

1 Sample exam questions: **Dean Unit, Biology 2290F/G** (Answers start on page 6)
 (These are **sample** exam questions. There are more questions than on the exam)

Question 1: *Chlorella* is a unicellular alga that grows well in liquid culture media. Using a haemocytometer, the cell density in a *Chlorella* culture was determined to be 5.625×10^7 cells/ml.

To make a standard curve of its absorbance at 750 nm (A_{750}) against the number of cells per milliliter, the culture was diluted **15-fold**. As shown in the table below, this diluted sample gave an A_{750} reading of 0.390. Complete the table to show (i) the number of cells per milliliter (#cells/ml) that correspond to each A_{750} reading, and (ii), the volumes of diluted culture and water in the blank spaces in columns 1 and 2 of the table. All tubes contain a total volume of 5.0 ml (**4 marks**)

<u>ml diluted culture</u>	<u>ml water</u>	<u>#cells/ml</u>	<u>A_{750}</u>
5.0	0		0.390
			0.234
			0.117
			0.039

Question 2: The following table shows the contents of the experimental reaction mixture used to determine the rate of photoreduction of DCPIP at 25 cm from a 100 watt lamp.

<u>Tube #</u>	<u>2×10^{-4} M DCPIP (ml)</u>	<u>Reaction buffer pH 7.0 (ml)</u>	<u>Distilled water (ml)</u>	<u>Chloroplast preparation (μl)</u>
1. Experimental	2.0	2.0	6.0	*
2. Dark Control				*
3. Blank				*

(* volume to be determined)

(a) Complete the table to show the contents of tubes numbered 2 and 3 (**2 marks**)

Chloroplasts were isolated according to the Biology 2290 protocol. Fifty (50) μ l of this preparation was added to 5.0 ml of 80% (v/v) acetone, the tube was shaken to dissolve the chlorophyll and centrifuged at $1300 \times g$ for 3 minutes. The supernatant was transferred to a cuvette and its A_{652} was determined to be **0.720**.

(b) What is the appropriate blank for this A_{652} determination?

(1 mark)

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The concentration of chlorophyll in the chloroplast preparation was determined from the following equation:

$$A_{652} \times \frac{100}{34.5} = \text{mg of chlorophyll/ml of chloroplast preparation.}$$

- (c) A volume of chloroplast preparation that contains 50 μg of chlorophyll was added to each tube in the preceding table. Calculate the required volume in μl and include this value in the table at the start of this question (Show calculation, **2 marks**).

In the experiment, the following data were obtained:

<u>(i) Experimental (tube #1, 25 cm from lamp).</u>			<u>(ii) Control (dark) (tube #2).</u>		
<u>Time (min)</u>	<u>A600</u>	<u>$\Delta A600$</u>	<u>Time (min)</u>	<u>A600</u>	<u>$\Delta A600$</u>
0	0.860		0	0.860	
2	0.460		2	0.760	

The absorption coefficient (E) for DCPIP in the conditions of the experiment was 18,000 litres/mole.cm.

- (d) Calculate the rate of photoreduction of DCPIP in tube #1 in moles of DCPIP photoreduced/minute/ μg of chlorophyll (Show and *briefly explain* your calculations, **5 marks**).

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(e) The molecular weight of DCPIP is 290.1. What total weight of DCPIP (in grams) was reduced, **as a result of the activity of all of the reducing agents**, in **two** minutes in tube #1? (show calculation, 3 marks).

