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4 **EUROPEAN OLYMPIAD OF**
EXPERIMENTAL SCIENCE
LUXEMBOURG

Task 1

MARKING SCHEME

CANCER

The origins of a genetic disorder

EOES 2024, 09.04.2024

Team (Country + A/B) _____

Students: _____

Problem 1 – Quantification of Fe²⁺ ions (27 points)

General information for all calculations in Task 1 Problem 1:

-50% on the mark if units are missing

Step 1: Generating a calibration curve for Fe²⁺ in mg/L

- **Table 1.1.1.:** Fill in the following table and detail your calculations for the first line. (4P + 2P)

$\beta(\text{Fe}^{2+})$ (mg/L)	V(A) (mL)	V(B) (mL)	V(H ₂ O) (mL)	V(buffer) (mL)	V(o-phenanthroline) (mL)
12.0	1.20	X	2.80	5.00	1.00
10.0	1.00	X	3.00	5.00	1.00
5.00	0.50	X	3.50	5.00	1.00
1.50	X	3.00	1.00	5.00	1.00
1.00	X	2.00	2.00	5.00	1.00
0.500	X	1.00	3.00	5.00	1.00
0.250	X	0.50	3.50	5.00	1.00
0	X	0	4.00	5.00	1.00

Detailed calculation for the first line:

$\beta_A = 100 \frac{\text{mg}}{\text{L}}$ $\beta_{\text{sol1}} = 12.0 \frac{\text{mg}}{\text{L}}$ $V_{\text{sol1}} = 10.0 \text{ mL}$ $V(A) = \frac{\beta_{\text{sol1}} \cdot V_{\text{sol1}}}{\beta_A} = \frac{12.0 \frac{\text{mg}}{\text{L}} \cdot 10.0 \text{ mL}}{100 \frac{\text{mg}}{\text{L}}} = 1.20 \text{ mL (1 pt)}$ $V(\text{H}_2\text{O}) = V_{\text{sol1}} - V(\text{o-phenanthroline}) - V(\text{buffer}) - V(A)$ $= 10.0 \text{ mL} - 1.00 \text{ mL} - 5.00 \text{ mL} - 1.20 \text{ mL} = 2.80 \text{ mL (1 pt)}$	Marks
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Step 2: Colorimetric determination of the concentration of an Fe²⁺ solution

- **Table 1.2.1.: Measured absorbances A at λ = 492 nm (3P)**

The coefficient of determination (R²) is used to evaluate the predictive accuracy of the calibration curve using the generated samples. Values with A>2.0 must be excluded from the calculation of R²

$$0.9990 \leq R^2 \leq 1 \quad (3\text{pts})$$

$$0.9980 \leq R^2 < 0.9989 \quad (2\text{pts})$$

$$0.9970 \leq R^2 < 0.9979 \quad (1\text{pts})$$

$\beta(\text{Fe}^{2+})$ (mg/L)	A
12.0	
10.0	
5.00	
1.50	
1.00	
0.500	
0.250	
0	

- **Question 1.2.2: Measured absorbance A at λ = 492 nm for sample F3**

OWN MEASUREMENT

- **Graph 1.2.3: Draw a calibration graph (plot the absorbance against the mass concentration) on graph paper. (4P for the graph)**

(You will only receive 1 sheet of graph paper. You can ask for a second one, but this will cost you 0.5 points).

Possible penalty points:
Signature of a supervisor:

1 pt: labelling of axes (A (492nm) / Fe²⁺ (mg/L))

1 ps: regression line (placement of the regression line on the graph (eye best fit), must not consider values A>2)

2 pts: precision of the graph (spacing on the axes (1pt) and positioning of the points (within 0.5mm of true value; -0.25 pt for each error)

- **Question 1.2.4.: Determine the mass extinction coefficient (ϵ_m) from the graph and calculate the molar extinction coefficient (ϵ). Write your calculation details in the box below and add your details to the graph (1.2.3.). (3P)**

(! For the calculations in points 1.2.4 to 1.2.8, indicate your final results using the scientific notation with 2 decimal places (example: $1.23 \cdot 10^{-5}$)

<p>1 pt: determination of the slope of the regression line (!Absorption values over 2.0 should be excluded!)</p> $A = \epsilon_m \cdot b \cdot \beta \quad \epsilon_m = \text{mass absorption coefficient (L} \cdot \text{mg}^{-1} \cdot \text{cm}^{-1}\text{)}$ <p>β = concentration in mg/L</p> <p>slope = $\epsilon_m \cdot b$ (0.5 pt) ϵ = molar absorption coefficient (L\cdotmol$^{-1}$$\cdotcm^{-1}$)</p> $\epsilon_m = \frac{\text{slope}}{b}$ (0.5 pt) M = molar mass of the absorbing species (55.85g/mol) $\epsilon = \epsilon_m \cdot 1000 \cdot M$ (1 pt) <p>(1 point for ϵ given if within 5% of true value, 0.5pt given if within 8%)</p>	<p>Marks</p>
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- **Question 1.2.5.: Calculate the molar extinction coefficient using the Lambert-Beer Law. Show your calculation details. (2P)**

<p>Chosen point must be located on the regression line or as close as possible to the regression line (1 pt)</p> $c = \frac{\beta \cdot 1000}{M}$ (0.5 pt) $A = \epsilon \cdot b \cdot c \Leftrightarrow \epsilon = \frac{A}{b \cdot c}$ (0.5 pt)	<p>Marks</p>
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- **Question 1.2.6: Determine the mass concentration of the unknown sample solution F3 (β_{F3}) graphically from the calibration curve (Graph 1.2.3.) and calculate its molar concentration (c_{F3}). Show details on the graph paper and write the mass concentration and the calculation details for c_{F3} in the box below. (2P)**

<p>0.5 pt : details correctly shown on graph paper 1pt: value of β_{F3} 0.5 pt : value of c_{F3}</p>	<p>Marks</p>
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- **Question 1.2.7: Calculate the molar concentration of the unknown sample solution F3 (c_{F3}) using the molar extinction coefficient! Show your calculation details. (2P)**

$A = \varepsilon \cdot b \cdot c \Leftrightarrow c = A/(\varepsilon \cdot b)$ (1 pt)	Marks
(molar extinction coeff. determined in 1.2.4. should be used) 1 pt : value of c_{F3} .	

