HELICATE · AS AND A-LEVEL BIOLOGY · TOPIC BANK

3.1.4 Proteins

Mark scheme

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TOTAL MARKS	QUESTIONS
74	13

How to use this mark scheme

;	Separates marking points. Each semicolon marks the end of one creditable point, and each point is worth one mark.
;;	Two marks for this answer (used for calculations).
/	Alternative words or phrases that are credited for the same point.
OR	An alternative answer for the same mark.
( )	Words in brackets are not needed to gain the mark.
max	More marking points are listed than marks available. Award up to the number of marks shown.
Accept	An alternative answer that gains credit.
Reject	A wrong answer. It does not gain credit and cancels a correct point it contradicts.
Ignore	Neither credited nor penalised. Mark the rest of the answer.
ECF	Error carried forward: credit correct working that uses an earlier wrong answer.
AO	Assessment objective: AO1 knowledge and understanding; AO2 applying knowledge; AO3 analysing, interpreting and evaluating.

Where an answer is correct but not listed, credit it if it is biologically accurate and answers the question at the depth the marks require. Right + wrong = wrong: a contradiction cancels a correct point.

Amino acids, peptide bonds and the biuret test 3.1.4.1

Question	Marking guidance	Mark	Comments
01.1	1. Central carbon atom bonded to H, R (group), NH_2 **and** COOH; 2. NH_2 labelled amine (group) **and** COOH labelled carboxyl (group);	**2** (2 × AO1)	1. **Accept** a displayed or structural formula, eg COOH drawn as C double-bonded to O and single-bonded to OH 1. **Accept** the ionised form, NH_3^+ and COO^- 1. **Reject** an H atom from the amine or carboxyl group drawn on the central carbon, or a central carbon with more than four bonds 2. **Accept** amino (group) for amine and carboxylic acid (group) for carboxyl 2. Both labels are needed; award point 2 even if point 1 is not awarded, provided the two groups are drawn

Question	Marking guidance	Mark	Comments
01.2	$C_5H_{10}N_2O_3$;	**1** (1 × AO2)	One molecule of water (H_2O) is removed in the condensation reaction that forms the peptide bond

Question	Marking guidance	Mark	Comments
02.1	Correct answer with working for 3 marks: lysine, proline **and** phenylalanine;;; Accept for 2 marks: R_f values of 0.14, 0.43 **and** 0.68 **OR** two of the amino acids correctly identified, with working Accept for 1 mark: distance moved by the solvent front = 7.7 ÷ 0.55 = 14 (cm)	**3** (3 × AO2)	**Accept** the alternative method: expected distances = R_f × 14 cm, eg lysine 2.0 cm, proline 6.0 cm, phenylalanine 9.5 cm **Ignore** the order in which the amino acids are given Identification without any working gains 1 mark only

Question	Marking guidance	Mark	Comments
02.2	1. Order: phenylalanine – proline – lysine; 2. Proline is in the middle (of the tripeptide) because it is in both dipeptides/is bonded to both other amino acids; 3. Phenylalanine is at one end (the amine end), so lysine is at the other end, with the free carboxyl group; 4. Peptide bonds form between the amine group (attached to the central carbon) of one amino acid and the carboxyl group of the next, so only one end of the chain has a free amine group OR the carboxyl group of phenylalanine forms the peptide bond with proline (and the carboxyl group of proline with lysine);	**3 max** (1 × AO1) (1 × AO2) (1 × AO3)	1. **Accept** Phe–Pro–Lys, or the order written from the carboxyl end (Lys–Pro–Phe) if lysine is identified as the free-carboxyl end 4. **Accept** the amine groups (not those in R groups) of proline and lysine are used in (forming) peptide bonds **Ignore** references to amine groups in R groups, eg in lysine; do not penalise a correct statement that lysine's R group has a free amine group 4. **Reject** peptide bonds form between R groups

Question	Marking guidance	Mark	Comments
02.3	Repeat the chromatography (of the sample and the standards) using a different solvent and compare the R_f values / positions of the spots with the standards again;	**1** (1 × AO3)	**Accept** run the hydrolysed sample and the standard of each suspected amino acid as a mixed spot, so a single spot confirms the identity **Ignore** repeat the experiment (with the same solvent) unqualified