Question 3: In microscopy, what is the meaning of resolution? **(1 mark)**

Abbe's equation: $d = \frac{0.612\lambda}{n \sin \alpha}$

What does each of the following terms in Abbe's equation represent? **(2 marks)**

(i) $d =$

(ii) $n =$

(iii) $\alpha =$

(iv) $\lambda =$

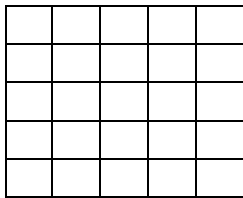
What is the value for **d** (in μm) if $n = 1.5$, $\sin \alpha = 0.85$ and $\lambda = 520 \text{ nm}$? **(1 mark)**

4 Sample exam questions: **Dean Unit, Biology 2290F/G** (Answers start on page 6)
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Question 4: You have a culture of cells that contain too few cells to count with the haemocytometer. You take 10 ml of this suspension, pellet the cells by centrifugation, discard the supernatant and resuspend the cells *up to* 4 ml with water. You determine that the concentrated cell suspension contains 4.2×10^6 cells/ml. How many cells/ml were there in the original culture? (1 mark).

Question 5: The diagram below represents the central part of the grid on a modified haemocytometer chamber. The largest square is 0.5×0.5 cm. When the chamber cover is in place, the depth of the chamber is 0.2 mm. What volume (in μm^3) is contained above each of the smallest squares shown in the diagram? (2 marks)

_____ 0.5 cm _____



Question 6: State Beer's Law in words

5 Sample exam questions: **Dean Unit, Biology 2290F/G** (Answers start on page 6)
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Question 7: The percent transmittance (%T) of a 1×10^{-4} M solution of a green dye is 1% at 520 nm. Predict the absorbance (A_{520}) of a 1×10^{-5} M solution of the same dye. (Note: both concentrations lie in the part of the standard curve for green dye at 520 nm where the relationship between concentration and absorbance is linear). (show calculations, **2 marks**)

Question 8: The pH of a solution is 6.73. Calculate its $[H^+]$, $[OH^-]$ and pOH (**3 marks**).

6 Sample exam questions: **Dean Unit, Biology 2290F/G** (Answers start on page 6)
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Question 1: *Chlorella* is a unicellular alga that grows well in liquid culture media. Using a haemocytometer, the cell density in a *Chlorella* culture was determined to be 5.625×10^7 cells/ml.

To make a standard curve of its absorbance at 750 nm (A_{750}) against the number of cells per milliliter, the culture was diluted **15-fold**. As shown in the table below, this diluted sample gave an A_{750} reading of 0.390. Complete the table to show (i) the number of cells per milliliter (#cells/ml) that correspond to each A_{750} reading, and (ii), the volumes of diluted culture and water in the blank spaces in columns 1 and 2 of the table. All tubes contain a total volume of 5.0 ml (**4 marks**)

<u>ml diluted culture</u>	<u>ml water</u>	<u>#cells/ml</u>	<u>A_{750}</u>
5.0	0	3.75×10^6	0.390
3.0	2.0	2.25×10^6	0.234
1.5	3.5	1.125×10^6	0.117
0.5	4.5	0.375×10^6	0.039

The #cells/ml in the first row is 5.625×10^7 divided by 15 = 3.75×10^6 cells/ml. Since absorbance is proportional to the #cells/ml, $\frac{0.234}{0.390} \times 3.75 \times 10^6 = 2.25 \times 10^6$ cells/ml

and $\frac{0.117}{0.390} \times 3.75 \times 10^6 = 1.125 \times 10^6$ and so on...

The dilution factors that generate these numbers of cells/ml can also be determined from the ratios of the A_{750} values:

e.g. 0.234 is 60% (3/5ths) of 0.390, therefore 3.0 ml of the original diluted solution when added to 2.0 ml of water will produce a concentration of 2.25×10^6 cells/ml. And so on.....

Question 2: The following table shows the contents of the experimental reaction mixture used to determine the rate of photoreduction of DCPIP at 25 cm from a 100 watt lamp.