- **Question 1.2.8: Calculate the corresponding molar concentrations c_{F2} and c_{F1} of the solutions F2 and F1. Show your calculation details. (5P)**

$c_{F3} = \text{result 1.2.8.}$	Marks
$V_{F3,F} = 50 \text{ mL}$	
$V_{F2,P} = 5 \text{ mL}$ correct values: 0,5 pt	
$c_{F2} \cdot V_{F2,P} = c_{F3} \cdot V_{F3,F}$ (0,5 pt)	
$\Leftrightarrow c_{F2} = \frac{c_{F3} \cdot V_{F3,F}}{V_{F2,P}} = \frac{c_{F3} \cdot 50 \text{ mL}}{5 \text{ mL}}$ (1 pt)	
$c_{F2} = \text{previous result}$	
$V_{F2,F} = 100 \text{ mL}$	
$V_{F1,P} = 7 \text{ mL}$ correct values: 0,5 pt	
$c_{F1} \cdot V_{F1,P} = c_{F2} \cdot V_{F2,F}$ (0,5 pt)	
$\Leftrightarrow c_{F1} = \frac{c_{F2} \cdot V_{F2,F}}{V_{F1,P}} = \frac{c_{F2} \cdot 100 \text{ mL}}{7 \text{ mL}}$ (1pt)	
Accuracy of the result for c_{F1} based on % deviation from the true value.	
1 pt: within 5% of reference	
0.5 pt: within 8% of reference	

Problem 2 – Solve the carcinogen chaos (23 points)

- **Table 2.1.1.: Fill in the following table with your observations for the CAN test (2.5P)**

substance	Observation: formation of a red complex? Use the following symbols: ✓ → Yes and X → No
control	✓ (0.5 pt)
X1	✓ (0.5 pt)
X2	X (0.5 pt)
X3	X (0.5 pt)
X4	✓ (0.5 pt)

- **Table 2.1.2.: Fill in the following table with your observations for the FeCl₃ test (2.5P)**

substance	Observation: colour of the solution changes to blue/purple/lilac ? Use the following symbols: ✓ → Yes and X → No
control	✓ (0.5 pt)
X1	✓ (0.5 pt)
X2	X (0.5 pt)
X3	X (0.5 pt)
X4	X (0.5 pt)

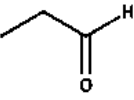
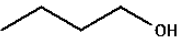
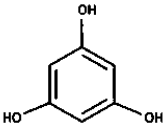
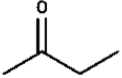
- **Table 2.1.3.: Fill in the following table with your observations for the Brady test (2.5P)**

substance	Observation: formation of a yellow to red precipitate? Use the following symbols: ✓ → Yes and X → No
control	✓ (0.5 pt)
X1	X (0.5 pt)
X2	✓ (0.5 pt)
X3	✓ (0.5 pt)
X4	X (0.5 pt)

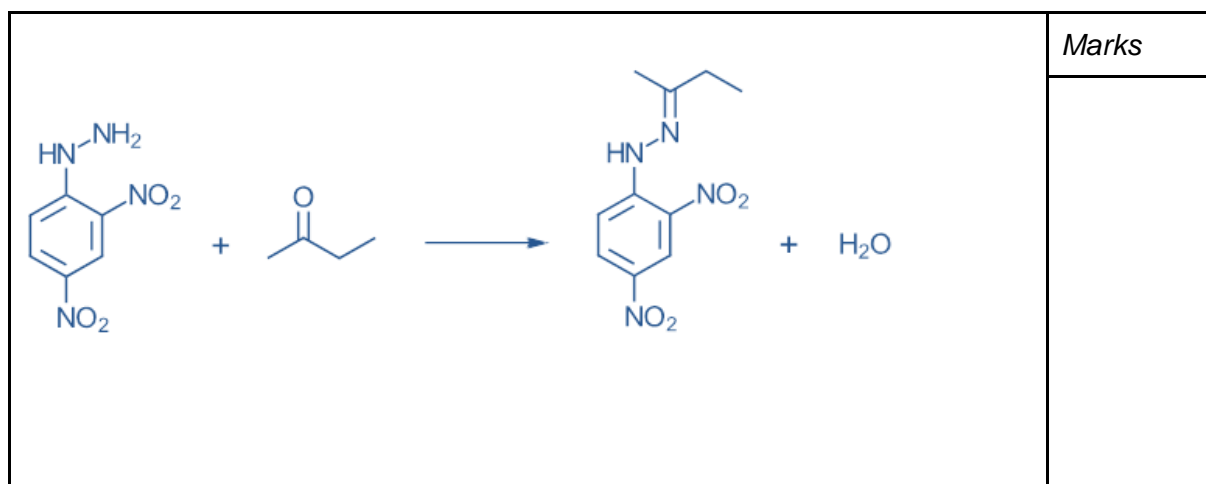
- **Table 2.1.4.: Fill in the following table with your observations for the Fehling test (2.5P)**

substance	Observation: formation of a brick red precipitate? Use the following symbols: ✓ → Yes and X → No
control	✓ (0.5 pt)
X1	X (0.5 pt)
X2	✓ (0.5 pt)
X3	X (0.5 pt)
X4	X (0.5 pt)

