PAG 6 Chromatography and Electrophoresis: OCR A Level Biology Exam

18 questions · 40 marks · about 48 minutes. Answer in the spaces, then check the mark schemes at the end.

Question 1 · A level Paper 3 · June 2023 · Q2(a)(i) · 1 mark

2 Vitamins are molecules that are consumed in the diet of animals and have essential roles in the body.

(a) Thin layer chromatography (TLC) was used to separate a mixture of vitamins from a vitamin supplement tablet.

(i) Most vitamins are colourless.

Suggest **one** method for visualising the vitamins in TLC.

.....

..... **[1]**

Question 2 · A level Paper 3 · June 2017 · Q1(b)(i) · 1 mark

(b) Thin layer chromatography can also be used to separate photosynthetic pigments.

(i) State a material that can be used as the stationary phase in thin layer chromatography.

..... **[1]**

Question 3 · AS Paper 2 · June 2025 · Q1(c) · 2 marks

(c) Outline the role of the solvent in paper chromatography.

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..... **[2]**

Turn over

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(c) A group of students wanted to use thin layer chromatography to identify four amino acids.

To produce the chromatogram, the students:

- drew a pencil line 1 cm from the bottom of the chromatography plate and put solvent into the beaker to a height of approximately 0.9 cm
- held the chromatography plate firmly in the middle with their hands and lowered it into the beaker
- left the apparatus to stand as shown in Fig. 1.4.

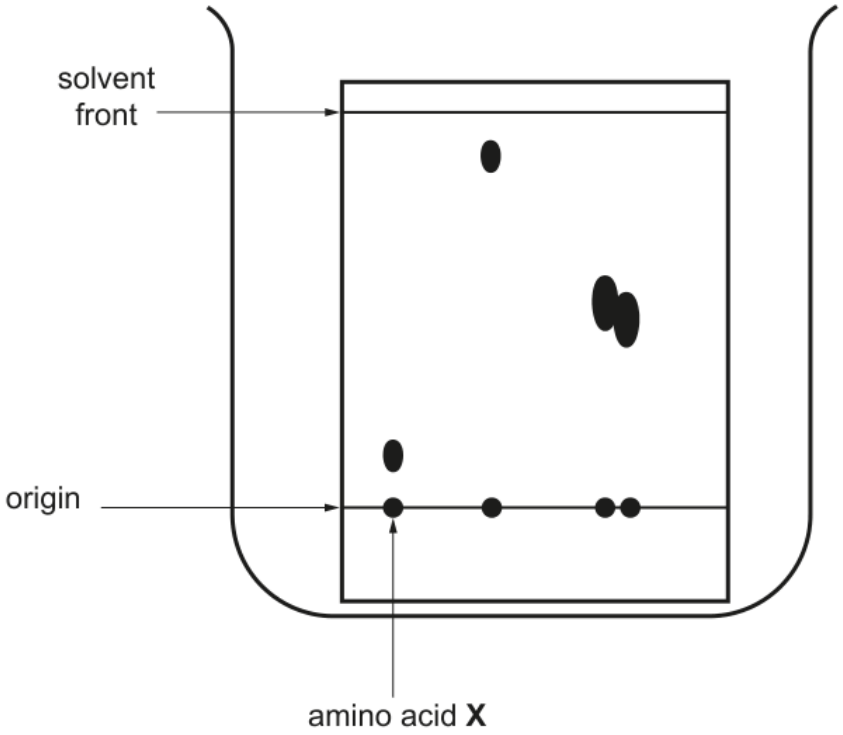


Fig. 1.4

7

(i) Describe **four** ways you would refine the method used by the students. For each change you suggest, give a reason why this would improve the results of the experiment.

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[4]

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Question 5 · AS Paper 2 · June 2018 · Q1(c)(ii) · 2 marks

(ii) Table 1 shows the R_f values of some amino acids.

Name of amino acid	R_f value
Alanine	0.31
Cysteine	0.40
Glutamine	0.13
Phenylalanine	0.59

Table 1

Using the information in Table 1 and Fig. 1.4, identify amino acid **X** by calculating its R_f value.

Show your working.

R_f value of amino acid **X** .....

Name of amino acid **X** .....