Question	Marking guidance	Mark	Comments
03.1	1. Add biuret (reagent/solution) to the sample; 2. Purple/lilac/violet/mauve (colour);	**2** (2 × AO1)	1. **Accept** add sodium hydroxide (solution), then (dilute) copper sulfate (solution) 1. **Reject** heating; reject Benedict's, iodine or ethanol 1. **Reject** 'burette' 2. **Accept** blue to purple 2. **Reject** pink, red or blue-black

Question	Marking guidance	Mark	Comments
03.2	Correct answer for 2 marks: 3.3 (g per 100 cm^3);; Accept for 1 mark: 33 mg cm^{-3} (reading × 5, not converted) **OR** 0.66 (g per 100 cm^3) (reading converted correctly but dilution not taken into account)	**2** (2 × AO2)	Reading from the line of best fit: 6.6 mg cm^{-3}; × 5 (1 cm^3 drink in a total of 5 cm^3) = 33 mg cm^{-3} = 33 × 100 ÷ 1000 g per 100 cm^3 **Accept** readings of 6.5–6.7 mg cm^{-3}, giving 3.25–3.35, for 2 marks 2.64 OR 2.6 (g per 100 cm^3) (multiplied by 4 instead of 5, units converted correctly) gains 1 mark Rule for 1 mark: a reading correctly converted to g per 100 cm^3 OR the reading correctly multiplied by 5

Question	Marking guidance	Mark	Comments
03.3	(Undiluted) absorbance would be higher than that of the most concentrated standard/off the (calibration) graph, so the concentration could not be read from it;	**1** (1 × AO3)	**Accept** the protein concentration of the drink is above 10 mg cm^{-3}/outside the range of the standard solutions **Accept** so the reading is within the range of the calibration curve/line **Ignore** to make it more accurate unqualified

Question	Marking guidance	Mark	Comments
03.4	1. Particles absorb/scatter light, so the absorbance is higher (than from the protein alone) and the protein concentration is overestimated; 2. Filter/centrifuge the (diluted) drink before the test OR zero the colorimeter using the (diluted) oat drink mixed with water instead of biuret reagent / subtract the absorbance of the drink without biuret reagent;	**2** (2 × AO3)	1. Must state the direction of the error (too high) 2. **Accept** remove the particles before measuring the absorbance 2. **Ignore** use a blank unless the content of the blank is given

Protein structure and function 3.1.4.1

Question	Marking guidance	Mark	Comments
04.1	1. Holds them together: ionic bonds, because reagent 2 separated the polypeptides OR hydrogen bonds, because reagent 3 separated the polypeptides; 2. Does not: disulfide bridges, because reagent 1 did not separate the polypeptides;	**2** (2 × AO2)	Each point needs the bond **and** the reason 1. **Accept** ionic and hydrogen bonds 2. **Accept** sulfur bridges or S–S bonds for disulfide bridges **Reject** peptide bonds

Question	Marking guidance	Mark	Comments
04.2	1. Reagent 3 breaks hydrogen bonds, which hold the secondary **and** tertiary structure (of each polypeptide); 2. So the tertiary structure/shape of the binding site changes and it is no longer complementary to hormone T; 3. After reagent 2, (each separate) polypeptide still binds T (85%), so the binding site is (within) one polypeptide/the tertiary structure is (mostly) kept OR so quaternary structure is not needed for binding;	**3** (2 × AO2) (1 × AO3)	1. **Accept** hydrogen bonds hold the coiled shape/secondary structure of the polypeptide 1. **Accept** α-helix or β-pleated sheet for secondary structure; neither is required 2. **Reject** active site, enzyme or substrate for protein K or hormone T (once only) 2. **Accept** denatured for the change in tertiary structure 3. **Accept** ionic bonds are less important (than hydrogen bonds) in maintaining the shape of the binding site **Reject** peptide bonds broken

Question	Marking guidance	Mark	Comments
04.3	Disulfide bridges may be present within each polypeptide/in the tertiary structure, but the other bonds keep the shape when they are broken OR binding did fall slightly (to 96%), so disulfide bridges may have been broken;	**1** (1 × AO3)	**Accept** reagent 1 only shows that disulfide bridges do not join the two polypeptides **Ignore** only one reagent/one test used unqualified