- **Table 2.1.5.: Assign the unknown substances to their correct labels (X1 – X4).**

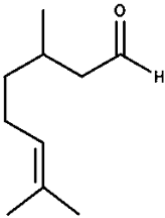
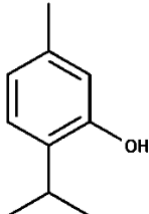
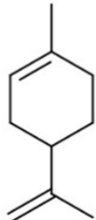
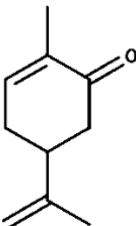
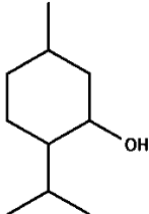
substance	Correct label
	X2
	X4
	X1
	X3

- **Question 2.1.6.: Formulate the reaction scheme for the reaction of the given ketone with the Brady reagent (2P).**



- **Table 2.1.7.: Do the presented natural fragrances show a reaction with the presented tests?**

Use the following symbols: ✓ → Yes and X → No (3P)

	CAN test	FeCl ₃ test	Brady test	Fehling test
citronella 	X	X	✓	✓
thymol 	✓	✓	X	X
limonene 	X	X	X	X
carvone 	X	X	✓	X
menthol 	✓	✓	X	X

0.15 pts for each correct mark

Problem 3: Ionizing radiation

Problem 3.1: Evidence for the existence of radon (23 points)

- **Table 3.1.1.: Background activity A_0 (1P)**

Measurements			Mean
A_{01} (counts/min)	A_{02} (counts/min)	A_{03} (counts/min)	A_0 (counts/min)
52	61	50	54

A_0 between 20 and 70: 1 point, otherwise 0 point.

- **Table 3.1.2.: Activity as a function of time (8P)**

Measurements				Mean	Effective activity
t (min)	A_1 (counts/min)	A_2 (counts/min)	A_3 (counts/min)	A_{mes} (counts/min)	$A=A_{mes}-A_0$ (counts/min)
0	284	275	282	280	226
15	256	259	250	255	201
30	222	186	216	208	154
45	154	111	170	145	91
60	166	142	143	150	96
75	129	151	120	133	79
90	135	109	110	118	64
105	91	95	103	96	42
120	80	84	89	84	30
135	69	78	74	74	20
150	65	85	66	72	18

You can "buy" measurement data of table 3.1.2. for 3 penalty points. Ask a supervisor!

0.5 point less for every row that is missing; 0.5 point less for every error in the calculated values

○ **Graph 3.1.3.: Activity as a function of time (5 P)**

On a sheet of graph paper, create a plot of the activity A as a function of time t for your balloon. You will only receive one sheet of graph paper for this plot. You can ask for a second one but this will cost you 0.5 points.

Possible penalty points:
Signature of a supervisor:

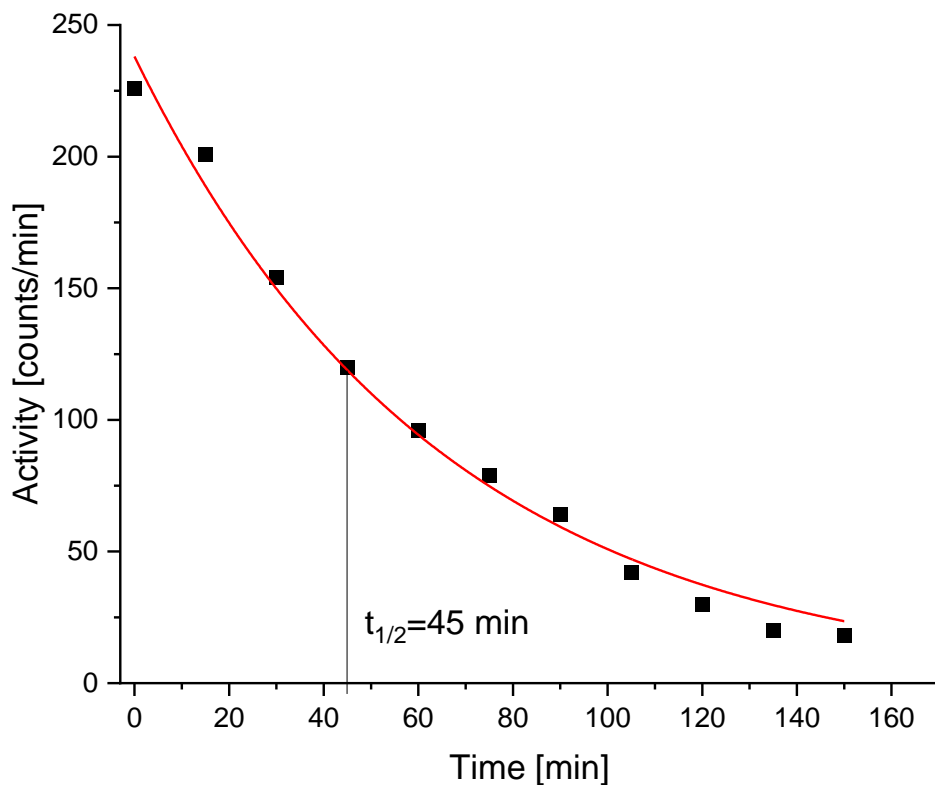
Too small (less than 0.5 page): -1

No labels: -0.5 per label; Units on the axis missing: -0.5 per axis

Data wrongly inserted: -2

Trendline wrong (e.g. no exponential form): -2

Graph G1



○ **Question 3.1.4. (3P)**

Half-life $t_{1/2}$
45 min

3 points if $t_{1/2}$ between 35 min and 55 min

2 points if $t_{1/2}$ between 30 min and 35 min or between 55 min and 60 min

1 point if $t_{1/2}$ between 25 min and 30 min or between 60 min and 65 min

- **Question 3.1.5. Tick (✓) the cell(s) under the different nuclei. (2P)**

Po-218	Pb-214	Bi-214	Po-214	Pb-210	Bi-210	Po-210	Pb-206
	✓	✓					

Every wrong or missing tick: -1

- **Question 3.1.6. (2P)**

Balloon with diameter	d ₁	d ₂	d ₃
Number of counts/min	100	310	1150

1 point less for every mistake. Values must be precise!