[2]

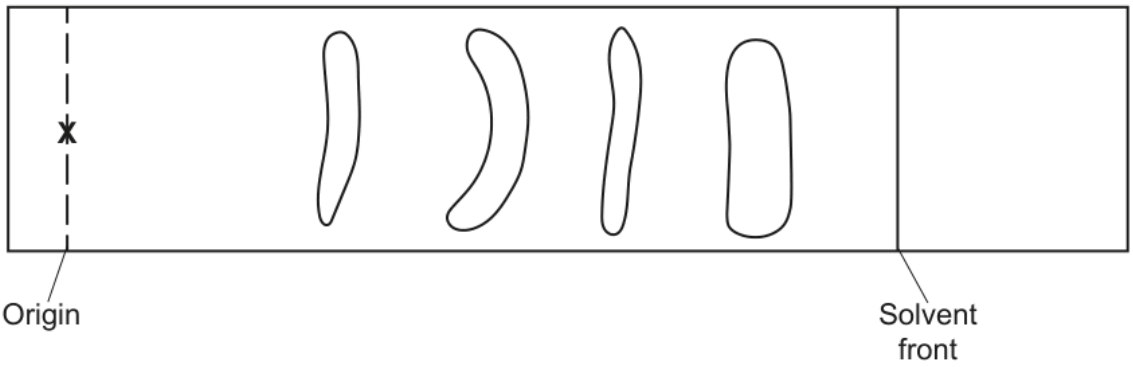
Turn over

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Question 6 · A level Paper 1 · June 2025 · Q16(b)(i) · 3 marks

(b) A student used thin layer chromatography (TLC) to separate the photosynthetic pigments in a chloroplast extract.

This is the result of the TLC.



(i) Calculate the R_f for the most soluble pigment.

Give your answer to **2** significant figures.

Distance travelled = ... mm

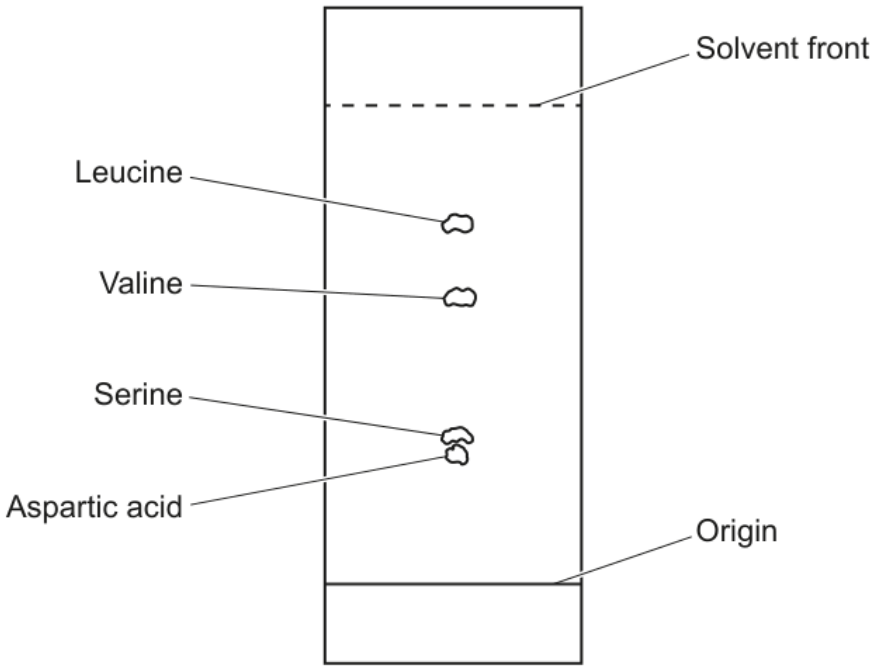
R_f = ...

[3]

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Question 7 · AS Paper 1 · June 2023 · Q23(b)(i) · 2 marks

(b) The diagram shows the chromatogram that was produced when using TLC to separate a mixture of amino acids.



(i) Use the chromatogram to calculate the Rf value of valine.

Rf = ... **[2]**

Question 8 · information from earlier in the question

2 Vitamins are molecules that are consumed in the diet of animals and have essential roles in the body.

(a) Thin layer chromatography (TLC) was used to separate a mixture of vitamins from a vitamin supplement tablet.

(i) Most vitamins are colourless.

Suggest **one** method for visualising the vitamins in TLC.