Question	Marking guidance	Mark	Comments
05.1	Description: The further folding of a polypeptide into a specific overall shape Bonds that maintain this structure: Hydrogen bonds, ionic bonds and disulfide bridges;	**1** (1 × AO1)	The first row describes primary structure, the second secondary structure and the fourth quaternary structure (wrongly held by peptide bonds)

Question	Marking guidance	Mark	Comments
05.2	1. Change in the primary structure/sequence of amino acids; 2. Disulfide bridge cannot form (between the R groups of the amino acids at positions 58 and 112); 3. Tertiary structure of the receptor changes OR the receptor folds differently; 4. Shape of the binding site changes, so it is no longer complementary to F; 5. So F cannot bind (to the receptor);	**4 max** (2 × AO1) (2 × AO2)	2. **Accept** sulfur bridge or S–S bond for disulfide bridge 2. **Reject** a disulfide bridge forms between serine and cysteine 3. **Ignore** 3D shape for tertiary structure; reject 3° or 3o 4. **Reject** active site, enzyme, substrate or enzyme-substrate complex (once only) **Reject** amino acids are formed/produced (for the change in sequence) **Ignore** protein not made/receptor not formed

Enzyme action and specificity 3.1.4.2

Question	Marking guidance	Mark	Comments
06.1	Hydrolysis of starch by amylase in saliva in the mouth;	**1** (1 × AO1)	

Question	Marking guidance	Mark	Comments
06.2	1. Substrate binds to the active site OR an enzyme-substrate complex forms; 2. Active site changes shape (slightly) as the substrate binds, so it becomes complementary to the substrate/moulds around the substrate; 3. This puts strain/tension on bonds in the substrate OR distorts/bends bonds in the substrate; 4. So less energy is needed to break the bonds (so activation energy is lower);	**3 max** (3 × AO1)	2. **Reject** the substrate changes shape 2. **Reject** the enzyme changes shape unless the active site is referred to 3. **Accept** brings reacting groups close together 3. **Ignore** weakens bonds **Accept** E-S complex / ES complex. E-S alone only if 'enzyme-substrate complex' is written elsewhere. Ignore ESC. **Ignore** alternative pathway unqualified

Question	Marking guidance	Mark	Comments
07.1	1. *d* decreases (from 1.6 to 1.1 nm) when the substrate binds, so the active site changes shape (when the substrate binds); 2. In the lock-and-key model the active site has a fixed/rigid shape (that is already complementary), so *d* would not change;	**2** (2 × AO3)	1. **Accept** the active site closes around/moulds to the substrate 2. **Accept** active site is flexible in the induced-fit model but not in the lock-and-key model **Reject** the substrate changes shape

Question	Marking guidance	Mark	Comments
07.2	1. M causes only a small change in the shape of the active site (*d* falls by only 0.1 nm) OR the active site does not close around/mould to M; 2. So no/little strain is put on the bonds in M OR the activation energy (of a reaction of M) is not lowered (enough);	**2** (2 × AO2)	1. **Accept** the active site does not become complementary to M 2. **Accept** bonds in M are not distorted/broken **Ignore** M is a competitive inhibitor unqualified

Question	Marking guidance	Mark	Comments
08.1	1. Positively charged R groups of lysine and arginine form ionic bonds with/are attracted to the negatively charged R group in the active site; 2. So these dipeptides bind to/are complementary to the active site and (more) enzyme-substrate complexes form; 3. Alanine and phenylalanine have uncharged R groups, so no ionic bond forms and few enzyme-substrate complexes form; 4. Glutamic acid has a negatively charged R group, which is repelled by the negative R group in the active site, so (almost) no enzyme-substrate complexes form;	**3 max** (3 × AO2)	**Accept** opposite charges attract and like charges repel, if linked to the correct amino acids **Accept** E-S complex / ES complex. E-S alone only if 'enzyme-substrate complex' is written elsewhere. Ignore ESC. **Reject** peptide bonds form with the active site

Question	Marking guidance	Mark	Comments
08.2	R group of arginine has a different shape/size (from lysine), so it is less complementary to the active site OR fewer enzyme-substrate complexes form;	**1** (1 × AO3)	**Accept** the ionic bond forms less easily / the charge is in a different position **Ignore** arginine has a weaker charge unqualified