- **Question 3.1.7. Tick (✓) the cell(s) under the different nuclei. (2P)**

Po-218	Pb-214	Bi-214	Po-214	Pb-210	Bi-210	Po-210	Pb-206
				✓	✓	✓	✓

Every wrong or missing tick: -1

Problem 3.2: Law of distance (15 points)

○ **Table 3.2.1.: Dark current (1 P)**

Current intensity in the dark: $I_0 = 60 \mu\text{A}$

○ **Table 3.2.2.: Intensity as a function of distance (6P)**

d (cm) Distance between lamp and phototransistor	I (mA) Intensity of current through phototransistor	I_L (mA) Intensity of current due to lamp light	$I_L \cdot d^2$ (mA \times cm 2)
20	277	221	88400
22	221	165	79860
24	183	127	73152
26	159	103	69628
28	144	88	68992
30	131	75	67500
32	120	64	65536
34	113	57	65892
36	107	51	66096
38	100	44	63536
40	95	39	62400
42	90	34	59976
44	87	31	60016
46	84	28	59248

48	81	25	57600
50	80	24	60000
52	78	22	59488
54	76	20	58320
56	75	19	59584
58	74	18	60552
60	73	17	61200
62	72	16	61504
64	71	15	61440
66	70	14	60984
68	69	13	60112
70	68.5	12.5	61250
72	68	12	62208
74	67	11	60236
76	66	10	57760
78	65.5	9.5	57798
80	65	9	57600

○ **Graph 3.2.3.: Intensity as a function of distance (3P)**

On a sheet of millimeter graph paper create the plot of I_L as a function of the distance d . Draw a trendline through the measurement points in Graph 3.2.3. but only through the points that in a first approximation fulfill the quadratic law of distance. You will only receive one sheet of graph paper for this plot. You can ask for a second one but this will cost you 0.5 points.

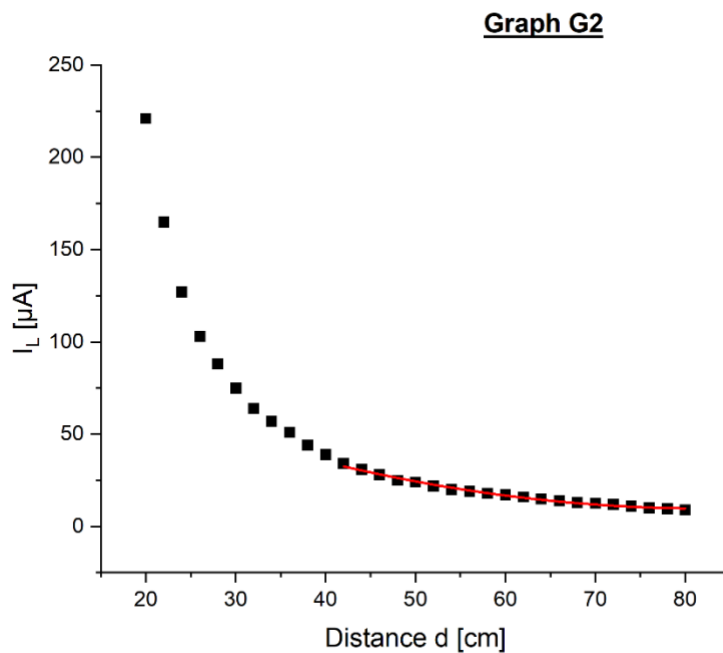
Possible penalty points:
Signature of a supervisor:

Too small (less than 0.5 page): -1

No labels: -0.5 per label; Units on the axis missing: -0.5 per axis

Data wrongly inserted: -2

Trendline wrong: -1

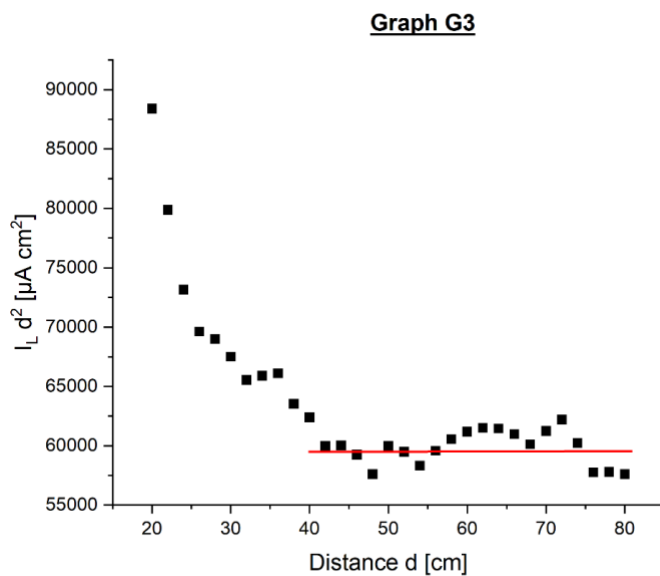


○ **Graph 3.2.4.: $I_L \cdot d^2$ as a function of distance (3P)**

On a sheet of millimeter graph paper create the plot of $I_L \cdot d^2$ as a function of the distance d . Draw a trendline through the measurement points in Graph 3.2.4. but only through the points that in a first approximation fulfill the quadratic law of distance. You will only receive one sheet of graph paper for this plot. You can ask for a second one but this will cost you 0.5 points.