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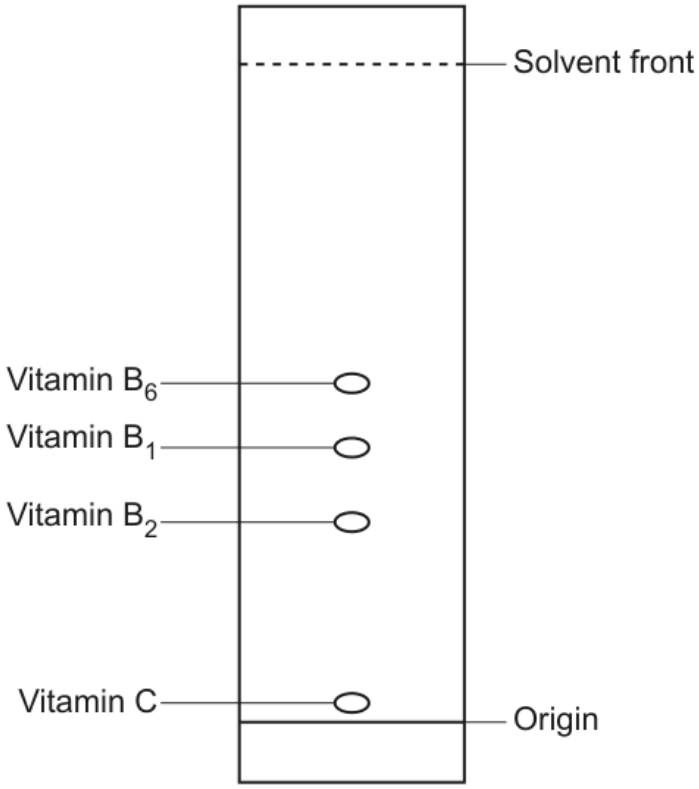
..... **[1]**

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Question 8 · A level Paper 3 · June 2023 · Q2(a)(ii) · 2 marks

(ii) The results of the TLC are shown in the chromatogram in **Fig. 2.1**.

Fig. 2.1



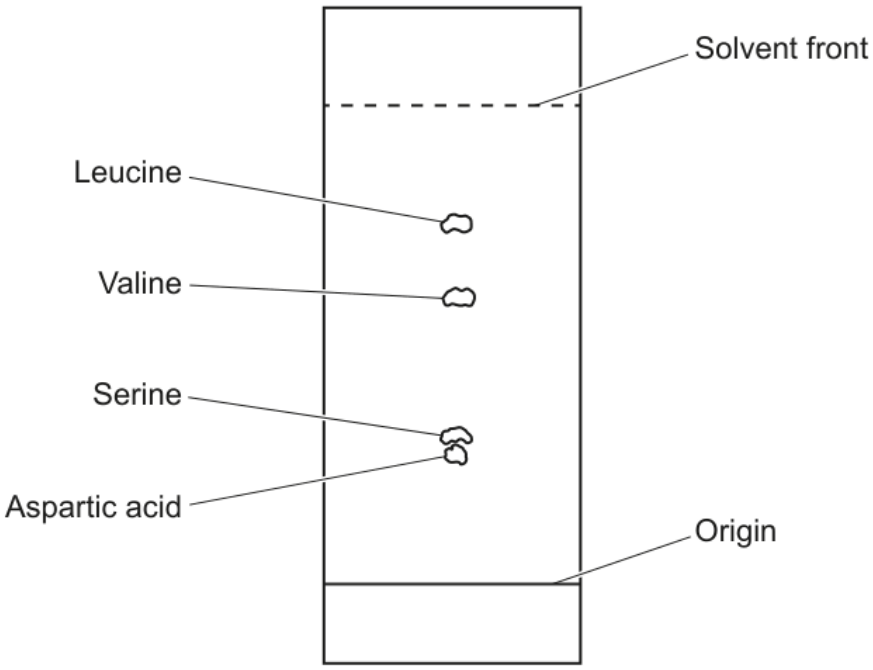
Use **Fig. 2.1** to calculate the R_f value of vitamin B_2.

R_f = ... **[2]**

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Question 9 · information from earlier in the question

(b) The diagram shows the chromatogram that was produced when using TLC to separate a mixture of amino acids.



(i) Use the chromatogram to calculate the Rf value of valine.

Rf = ... **[2]**

Question 9 · AS Paper 1 · June 2023 · Q23(b)(ii) · 1 mark

(ii) Serine and leucine have different R groups.

With reference to the chromatogram, suggest what can be concluded about the chemical properties of the R groups in serine and leucine.