Factors affecting the rate of enzyme-controlled reactions 3.1.4.2

Question	Marking guidance	Mark	Comments
09.1	Correct answer in the range 1.7 to 2.3 (mmol dm^{-3} min^{-1}) for 2 marks;; Accept for 1 mark: a straight-line tangent drawn touching the curve at 0 minutes (the origin), whatever answer is given	**2** (2 × AO2)	The tangent to the plotted curve at 0 minutes has a gradient of 2.0 mmol dm^{-3} min^{-1} A chord from 0 to 2 minutes gives 1.6; this is not a tangent and gains 0 marks **Ignore** the unit if the number is correct

Question	Marking guidance	Mark	Comments
09.2	1. Substrate is used up/converted to product, so the substrate concentration falls; 2. So fewer enzyme-substrate complexes form (per unit time) OR fewer collisions between enzyme and substrate (molecules);	**2** (2 × AO1)	1. **Accept** the curve levels off because (almost) all the substrate has been converted **Reject** the enzyme is used up/denatured **Accept** E-S complex / ES complex. E-S alone only if 'enzyme-substrate complex' is written elsewhere. Ignore ESC.

Question	Marking guidance	Mark	Comments
09.3	Initial rate of reaction: About twice as high Concentration of product when the curve levels off: About the same;	**1** (1 × AO2)	More active sites, so more enzyme-substrate complexes form per unit time; the amount of substrate is unchanged, so the same amount of product forms

Question	Marking guidance	Mark	Comments
10.1	1. 67 (%) at 2 mmol dm^{-3}; 2. 27 (%) at 20 mmol dm^{-3};	**2** (2 × AO2)	(25.0 − 8.3) ÷ 25.0 × 100 = 66.8; (45.5 − 33.3) ÷ 45.5 × 100 = 26.8 If neither answer is to two significant figures, award 1 mark for 66.8 and 26.8 **Reject** percentage increase; reject negative values unless described as a decrease

Question	Marking guidance	Mark	Comments
10.2	1. Drug D causes a smaller percentage decrease in rate at the high substrate concentration than at the low substrate concentration (27% compared with 67%); 2. Drug D has a similar shape to the substrate and binds to the active site; 3. At high substrate concentration, substrate (molecules) are more likely to collide with/occupy the active sites than drug D OR the substrate out-competes drug D, so more enzyme-substrate complexes form; 4. A non-competitive inhibitor would cause the same percentage decrease at both substrate concentrations;	**3 max** (1 × AO2) (2 × AO3)	1. Must be a comparison; accept the effect of D is reduced/overcome at high substrate concentration 1. **Accept** the same idea at any drug concentration, eg at 40 μmol dm^{-3} 2. **Accept** complementary to the active site; reject the same shape as the substrate **Accept** E-S complex / ES complex. E-S alone only if 'enzyme-substrate complex' is written elsewhere. Ignore ESC.

Question	Marking guidance	Mark	Comments
10.3	1. Drug E binds to the enzyme at a site other than the active site; 2. Changes the tertiary structure of the enzyme/the shape of the active site; 3. Active site is no longer complementary, so the substrate cannot bind OR fewer enzyme-substrate complexes form; 4. Substrate does not compete with drug E for the same site, so more substrate cannot displace drug E/reverse the change in the active site;	**3 max** (2 × AO1) (1 × AO2)	**Max 2** if point 4 is not given (the effect of substrate concentration must be explained) 1. **Accept** allosteric site; do not require it 2. **Do not accept** 'denatures the enzyme' for point 2 3. **Accept** E-S complex / ES complex. E-S alone only if 'enzyme-substrate complex' is written elsewhere. Ignore ESC. **Reject** drug E binds to the substrate

Question	Marking guidance	Mark	Comments
11.1	1. Activity is highest at pH 3 (the optimum) and decreases as the pH rises (to zero at pH 7); 2. As the pH rises, the concentration of hydrogen ions falls; 3. This changes the charges on the R groups of amino acids (in the active site); 4. Ionic bonds **and** hydrogen bonds (in the tertiary structure) are broken/altered; 5. Tertiary structure/shape of the active site changes; 6. Active site is no longer complementary to the substrate/protein, so fewer enzyme-substrate complexes form; 7. At pH 7 the protease is denatured (so there is no activity);	**5 max** (4 × AO1) (1 × AO2)	1. **Accept** optimum of 2.5–3.5 4. **Reject** peptide bonds broken 5. **Accept** the protease is denatured for point 5 if linked to a change in the shape of the active site **Reject** the enzyme is killed **Accept** E-S complex / ES complex. E-S alone only if 'enzyme-substrate complex' is written elsewhere. Ignore ESC.