Possible penalty points:
Signature of a supervisor:

- Too small (less than 0.5 page): -1*
- No labels: -0.5 per label; Units on the axis missing: -0.5 per axis*
- Data wrongly inserted: -2*
- Trendline wrong: -1*



○ **Question 3.2.5. ✓ → Yes (2P)**

If instead of a point-like source, you were to use an extended light source. Which of the following statements would be true? Tick (✓ → Yes) the correct cells!

The intensity would decay slower than with a point-like source	✓
The intensity would decay faster than with a point-like source	
The decay of the intensity is the same as for a point-like source	

Every wrong or missing tick: -1

Problem 3.3.: Absorption of radiation (12 points)

- **Table 3.3.1.: Intensity as a function of the number of plates (3P)**

Incorrect measurement values: -1 per value

Incorrect calculus: -1 per error

N Number of plates	I (mA) Intensity of current through phototransistor	I_L (mA) Intensity of current due to lamp light
0	1251	1196
1	1099	1044
2	985	930
3	856	801
4	807	752
5	751	696
6	670	615
7	625	570
8	585	530
9	545	490
10	505	450

○ **Graph 3.3.2.: Intensity as a function of the number of plates (3P)**

On a sheet of graph paper, create a plot of I_L as a function of N , insert the extrapolation for the determination of $N_{1/2}$. You will only receive one sheet of graph paper for this plot. You can ask for a second one but this will cost you 0.5 points.

Possible penalty points:
Signature of a supervisor:

Too small (less than 0.5 page): -1

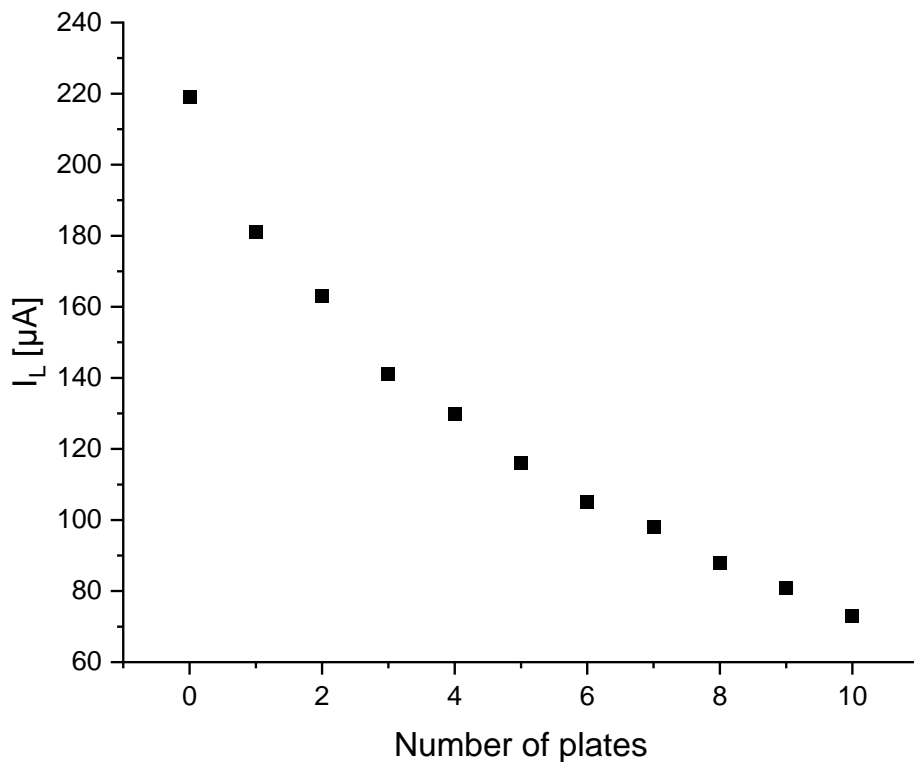
No labels: -0.5 per label; Units on the axis missing: -0.5 per axis

Data wrongly inserted: -2

Trendline à wrong: -1

$N_{1/2}$ wrong inserted: -1

Graph G4



○ **Question 3.3.3. (1P)**

$N_{1/2}$

1 point if the value is between 5, 6 or 7; 0.5. points if the value is 4 or 8, 0 points otherwise

○ **Question 3.3.4.: (3P)**

Sort the materials from 1 to 5 by how strongly they absorb radiation. Mark the best absorber with 1 and the worst with 5.

Iron	2
Cardboard	4
Lead	1
Glass	3
Air	5

-1 per error

○ **Question 3.3.5. Tick (✓) the correct cell(s). (2P)**

Imagine that a material for shielding radioactive radiation has a thickness $D_{1/2} = 2$ cm for absorbing half the radiation. Which of the following thicknesses is sufficient to reduce the radiation to less than 5% of its initial value?