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..... **[1]**

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Question 10 · information from earlier in the question

17 A student investigated photosynthetic pigments in spinach leaves using thin layer chromatography (TLC).

(a) Fig. 17.1 shows the student's plate at the end of the investigation.

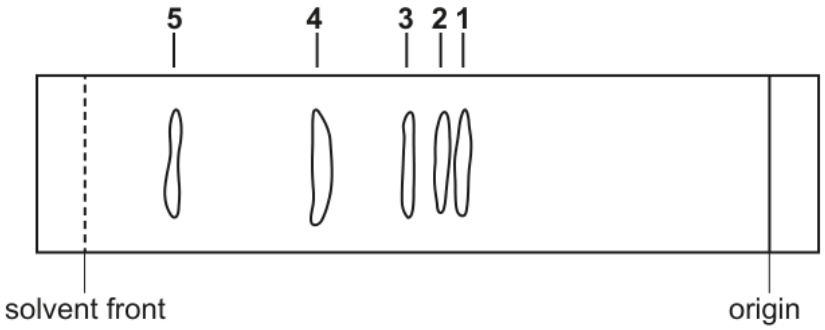


Fig. 17.1

The table shows colours and R_f values for several photosynthetic pigments.

Pigment	Colour	R_f
Carotene	Yellow-orange	0.90
Chlorophyll a	Blue-green	0.53
Chlorophyll b	Green	0.49
Pheophytin	Grey	0.65
Xanthophylls	Yellow	0.32–0.44

(i) Calculate the R_f value for spot **3** and use this to identify the pigment that spot **3** represents.

R_f = ...

pigment = ...

[3]

Question 10 · A level Paper 1 · November 2020 · Q17(a)(iii) · 2 marks

(iii) The solvent used for the separation was non-polar.

Identify the spot corresponding to the **least** polar pigment. Give a reason for your choice.

spot .....

reason .....

.....

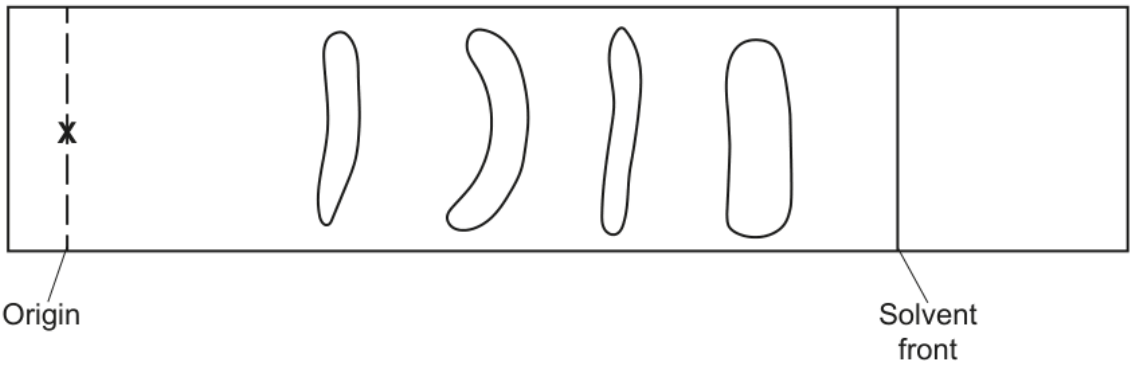
[2]

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Question 11 · information from earlier in the question

(b) A student used thin layer chromatography (TLC) to separate the photosynthetic pigments in a chloroplast extract.

This is the result of the TLC.



(i) Calculate the R_f for the most soluble pigment.

Give your answer to **2** significant figures.

Distance travelled = ... mm

R_f = ...

[3]

Question 11 · A level Paper 1 · June 2025 · Q16(b)(ii) · 2 marks

(ii) The student noticed that the spots were very faint and thought that there might be other pigments present in the mixture that were too faint to be seen.

Describe how the student could increase the intensity of the spots using the same chloroplast extract.

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..... **[2]**

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Question 12 · A level Paper 1 · June 2025 · Q16(b)(iv) · 1 mark

(iv) The student was not able to identify any of the pigments using R_f values from a data book.

Suggest an explanation for this.

.....

..... **[1]**

Question 13 · A level Paper 1 · June 2025 · Q16(b)(v) · 2 marks

(v) Describe how the student could use TLC to identify the pigments present in the mixture without calculating R_f values.

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..... **[2]**

Turn over

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Question 14 · A level Paper 3 · June 2017 · Q1(c) · 3 marks

(c) When sequencing DNA, fragments of DNA are separated by electrophoresis.

