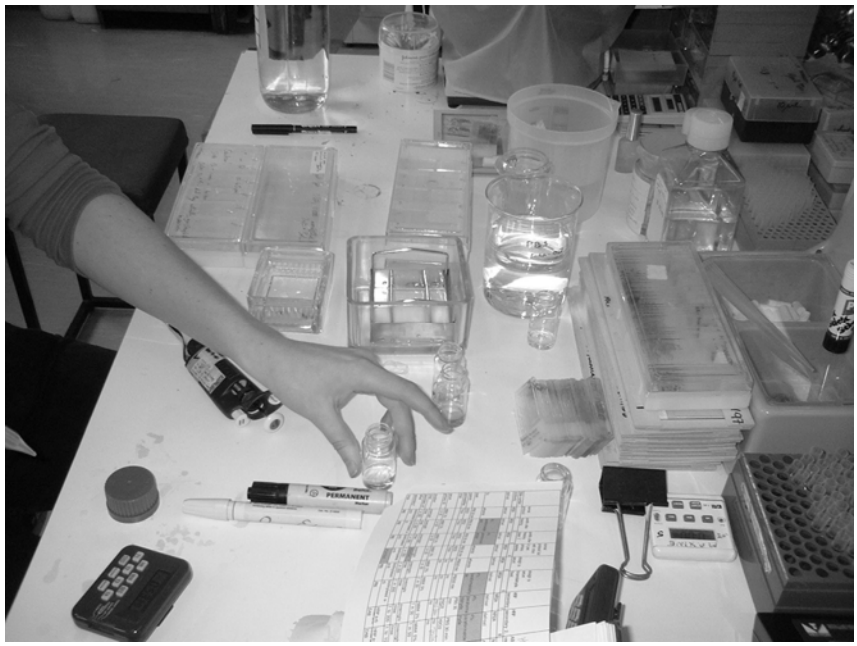
Laboratory Skills Workbook



Dr M. Lintern

Introduction

This workshop will give you the opportunity to have go at some basic lab skills, improve your technique and give you a bit more confidence for when you start your research projects next semester. The workshop is split into 5 units, each addressing a different skill or activity. In small groups you will work through each unit recording your results in this workbook. After 15-20 minutes you will finish the activity and move onto the next unit. It does not matter what order you work through the units.

You will not be marked on your results, but I will want to check your progress and see your data as you move through the workshop.

Please ask questions whenever you need to, and discuss your ideas and results with your group.

Unit 1	Weights and Measures
Unit 2	Volumes and Pipettes
Unit 3	Data Handling
Unit 4	Buffers and pH
Unit 5	Serial Dilutions

Unit 1 Weights and Measures

Aims

The aim of this unit is to practice weighing out set amounts of different substances. You must choose the correct balance, size of weighing boat, spatula and try and weigh out each of the following masses for 3 substances, NaCl, sucrose and protein M. Record on the sheet how close you get, and sketch what it looks like. Make a note of any problems or helpful tips.

Target weight **500mg**

	NaCl	sucrose	prot M
Weight achieved	_____	_____	_____
How did it look?			

Notes

Target weight **125mg**

	NaCl	sucrose	prot M
Weight achieved	_____	_____	_____
How did it look?			

Notes

Target weight **10mg**

	NaCl	sucrose	prot M
Weight achieved	_____	_____	_____
How did it look?			

Notes

Target weight **10µg**

	NaCl	sucrose	prot M
Weight achieved	_____	_____	_____
How did it look?			

Notes

Take home

- a visual idea of what these amounts can look like.
- an understanding that sometimes the same mass of different stuff looks different.
- an idea of the limitations of how low you can go!

Unit 2 Volumes and Pippettes

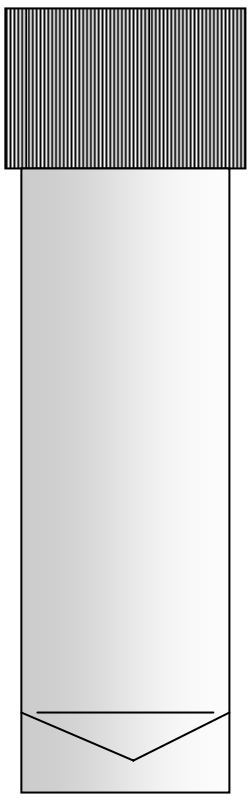
Aims

The aim of this unit is to gain experience at pipetting different volumes of distilled water. You need to choose the correct pipette, the correct tip and the right size vessel and aliquot out the required volume in duplicate. You can then weigh the vessels to see how accurate you have been. Record your findings below.

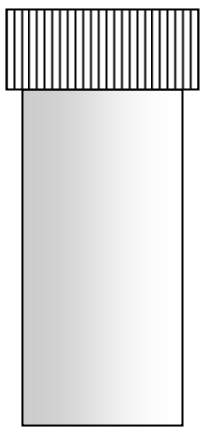
Pipette options	0-10µl, 0-20µl, 50-200µl, 50-1000µl, 5000µl, 10000µl
Tip options	size 1 (<10µl), size 2 (<200µl), size 3 (<1000µl), size 4 (<5000µl), size 5 (<10000µl)
Vessels	500µl microtubes, 1.5ml microtubes, bijoux, universals

Volume	Pippette	Tip	Vessel	Mass 1	Mass 2
5µl					
17µl					
82µl					
125µl					
250µl					
745µl					
1037µl					
1.42ml					
4.735ml					
12.26ml					

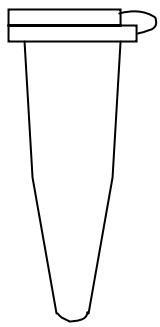
On the diagrams below, sketch the approximate levels of the volumes you have measured out onto the appropriate vessel



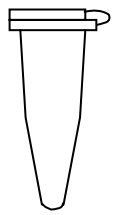
Universal tube



Bijoux



1.5ml microtube



500µl microtube

Take home

- an understanding of the range of pipettes, tips and tubes.
- an understanding of to use pipettes safely and accurately.
- the comparative accuracy of repetition.