8 cm	
9 cm	
7 cm	
10 cm	

Every wrong or missing tick: -1

Problem 4 – Effect of UV light on cell growth

(16 points)

4.1 Experimental set-up and UV exposure

- **Question 4.1.1.: Show your steps of the theoretical calculation. Round to one decimal place. (2P)**

<p style="color: green;">Initial Volume* Initial OD = Final Volume * Final OD $X \cdot 0.9 = 20 \cdot 0.2$ $X = (20 \cdot 0.2) / 0.9$ $X = 4.5 \text{ mL}$</p> <p style="color: green;">+1p if intermediate calculation step is shown. +0.5p if result is correct. +0.25P if rounded to one decimal place. +0.25p if unit is present.</p>	<p>Marks</p>
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- **Question 4.1.2: Measured OD before the treatment:**

4.2. Growth analysis by determination of OD600nm

- **Table 4.2.1.: Fill in the table (4 P)**

	0'	30'	60'	90'	Marks
OD sample 1					
OD sample 2					
OD sample 3					
OD sample 4					
				Total marks	

-0.25p if not rounded to 3 decimal places.

+1p The values of sample 1,3 & 4 need to increase after every 30 minutes

-0.5p if a value is higher than the next measurement in that sample. Applies for samples 1,3 & 4

+1p Sample 2 at 30' needs to be the lowest value at the timepoint AND cannot be more than +-10% of the value of sample2 at the 0' measurement.

For outliers -0.5p All values of sample 2 cannot deviate more than +-30% of the value from time point 0'

+1p value of Sample 4 at 30' needs to be 25% lower than the value of Sample 3 at 30'. A difference of max +-15% of that calculated value is allowed.

+1p Taking the value of sample 1 at 90' as reference. The measured value for sample 3 needs to be in the range defined as: value of Sample 1 at 90' +-15%.

- **Graph 4.2.2.: Using the graph paper provided draw the four different growth curves. (8 P)**

Addition graph paper can be purchased but costs 0.5 P

Possible penalty points: Signature of a supervisor:
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+4p Points all drawn correctly. 0.25p per correct data point drawn. Total :16 data points, per mistake (-0.5P)

+1p Line points equally spaced (0.25p for each axis)

+1p if points are connected. No points are awarded if points are not connected or a line of best fit is drawn.

+0.5 p Legend present

+0.5p Both axes start at 0

+0.5p Correct size (at least half the page)

+0.5p Absorbance plotted on the Y axis, time on the x axis

-2p if graph has no group sticker on it i.e there is no sticker identifying to which team it belongs to.

-1p lack of numbers on the axis

- **Question 4.2.3.: Optical density (1 P)**

Letter (A, B, C, D)	Marks
A	

Why is the optical density (OD) measured at 600 nm? More than 1 correct answer may be possible.

A - The wavelength minimizes damage to the bacteria

B - The wavelength favours the growth of bacteria

C - A lower wavelength would not penetrate the solution

D - 600nm corresponds to the absorbance of proteins

-1p for a wrong answer

○ **Question 4.2.4.: Sun protection factor (1 P)**

Letter (A, B, C, D, E)	Marks
B	

What does SPF 50 mean? More than 1 correct answer may be possible.

A - The skin is completely protected from UV radiation for 50 minutes

B - It allows only 2% of UV to pass through

C - It is the max sun protection we can use

D - It allows 50% UV to pass through after 1 hour

E - 50 corresponds to the concentration of titan dioxide

Problem 5 – Effect of UV exposure on genetic material (34 points)

5. Effect of UV exposure on genetic material

5.1. Cell counting

○ **Question 5.1.1.: Counting slide set up (1P)**

0.5 penalty points for using a 2nd try

<table border="1" style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 50%; text-align: center; padding: 5px;">1st try</td> <td style="width: 50%; text-align: center; padding: 5px;">2nd try</td> </tr> <tr> <td style="height: 100px;"></td> <td style="height: 100px;"></td> </tr> </table>	1st try	2nd try			<table border="1" style="width: 100%; border-collapse: collapse;"> <tr> <td style="text-align: center; padding: 5px;">Validated</td> </tr> <tr> <td style="height: 100px;"></td> </tr> </table>	Validated		<p><i>Marks</i></p>
1st try	2nd try							
Validated								

Orientation of cover slip vertical horizontal?

Addition of water too much water

1p If stamp at first try and validated

0.5p if stamp at first try, second try and validated

0p if stamp at first try, second try but NOT at validated. i.e. needed more than two tries.

○ **Question 5.1.2.: Picture of counting slide.**

if no stamp present, then only a maximum of 1.5 P possible for 5.1.3

<table border="1" style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 50%; text-align: center; padding: 5px;">Stamp</td> <td style="width: 50%; text-align: center; padding: 5px;">Time:</td> </tr> <tr> <td style="height: 100px;"></td> <td style="height: 100px;"></td> </tr> </table>	Stamp	Time:			<p><i>Marks</i></p>
Stamp	Time:				

Verification of cell loading quality onto the counting slide (photo taken)

If not checked by the supervisor: -1.5P marks on 5.1.3

- **Table 5.1.3.: Report your cell counting results in the table below. Round to one decimal place for the average. (3P)**

Count	Cells per grid 1	Cells per grid 2	Cells per grid 3	Cells per grid 4	Cells per grid 5	Average	Marks
Number of living cells							
Number of dead cells							
Total marks							

+1p for the correct number of living cells in all 5 grids. Number of cells needs to match the number of cells on the picture taken in 5.1.2.

+1p for the correct number of dead cells in all 5 grids. Number of cells needs to match the number of cells on the picture taken in 5.1.2

-0.5p per grid if value of participant does not match the number of cells on the picture taken in 5.1.2.

1p for correct calculated averages+-25% . 0.5p per average.