Describe **three** differences between the process of thin layer chromatography and the form of electrophoresis used to sequence DNA.

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..... **[3]**

Turn over

Question 15 · A level Paper 2 · November 2021 · Q16(b)(i) · 4 marks

(b) DNA profiling makes use of the fact that DNA varies between individuals.

(i) When creating a DNA profile, DNA is first extracted from a sample of tissue.

Outline the subsequent steps involved in producing a DNA profile.

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..... **[4]**

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Question 16 · A level Paper 3 · June 2018 · Q4(c)(v) · 2 marks

(v) DNA fragments are separated to produce a DNA profile using electrophoresis.

A student wrote the following description of the electrophoresis procedure:

> We will set up an agarose gel plate and place the DNA samples in the wells at the cathode. Voltage will be passed through the gel for one minute. The gel will then be placed in purified water and we will be able to see the banding pattern of each DNA sample.

Describe **two** changes you would make to the student's procedure and explain how these changes would improve electrophoresis.

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..... **[2]**

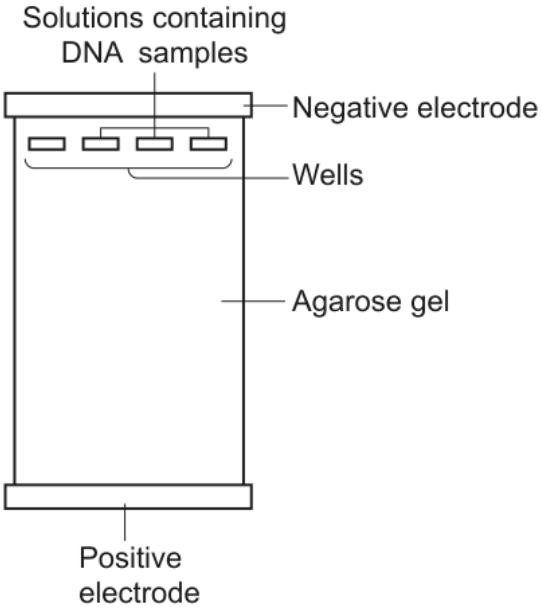
Turn over

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Question 17 · A level Paper 3 · June 2025 · Q4(d)(i) · 4 marks

(d) Electrophoresis is used to separate fragments of DNA so that the separated DNA fragments can be compared and analysed.

(i) The diagram shows an electrophoresis box. Solutions of DNA samples have been added to three of the wells in the box.



State **two other** solutions that should be added to the box before performing electrophoresis **and** explain the purpose of each solution.

Solution .....

Explanation .....

.....

Solution .....

Explanation .....

.....

[4]

Turn over

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Question 18 · A level Paper 3 · June 2025 · Q4(d)(ii) · 2 marks

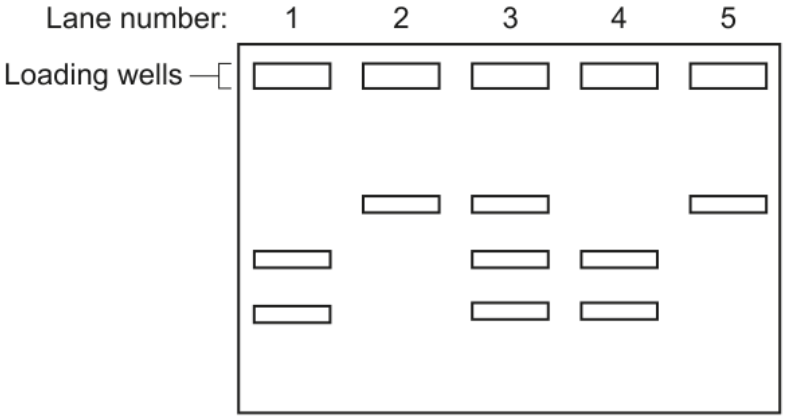
(ii) Electrophoresis can be used to detect disease-causing alleles, such as the mutant *HBB* allele that causes sickle cell disease.

Restriction enzymes are used to produce fragments of *HBB* alleles for analysis by electrophoresis.

These restriction enzymes:

- produce one fragment when the sickle cell *HBB* allele is present
- produce two smaller fragments when the normal *HBB* allele is present.

The diagram shows the results of electrophoresis of DNA samples from five people. It only shows DNA fragments from the *HBB* gene.