Question	Marking guidance	Mark	Comments
11.2	Correct answer for 2 marks: 32 OR 31;; Accept for 1 mark: $[H^+]$ of 1(.0) × 10^{-3} **and** 3.2 × 10^{-5} (mol dm^{-3}) **OR** $10^{1.5}$ OR 31.6 (not to two significant figures)	**2** (2 × AO2)	31 comes from using the rounded value 3.2 × 10^{-5}; accept either **Reject** 1.5 (difference in pH)

Required practical 1: rate of an enzyme-controlled reaction 3.1.4.2 (RP1)

Question	Marking guidance	Mark	Comments
12.1	1. Concentration of starch (solution); 2. Concentration of amylase (solution); 3. pH (using a buffer); 4. Volume/size of the drops of mixture or of iodine solution; 5. Concentration of iodine solution;	**2 max** (1 × AO1) (1 × AO3)	**Accept** source/type of amylase or of starch **Ignore** temperature: it is the independent variable **Ignore** volume unqualified, time, or amount

Question	Marking guidance	Mark	Comments
12.2	Correct answer for 2 marks: 21 OR 21.4 (%);; Accept for 1 mark: 30 ÷ 140 (× 100)	**2** (2 × AO2)	30 ÷ 140 × 100 = 21.43 **Accept** ±21% 10.7 (%) (half the interval used) gains 1 mark

Question	Marking guidance	Mark	Comments
12.3	Test drops of the mixture more often/at shorter intervals, eg every 10 seconds;	**1** (1 × AO3)	**Accept** use a lower concentration of amylase (or a larger volume of starch) so that the times are longer **Ignore** repeat more times **Ignore** use a colorimeter (not the same method)

Question	Marking guidance	Mark	Comments
12.4	A line graph with temperature on the *x*-axis, because temperature is a continuous variable;	**1** (1 × AO2)	

Question	Marking guidance	Mark	Comments
12.5	1. Supports: the mean time is shortest/rate is highest at 40 °C (140 s); 2. Against: temperatures were only 10 °C apart, so the optimum could be anywhere between 30 °C and 50 °C; 3. Against: more temperatures (eg at 1 °C or 2 °C intervals) between 30 °C and 50 °C are needed;	**2 max** (2 × AO3)	**Max 1** if only supporting or only opposing points are given 2. **Accept** the optimum could be between 40 °C and 50 °C **Ignore** uncertainty in the times or variation between repeats: even if each time could be up to 30 s shorter, the ranges at 40 °C (110–140 s) do not overlap those at 30 °C or 50 °C, so 40 °C is still the fastest temperature tested **Ignore** only three repeats unqualified

Question	Marking guidance	Mark	Comments
13	1. Dilute the stock solution with (distilled) water to make a range of (at least five) copper sulfate concentrations; 2. Include a control with water instead of copper sulfate (solution) OR a control with a sulfate salt of another metal (eg sodium sulfate) at the same concentrations, to show the effect is due to copper ions; 3. Add the same volume of each copper sulfate solution to each reaction mixture OR keep the total volume the same; 4. Use the same volume **and** concentration of starch solution and of amylase solution each time; 5. Keep the temperature constant with a water bath (below the optimum, so the amylase is not denatured) and let the solutions reach this temperature before mixing; 6. Keep the pH constant with a buffer; 7. Test a drop every 10–30 seconds and record the time until the iodine solution stays orange-brown; 8. Calculate the rate as 1 ÷ time; 9. Repeat (at least three times) at each concentration and calculate a mean;	**6 max** (2 × AO1) (4 × AO3)	**Max 4** if no control variables are described (points 3–6) 1. **Accept** a stated range, eg 0.1 to 1.0 mmol dm^{-3}, or a dilution series 5. **Accept** 40 °C; ignore room temperature unless a way of keeping it constant is given 7. **Accept** use a colorimeter to measure the change in colour of a starch–iodine mixture 8. **Accept** 1000 ÷ time **Ignore** same amount unqualified

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