-0.25p if not rounded to one decimal place

- **Question 5.1.4.: What is the percentage of living cells? Round to one decimal place. (2P)**

+1p for correct result +1p if all calculation steps are shown Total of living/Total number of counted cells * 100 (Calculations are based on participants actual data) -1p if unit (%) is missing -0.25 p If decimal place is missing	Marks

- **Question 5.1.5.: What is the concentration of living cells in your tube “HC”? Round to two decimal places (5P)**

Average number of living cells +1p Multiply by two due to the dilution by trypan blue +0.5p Determine the area $0.25 \times 0.25 = 0.0625 \text{mm}^2$ +0.5p Determine the volume $0.0625 \times 0.1 = 0.00625 \text{mm}^3$ +1p Convert mm^3 to mL 0.00000625mL 6.25×10^{-6} +1p Divide by $6.25 \times 10^{-6} \rightarrow \text{cells/mL}$ (1P) +1p Unit present -0.25p if not rounded to two decimal places	Marks

- **Question 5.1.6.: What is the total number of living cells in your tube “HC”? Round to two decimal places (1P)**

+1p Previous calculated value x total amount of liquid (0.75 mL) -0.5P if calculations are not shown -0.25P if units are missing -0.25p if not rounded to two decimal places	Marks

5.2. Extraction of genetic material

- **Question 5.2.1.: What is the role of the PM solution? More than 1 correct answer may be possible (1P)**

Letter (A, B, C, D)	Marks
A	

A - To break down the cell membrane of the bacteria

B - To uncoil the DNA for the next step of the procedure

C - To prevent any damage to the DNA during the heating process

D - To amplify the DNA

-1p if answer is not correct

- **Table 5.2.2.: Write down the DNA concentration and OD260/OD280 ratio measured with the help of the Nanodrop (3 P)**

DNA concentration 1p	OD260/OD280 ratio 2p	Stamp & signature of lab assistant
1 value needs to be over 10ng or under 100ng otherwise dilution was not done properly (0.25P)	Between 1.6 and 2.1 (0.5P)	
2 value needs to be over 10ng or under 100ng otherwise dilution was not done properly (0.25P)	Between 1.6 and 2.1 (0.5P)	
3 value needs to be over 10ng or under 100ng otherwise dilution was not done properly (0.25P)	Between 1.6 and 2.1 (0.5P)	
4 value needs to be over 10ng or under 100ng otherwise dilution was not done properly (0.25P)	Between 1.6 and 2.1 (0.5P)	
Total marks		

5.3 Preparation of samples for PCR

○ **Table 5.3.1.:**

Calculate the volume of DNA and water that is required to have 400 ng of DNA in a 20 μL solution. Measure the DNA concentration afterwards using the Nanodrop. Round to one decimal place (4P)

	Required DNA volume (μL) <i>1.5 p</i>	Required water volume (μL) <i>0.5p</i>	Measured DNA concentration ($\text{ng}/\mu\text{L}$) <i>2 p</i>	Stamp & Signature of lab assistant
Sample 1	<i>Volume of DNA : 400/measured concentration</i>	<i>Volume of water: 20-calculated volume of DNA</i>		
Sample 2	<i>Volume of DNA</i>	<i>Volume of water:</i>		
Sample 3	<i>Volume of DNA</i>	<i>Volume of water:</i>		
Sample 4	<i>Volume of DNA</i>	<i>Volume of water:</i>		
Total marks				

+1.5p if all calculations in the "required DNA volume" column are correct

-0.5p for each mistake in the "required DNA volume" column

+0.5p if all calculations in the "required water volume" column are correct

-0.25p for each mistake in "required water volume" column

-0.25 p if not rounded to one decimal place.

5.4. Preparation for gel electrophoresis

○ **Question 5.4.1.: What's the role of the loading dye? More than 1 correct answer may be possible (1 P)**

A - Make the sample visible in the gel

B - Facilitate the entry of the DNA in the agarose gel

C - It's a DNA staining dye

D - Keep the DNA at the bottom of the gel wells

E - Protect the DNA from the electrical current

-0.5p if answer is not correct

Letter(s) (A, B, C, D, E)	Marks
<i>A & D</i>	

○ **Question 5.4.2.: Loading of the gel electrophoresis (4P)**

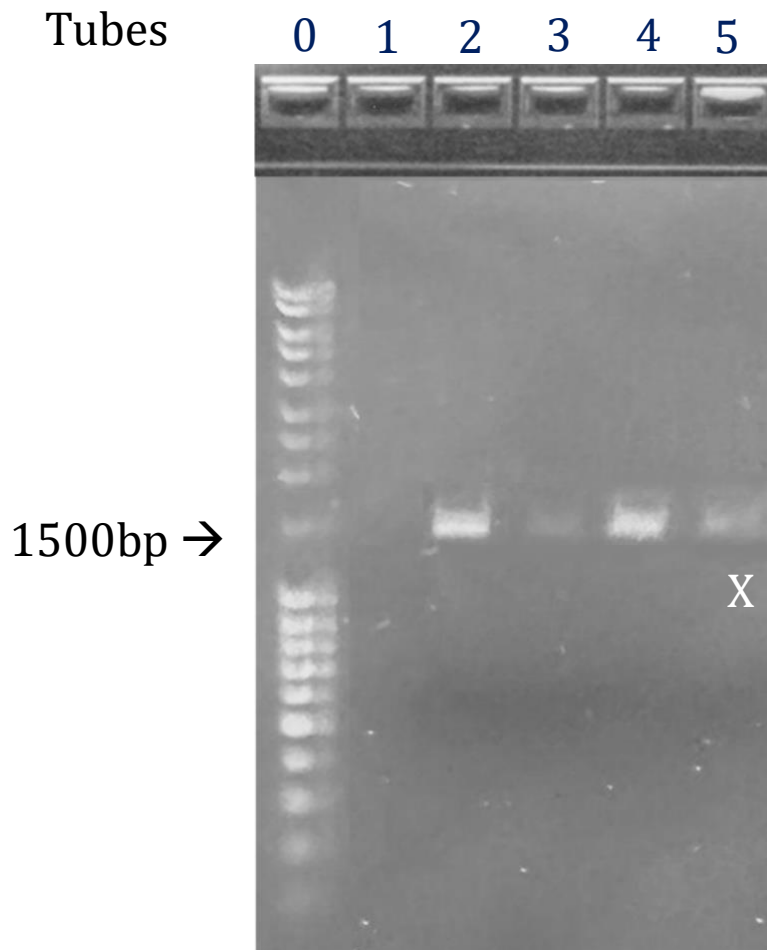
<p>6 Slots filled (check presence of dye (1) Order of slots correct (1P) Quality of results: Results as expected (2P)</p>	<p>Stamp Start Time: Gel electrophoresis has been started.</p>	<p>Marks</p>
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5.5. Analysis of PCR results