State the lane numbers that indicate:

- a genotype that is homozygous for the sickle cell *HBB* allele
- a genotype that is homozygous for the normal *HBB* allele
- a heterozygous genotype for the *HBB* gene.

Homozygous sickle cell .....

Homozygous normal .....

Heterozygous .....

[2]

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Mark schemes

Question 1 · A level Paper 3 · June 2023 · Q2(a)(i) · 1 mark

2	**(a)**	**(i)**	stain(s) / dye(s) ✓ (exposure to) UV light ✓	**1**	2.7	**ALLOW** any named stain

Question 2 · A level Paper 3 · June 2017 · Q1(b)(i) · 1 mark

b	**i**	silica (gel) ✓	**1**	**ACCEPT** aluminium oxide/ alumina/ cellulose/ zirconium oxide / silicon dioxide **DO NOT ACCEPT** paper/ silicon / aluminate **IGNORE** plastic / perspex

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Question 3 · AS Paper 2 · June 2025 · Q1(c) · 2 marks

	(c)	(solvent) <u>dissolves</u> the carbohydrates / acts as <u>mobile</u> phase ✓ (solvent) carries, (dissolved) carbohydrates through <u>stationary</u> phase ✓ different carbohydrates will travel different distances, depending on their solubility in the solvent ✓ different carbohydrates will have, high**er** / low**er**, affinity for solvent / solubility in solvent ✓
		Total

2 max	**2.7**	**IGNORE** general references to separates the carbohydrate(s) – as this relates to the whole process of chromatography, not specifically the solvent **ALLOW** 'solute' for carbohydrate(s) in all MP **IGNORE** 'samples' (in place of carbohydrate) **ALLOW** carries, carbohydrate / solute(s), up the paper **ALLOW** e.g. more soluble sunstances move furth**er ORA** **IGNORE** 'different carbohydrates have different solubilities' unqualified
7		

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Question 4 · AS Paper 2 · June 2018 · Q1(c)(i) · 4 marks

1(c)(i)	*(suggestion(**S**) PLUS reason (**R**) needed)* **1 S** put pencil line / origin / amino acids, higher (than the solvent / 1cm) **+** **1 R** to stop, spots / samples / amino acids, dissolving into / mixing with / touching, solvent ✓ **2 S** put, amino acids / spots / them, further apart / on separate plates **+** **2 R** to stop them, merging / touching / clashing / AW ✓ **3 S** touch plate edges / wear gloves / use forceps / don't touch middle, **+** **3 R** to prevent, contamination / transfer of substances from hands ✓ **4 S** place, lid / cover, over beaker **+** **4 R** to prevent evaporation (of solvent) ✓ **5 S** support the plate / attach plate to beaker **+** **5 R** to keep plate, vertical / still / at constant height ✓ **6 S** use ninhydrin **+** **6 R** to, see / visualise, amino acids ✓ **7 S** repeat and find, mean / average (Rf value) **+** **7 R** to improve, accuracy / check for repeatability / exclude anomalies ✓ **8 S** label, amino acids / spots / samples (in pencil / on beaker) **+** **8 R** to know which is which / avoid confusion ✓

Question	Answer
4 max	**Read all and mark as prose**. **ALLOW** paper / chromatogram / gel, for 'plate' **IGNORE** measure in mm instead of cm **ALLOW** 'or otherwise x would happen' in place of the reason 'to stop x' throughout **ALLOW 1S** ORA less solvent / make solvent lower **OR** make plate / paper, higher **DO NOT ALLOW 1S** pen / permanent marker, line **ALLOW 1R** so only bottom of plate touches solvent **ALLOW 2S** put same distance apart / spread them apart **ALLOW 2R** ORA so they are, distinguishable / clear **ALLOW 3R** amino acids / oils for 'substances' **ALLOW 3R** *idea of* not damaging, stationary phase / silica gel / alumina / AW **ALLOW 4S** close beaker / line beaker with filter paper soaked in solvent **ALLOW 5S** description e.g. use clips / pencil / clamp / rod **ALLOW 5R** ORA to stop plate, tilting / trembling / moving **IGNORE 6S** UV / iodine / permanganate **ALLOW** *'no need, to stain / for ninhydrin, as spots shown up already'* (on Fig. 1.4) **= 1 mark**
Mark	**Guidance**

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Question 5 · AS Paper 2 · June 2018 · Q1(c)(ii) · 2 marks

1(c)(ii)	**1** *answer must lie within this range:* 0.1(0) to 0.15 **AND** *supporting calculation must be shown, e.g:* $\frac{0.65}{4.95}$ (= 0.13) ✓ **2** glutamine ✓	
Question	**Answer**	**Mark**

No mark for figure in correct range unless it also shows the working out of this calculation:

$$\frac{\text{distance from origin to spot}}{\text{distance from origin to solvent front.}}$$

ALLOW figures given in **mm**

ALLOW figures with no unit shown

ALLOW variation in measurements taken **so long as** the final answer falls within the allowed range.