<u>Tube #</u>	<u>2×10^{-4} M DCPIP (ml)</u>	<u>Reaction buffer pH 7.0 (ml)</u>	<u>Distilled water (ml)</u>	<u>Chloroplast preparation (μl)</u>
1. Experimental	2.0	2.0	6.0	23.9 *
2. Dark Control	2.0	2.0	6.0	23.9 *
3. Blank	0	2.0	8.0	23.9 *

(* volume to be determined)

(a) Complete the table to show the contents of tubes numbered 2 and 3 (**2 marks**)

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Chloroplasts were isolated according to the Biology 2290 protocol. Fifty (50) μl of this preparation was added to 5.0 ml of 80% (v/v) acetone, the tube was shaken to dissolve the chlorophyll and centrifuged at $1300 \times g$ for 3 minutes. The supernatant was transferred to a cuvette and its A_{652} was determined to be 0.720.

(b) What is the appropriate blank for this A_{652} determination?

80% (v/v) aqueous acetone (1 mark)

The concentration of chlorophyll in the chloroplast preparation was determined from the following equation:

$$A_{652} \times \frac{100}{34.5} = \text{mg of chlorophyll/ml of chloroplast preparation.}$$

(c) A volume of chloroplast preparation that contains 50 μg of chlorophyll was added to each tube in the preceding table. Calculate the required volume in μl and include this value in the table at the start of this question (Show calculation, **2 marks**).

$$0.720 \times \frac{100}{34.5} = 2.09 \text{ mg/ml} = 2.09 \mu\text{g}/\mu\text{l}$$

$$50 \mu\text{g divided by } 2.09 \mu\text{g}/\mu\text{l} = 23.9 \mu\text{l}$$

In the experiment, the following data were obtained:

<u>(i) Experimental (tube #1, 25 cm from lamp).</u>			<u>(ii) Control (dark) (tube #2).</u>		
<u>Time (min)</u>	<u>A₆₀₀</u>	<u>ΔA_{600}</u>	<u>Time (min)</u>	<u>A₆₀₀</u>	<u>ΔA_{600}</u>
0	0.860	0	0	0.860	0
2	0.460	0.400	2	0.760	0.100

The absorption coefficient (E) for DCPIP in the conditions of the experiment was 18,000 litres/mole.cm.

(d) Calculate the rate of photoreduction of DCPIP in tube #1 in moles of DCPIP photoreduced/minute/ μg of chlorophyll (Show and *briefly explain* your calculations, **5 marks**).

(i) $0.400 - 0.100 = 0.300$ (corrected $\Delta A_{600}/2$ minutes)

(ii) $\frac{0.300}{2} = 0.150$ ($\Delta A_{600}/\text{minute}$)

(iii) $\frac{0.150}{18000} = 8.33 \times 10^{-6}$ moles/litre of DCPIP photoreduced/minute

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(iv) $8.33 \times 10^{-6} \times \frac{10.02 \text{ml}}{1000 \text{ml}} = 8.35 \times 10^{-8} \text{ moles of DCPIP photoreduced/minute}$

(v) *divide by 50 μg of chlorophyll =*
 $1.671 \times 10^{-9} \text{ moles of DCPIP photoreduced/minute/}\mu\text{g of chlorophyll}$

(e) The molecular weight of DCPIP is 290.1. What total weight of DCPIP (in grams) was reduced, **as a result of the activity of all of the reducing agents**, in two minutes in tube #1? (show calculation, 3 marks).

Notice that this question deals with TOTAL weight reduced as a result of ALL OF THE REDUCING ACTIVITY in 2 MINUTES.

