RE 'coen	RE 'coen	RE 'coen	RE 'coen	RE 'coen	RE 'coen	RE 'coen	RE 'coen	RE 'coen	RE 'coen	RE 'coen:	RE 'coen:	2% for the	44	36	80	2.0 min	f maltose	malfose concentration. 3 ACCEPT calculated difference 3 DO NOT CREDIT if '%' and 'm 3 ACCEPT any concentration wi within ± 0.05 min.	altose con ne or two	PT any ti	
ription	s to, activ		e differen	48	40	88	2.5 min) (%) pre	ation. lated diffe	3 two maltose concentrations (+ or – chloride) for a given time or two times (+ or – chloride) for given	me betwe													
	e site		ce in mal	50	45	95	3.0 min	sent eve	rence and 'min.' ion withir	ns (+ or - or – chlor	en 3.45 €													
			nd a +/- 2% for the difference in maltose concentrations	52	48	100	3.5 min	ration of maltose (%) present every half a minute	malfose concentration. 3 ACCEPT calculated difference 3 DO NOT CREDIT if '%' and 'min.' not given 3 ACCEPT any concentration within ± 1 % and time within ± 0.05 min.	- chloride ide) for g	ACCEPT any time between 3.45 and 3.55 min.													
			entrati	50	50	100	4.0 min	minute	nd time) for a iven	min.													

1 temperature; 2 pH; 3 enzyme / amylase / chloride, concentration; 5 constant / regular, stirring; 6 (fixed) yolume of solution (removed each time for sampling); 3 max	۵	Question	tion	
temperature; pH; enzyme / amylase / chloride, <u>concentration</u> ; substrate / starch / amylose, <u>concentration</u> ; constant / regular, stirring; (fixed) <u>volume</u> of solution (removed each time for sampling);		(c)	(E)	<u></u>
pH; enzyme / amylase / chloride, <u>concentration</u> ; substrate / starch / amylose, <u>concentration</u> ; constant / regular, stirring; (fixed) <u>volume</u> of solution (removed each time for sampling);				<u></u>
enzyme / amylase / chloride, <u>concentration</u> ; substrate / starch / amylose, <u>concentration</u> ; constant / regular, stirring; (fixed) <u>volume</u> of solution (removed each time for sampling);				2
substrate / starch / amylose, <u>concentration</u> ; constant / regular, stirring; (fixed) <u>volume</u> of solution (removed each time for sampling);		-		
constant / regular, stirring; (fixed) <u>volume</u> of solution (removed each time for sampling);				
(fixed) <u>volume</u> of solution (removed each time for sampling);				
		41.7		

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0	Question	tion	Expected Answer	Mark	Additional Guidance
4	(g)	(E) 2	non-competitive (inhibitor); (α-amanitin / inhibitor / toxin) fits into, allosteric site / a place other than active site;		3. ACCEDT 'Alefortion of active eite'
		· •	active site originals, strape / comparation / comornialion / structure ;		
		4	substrate no longer, ins / complementary to, <u>active site</u> ,	2 max	
4	(p)	E	inhibits production of mRNA / mRNA not produced;		
		N W	prevents protein synthesis / AW; e.g. of, specific named protein / (vital) process, that may be affected;	2 max	 3 CKEDII translation 3 e.g. respiration / photosynthesis (as question refers to 'an organism') / haemoglobin / cytochrome C oxidase
4	(၁)	©	sequence / order, of amino acids;	~	IGNORE number / organisation
	(3)	(II)			ALLOW phonetic spelling
			A = ionic; B = hydrogen; C = $\overline{disulfide}$ (bond / bridge);	က	DO NOT CREDIT disul <u>fate</u>
4	(p)	- 6	increased <u>kinetic</u> energy ;		1 must contain the idea of more than normal
		1 (7)	hydrophilic / hydrophobic / hydrogen / ionic, bonds / interactions, break;		3 IGNORE Van der Waals DO NOT CREDIT if disulfide / covalent / peptide
		4	change in, <u>3D</u> shape / conformation (of protein) ;		4 IGNORE tertiary / structure (as in question) IGNORE refs to, active site / enzymes
		က	denatures;	3 max	
			Total	17	

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shape 1 ACCEPT more successful collisions between substrate 3 DO NOT CREDIT refs to PABA and sulfonamide being complementary to each other or to the enzyme (alone) 1 ACCEPT similar structure DO NOT CREDIT same DO NOT CREDIT 'optimum rate' or 'rate levels off' 3 ACCEPT active sites filled / no free active sites sulfonamide has SONH₂ but PABA has N₂ sulfonamide has 1 more NH2 group Additional Guidance DO NOT CREDIT active sites run out 4 ACCEPT 'cannot work any quicker' 6 e.g. only sulfonamide contains S only PABA has COOH group and active site Marks 3 max 3 max limiting; | 1 substrate / PABA, and, inhibitor / sulfonamide, similar shape; 6 correct ref to a difference between sulfonamide and PABA; at high concentration all active sites occupied; (at high substrate concentration) enzyme concentration 2 more, enzyme substrate complexes / ESCs, formed; 1 more, PABA / substrate, molecules enter active site; at low concentration not all active sites occupied / achieves / reaches, max (turnover) rate / Vmax; 2 able to, bind / fit into / block, active site; **Expected Answers** 4 both have, hex / benzene / 6-C, (ring); (shape) complimentary to active site; 5 both have, NH2 / amine; without inhibitor .. × ന \equiv $\widehat{\boldsymbol{\Xi}}$ \equiv Question (a) (a) **(**p က