ALLOW mp2 even if no attempt is made at working stage

Guidance

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16	(b)	(i)	**Distance travelled = 91 mm, award 1 mark** distance travelled by most soluble = 91 (mm) ✓ **FIRST CHECK THE ANSWER ON ANSWER LINE** **If answer = 0.83 award 2 marks** R_f = 91 ÷ 109 ✓ = 0.83 ✓	**3**	2.7

Question	Answer	Marks	Element

Question	Answer	Marks	Element

ALLOW +/- 0.5mm for each measured distance (soluble pigment and solvent front)

ALLOW values of 87 **or** 95

ALLOW 2 marks for

Rf value on answer line matches correct distance travelled answer (refer to table) to 2sf

Rf value on answer line correct to 2sf with calculation expressed in cm

e.g. *9.1÷ 10.9 = 0.83*

incorrect soluble pigment distance measured but subsequent calculation correct with answer to 2sf

e.g. *36 ÷ 109 = 0.33*

ALLOW one mark for

incorrect solvent front distance measured but subsequent calculation correct with answer to 2sf

e.g. *91 ÷ 111 = 0.82*

Guidance

incorrect soluble pigment distance measured and incorrect solvent front distance measured but subsequent calculation correct with answer to 2sf

e.g. *73 ÷ 114 = 0.64*

answer not to 2sf but calculation shows correct distances measured for both soluble pigment and solvent front.

e.g. *87 ÷ 109 = 0.798*

Pigment	Solvent front	Rf
86.5	108.5	0.80
86.5	109 / 109.5	0.79
87.0	108.5 / 109	0.80
87.0	109.5	0.79
87.5	108.5	0.81

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Question 7 · AS Paper 1 · June 2023 · Q23(b)(i) · 2 marks

(b)	(i)	**FIRST CHECK ON ANSWER LINE** **If answer = 0.6 award 2 marks** 3.6/6 ✓ = 0.6 ✓

2	**ALLOW** 0.58 – 0.62 3.5/6 = 0.58 3.7/6 = 0.62 **ALLOW** ecf for correct calculation and correct rounding after incorrect measurement of valine position and / or solvent front Allow answers to two decimal points, if more decimal points used max 1 mark

Question 8 · A level Paper 3 · June 2023 · Q2(a)(ii) · 2 marks

2	(a)	(ii)	**FIRST CHECK THE ANSWER ON ANSWER LINE** **If answer = 0.3 award two marks** 2.4/8 ✓ = 0.3 ✓	**2**	2.8

ALLOW 0.29 – 0.31 for 2 marks
2.5/8 = 0.31
2.3/8 = 0.29
Answers given to more than 2 sig figs = max 1

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Question 9 · AS Paper 1 · June 2023 · Q23(b)(ii) · 1 mark

	(b)	**(ii)**	different , polarity / charges / functional groups ✓
			Total

1	**ALLOW** serine more polar ORA **IGNORE** 'chemical properties different' **IGNORE** ref to size (of R group), **IGNORE** ref to solubility (of R group)
4	

Question 10 · A level Paper 1 · November 2020 · Q17(a)(iii) · 2 marks

(a)	**(iii)**	spot 5 ✓ (because) is most soluble in , mobile phase / solvent ✓	**2**	2.3 2.4

ALLOW is less attracted to stationary phase / TLC plate

Question 11 · A level Paper 1 · June 2025 · Q16(b)(ii) · 2 marks

16	**(b)**	**(ii)**		**max 2**	3.3
		1.	allow/ leave, (existing) spot/ extract (at origin), to dry ✓		
		2.	(once dry) add, another spot / more extract, to, X / origin / original spot ✓		
		3.	evaporate (some) solvent from (original) extract ✓		