(i) $\Delta A_{600}/2 \text{ minutes} = 0.400$

(ii) $\frac{0.400}{18000} = 2.22 \times 10^{-5} \text{ moles/litre of DCPIP reduced/2 minutes}$

(iii) $2.22 \times 10^{-5} \times \frac{10.02 \text{ml}}{1000 \text{ml}} = 2.22 \times 10^{-7} \text{ moles of DCPIP reduced/2 minutes}$

(iv) $2.22 \times 10^{-7} \text{ moles} \times 290.1 \text{ g/mole} = 6.46 \times 10^{-5} \text{ g}$

Question 3: In microscopy, what is the meaning of resolution? (1 mark) _____

Resolution is the ability to image two distinct objects as two distinct objects (or anything which means the same)

Abbe's equation: $d = \frac{0.612\lambda}{n \sin \alpha}$

What does each of the following terms in Abbe's equation represent? (2 marks)

(i) d = **the minimum distance between two objects that will allow them to be resolved (or imaged as two distinct objects)**

(ii) n = **the refractive index of the medium through which the radiation (light) passes on its way from the specimen to the lens**

(iii) α = **the angle subtended by the optical axis and the outermost rays that enter the lens (draw a sketch to illustrate α somewhere on the page if you prefer).**

(iv) λ = **the wavelength of the imaging radiation.**

What is the value for **d** (in μm) if $n = 1.5$, $\sin \alpha = 0.85$ and $\lambda = 520 \text{ nm}$? (1 mark)

$$d = \frac{0.612 \times 520 \text{ nm}}{1.5 \times 0.85} = 249.6 \text{ nm} = 0.25 \mu\text{m}$$

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$$C_1 \quad x \quad V_1 \quad = \quad C_2 \quad x \quad V_2$$

$$C_1 \quad x \quad 10 \text{ ml} \quad = \quad 4.2 \times 10^6 \quad x \quad 4 \text{ ml}$$

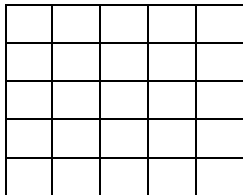
$$C_1 = 1.68 \times 10^6 \text{ cells/ml}$$

Question 5: The diagram below represents the central part of the grid on a modified haemocytometer chamber. The largest square is 0.5 x 0.5 cm. When the chamber cover is in place, the depth of the chamber is 0.2 mm. What volume (in μm^3) is contained above each of the smallest squares shown in the diagram? **(2 marks)**

The smallest squares shown in the diagram are 0.1 cm x 0.1 cm
 $= 1 \text{ mm} \times 1 \text{ mm}$
 $= 1000 \mu\text{m} \times 1000 \mu\text{m} = 1 \times 10^6 \mu\text{m}^2$

The depth is 0.2 mm = 200 μm , therefore the volume is
 $1 \times 10^6 \mu\text{m}^2 \times 200 \mu\text{m} = 2 \times 10^8 \mu\text{m}^3$

_____ 0.5 cm _____



Question 6: State Beer's Law in words *Absorbance is proportional to the number of absorbing particles in solution (Absorbance is linearly related to the concentration of an absorbing compound in solution).*

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Question 7: The percent transmittance (%T) of a 1×10^{-4} M solution of a green dye is 1% at 520 nm. Predict the absorbance (A_{520}) of a 1×10^{-5} M solution of the same dye. (Note: both concentrations lie in the part of the standard curve for green dye at 520 nm where the relationship between concentration and absorbance is linear). (show calculations, **2 marks**)

$$\text{Absorbance of } 1 \times 10^{-4} \text{ M solution} = \log \frac{1}{T} = \log \frac{1}{0.01} = 2.0$$

The 1×10^{-5} M solution is 10% of the concentration of the 1×10^{-4} M solution, therefore, since absorbance is proportional to concentration, 10% of 2.0 = 0.2

Question 8: The pH of a solution is 6.73. Calculate its $[H^+]$, $[OH^-]$ and pOH (**3 marks**).

$$[H^+] = 10^{-pH} = 10^{-6.73} = 1.86 \times 10^{-7} \text{ M}$$

$$[H^+].[OH^-] = 1 \times 10^{-14}, [OH^-] = \frac{1 \times 10^{-14}}{1.86 \times 10^{-7}} = 5.38 \times 10^{-8} \text{ M}$$

$$pH + pOH = 14, 6.73 + pOH = 14, 14 - 6.73 = 7.27$$