○ **Question 5.5.1.: Gel migration drawing (5P)**

<p>+1 P Correct placement of band in relation to Ladder +2 P Correct intensity of bands of control samples (+&-) (1+1) +3P Correct intensity of band for experimental samples -Intensity of sunscreen similar to intensity of + control (1P) - 20'UV + no lotion needs to be lowest signal (0.5p) - body lotion needs to be middle (0.5P)</p>	<p>Stamp Gel drawing has been given to the supervisor</p>	<p>Marks</p>
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PHOTO OF THE GEL ELECTROPHORESIS STUDENTS WILL GET AFTER HANDING IN 5.5.1. – EXPECTED PCR RESULTS



- **Question 5.5.2.: What will happen to the signals on the gel if you increase the volume of starting material in step 2, Problem 4.2? More than 1 correct answer may be possible (1P)**

A - The signals on the gel will appear similar to the one you observe on the official result

B - The signals on the gel will appear stronger

C - The signals on the gel will appear lower

D - It will depend on the volumes used

E - It will depend on the cell concentration

-1 p for incorrect answer

Letter (A, B, C, D, E)	Marks
A	

- **Question 5.5.3.: What will be observed in position X if you expose the bacteria for 40 minutes to UV radiation? More than 1 correct answer may be possible (1P)**

A - The signal on the gel will appear similar to the one you observe on the official result

B - The signal on the gel will appear stronger

C - The signal on the gel will appear lower

-1 p for incorrect answer

Letter (A, B, C)	Marks
C	

- **Question 5.5.4.: What could be observed for sample 3 if we would have used a sunscreen with a lower SPF such as SPF 15? More than 1 correct answer may be possible (1P)**

A - The signal on the gel would have appeared similar to the one you observe on the official result

B - The signal on the gel would have appeared stronger

C - The signal on the gel would have appeared lower

D - The signal on the gel would have completely disappeared

E - It depend on the brand of the sunscreen

-1 p for incorrect answer

Letter (A, B, C, D, E)	Marks
C	

- **Question 5.5.5.: How could you explain the result observed with sample 2?**
More than 1 correct answer may be possible (1P)

A - UV radiation induce unspecific mutations on the whole DNA strain preventing any recognition of the DNA sequence by the PCR primers

B - UV radiation induce nucleotide dimer formation preventing DNA reading by polymerase

C - UV radiation induce high denaturation of the cell DNA preventing DNA polymerisation

D - UV radiation depolymerize the DNA sequence

E - UV radiation impair the cell division

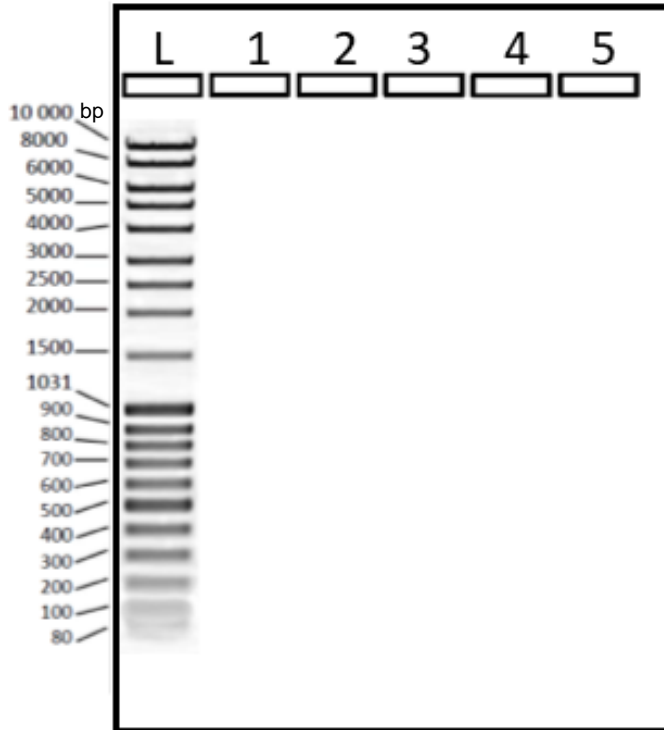
-0.5 p for incorrect answer

Letter (A, B, C, D, E)	Marks
<i>B & E</i>	

APPENDIX - 5.5.1. Expected PCR results

EXPLANATIONS FOUND BEFORE UNDER 5.5.1.

Draw the expected result of the gel electrophoresis.



Legend:

- L: Ladder**
- 1: Negative control without DNA**
- 2: sample 1 (no UV exposure)**
- 3: sample 2 (20 min UV exposure)**
- 4: sample 3 (sunscreen protection + 20 min UV exposure)**
- 5: sample 4 (body lotion + 20 min UV exposure)**

Intensity of the signal

Use the following notation to indicate the intensity of the signal/bands. The fewer diagonal lines there are in the box the weaker the signal.