ALLOW dots for spots

2 ALLOW 'pigment**s**' for 'spots'
2 ALLOW origin line for 'origin'

Question 12 · A level Paper 1 · June 2025 · Q16(b)(iv) · 1 mark

16	**(b)**	**(iv)**	data book used a different , solvent / (TLC) plate / temperature ✓ extract was prepared from different, plant /chloroplasts✓	**max 1**	3.1	
Question			**Answer**	**Marks**	**Element**	**Guidance**

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Question 13 · A level Paper 1 · June 2025 · Q16(b)(v) · 2 marks

16	**(b)**	**(v)**	run known pigments, on same TLC/ using same set up / with same solvent / at same time ✓ compare to position of known pigments **OR** compare colour to, known pigments / colour chart ✓	**2**	3.3	**ALLOW** 'same conditions' for 'same set up' **IGNORE** ref to colorimeter **ALLOW** colour in, data, book/base for 'colour chart'

Question 14 · A level Paper 3 · June 2017 · Q1(c) · 3 marks

c	1. separates by (relative) , adsorption / solubility / interaction with the stationary phase in TLC **and** (separates) by size in electrophoresis ✓ 2. TLC separates non - charged particles **and** electrophoresis (only) separates charged particles ✓ 3.electricity, used for electrophoresis / not used for TLC ✓ 4.buffer solution, used for electrophoresis / not used for TLC ✓ 5.dyes used in TLC **OR** radioactive / fluorescent , tags / nucleotides, used in electrophoresis ✓ *6.Idea of* electrophoresis is , automated / computerised / uses laser scanning (to analyse sequence) / TLC is not automated ✓

3 max	**Read as prose and look for any three correct mp's** **for mp1 and 2** **IGNORE** separates by size of charge on molecule **ACCEPT** mass/ length for size **ACCEPT** electrophoresis uses, current / voltage / charge / (named) electrode(s)

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Question 15 · A level Paper 2 · November 2021 · Q16(b)(i) · 4 marks

16	(b)	(i)	**1** amplify (DNA fragment) with PCR ✓ **2** cut / digest , with restriction enzyme ✓ **3** separate / AW , using, electrophoresis / electric current through gel ✓ **4** transfer (fragments) to , paper / membrane / nylon ✓ **5** add , radioactive / fluorescent , probe ✓ **6** use , x-rays / UV light , to view (position of DNA fragments) ✓

4 max	1.2	**IGNORE** steps in implausible sequence **4 ALLOW** (Southern) blot **6** viewing method must not contradict probe used.

Question 16 · A level Paper 3 · June 2018 · Q4(c)(v) · 2 marks

v	use, alkaline solution /buffer (solution) **AND** Solution carries charge / current (to separate fragments)✓ (use) Southern blotting / described **AND** to transfer fragments to a membrane ✓ use (radioactive / fluorescent) probes / tags / dyes / labels /stains **AND** to , visualise / AW , bands/ patterns ✓	**2 max**
	idea of testing for longer than one minute **or** carrying out preliminary tests to assess the optimum run time **AND** *idea of* (ensures) separation (of DNA fragments / bands) ✓	

Mark first two changes described **ALLOW** to see the position of the fragments

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4	**(d)**	**(i)**	**S1** buffer (solution) ✓ **E1** maintains / controls / for optimum , pH **or** carry / provides ions for , current (through gel) ✓ **S2** reference / marker / ladder , DNA (fragments in solution) ✓ **E2** (to) compare / know / observe / identify , position / mass / size (of sample fragments) ✓ **S3** dye / stain / fluorescent marker / fluorescent tag (solution) ✓ **E3** to , see / monitor progress of , bands / DNA / fragments ✓
Question			**Answer**

4 **max**	3.3 3.4	**Only award an explanation mark when it is paired with the correct solution** *mark as prose* **IGNORE** a stated pH value **ALLOW** AW, e.g. 'allows charge to pass across (gel)' **ALLOW** DNA fragments of known , length / mass / size **IGNORE** probe / UV **ALLOW** radioactive marker / colour marker / colour tag **ALLOW** named dyes, e.g. ethidium bromide
Mark	**AO Element**	**Guidance**

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Question 18 · A level Paper 3 · June 2025 · Q4(d)(ii) · 2 marks

4	**(d)**	**(ii)**	homozygous sickle cell = 2 and 5 ✓ homozygous normal = 1 and 4 **AND** heterozygous = 3 ✓	**2**	3.1

ALLOW 1 mark for:
homozygous sickle cell = 2 **or** 5
and
homozygous normal = 1 **or** 4
and
heterozygous = 3

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