Unit 3 Data Handling

Aims

The aim of this unit is to gain experience handling and plotting different kinds of data. This is a paper exercise where you look at the experimental data provided and decide how best to present the data. Make any calculations that are required and choose the most appropriate style of graph, the best scales, markers etc. Use the space below to make notes and sketches, and draw the graphs on the graph paper.

Take home

- an understanding of how to accurately present numerical data in graphical form
- an understanding of how to make the best choice of graph for the situation
- an understanding of how to handle blanks, legends, titles and labels

Data Set 1

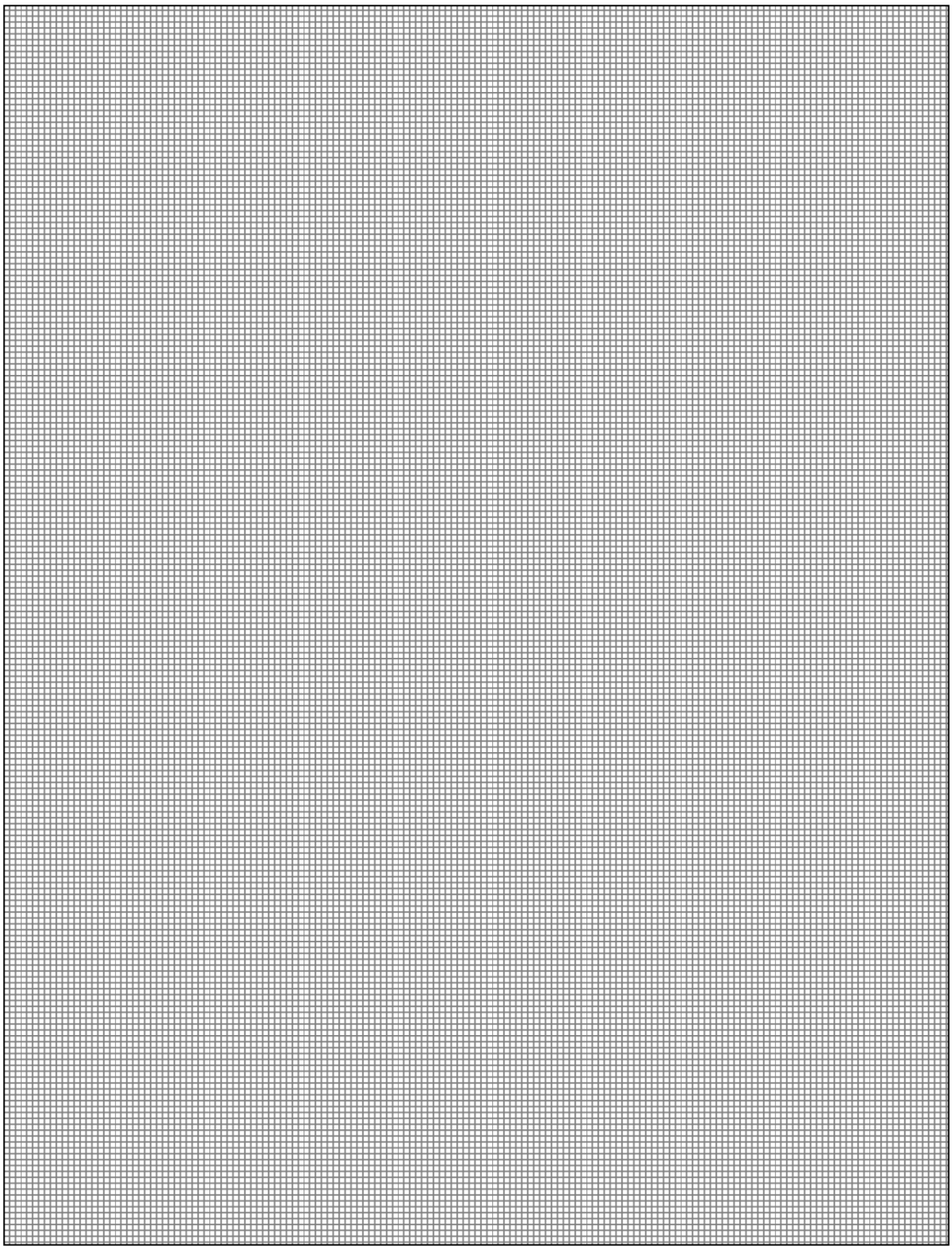
Cells in culture were treated with a single dose of drug X, drug Y or both drug X and Y. The effect on the activity of enzyme A was measured for 6 cultures for each case. Display the resulting data in graphical form, and use the graph to comment on the effects seen.

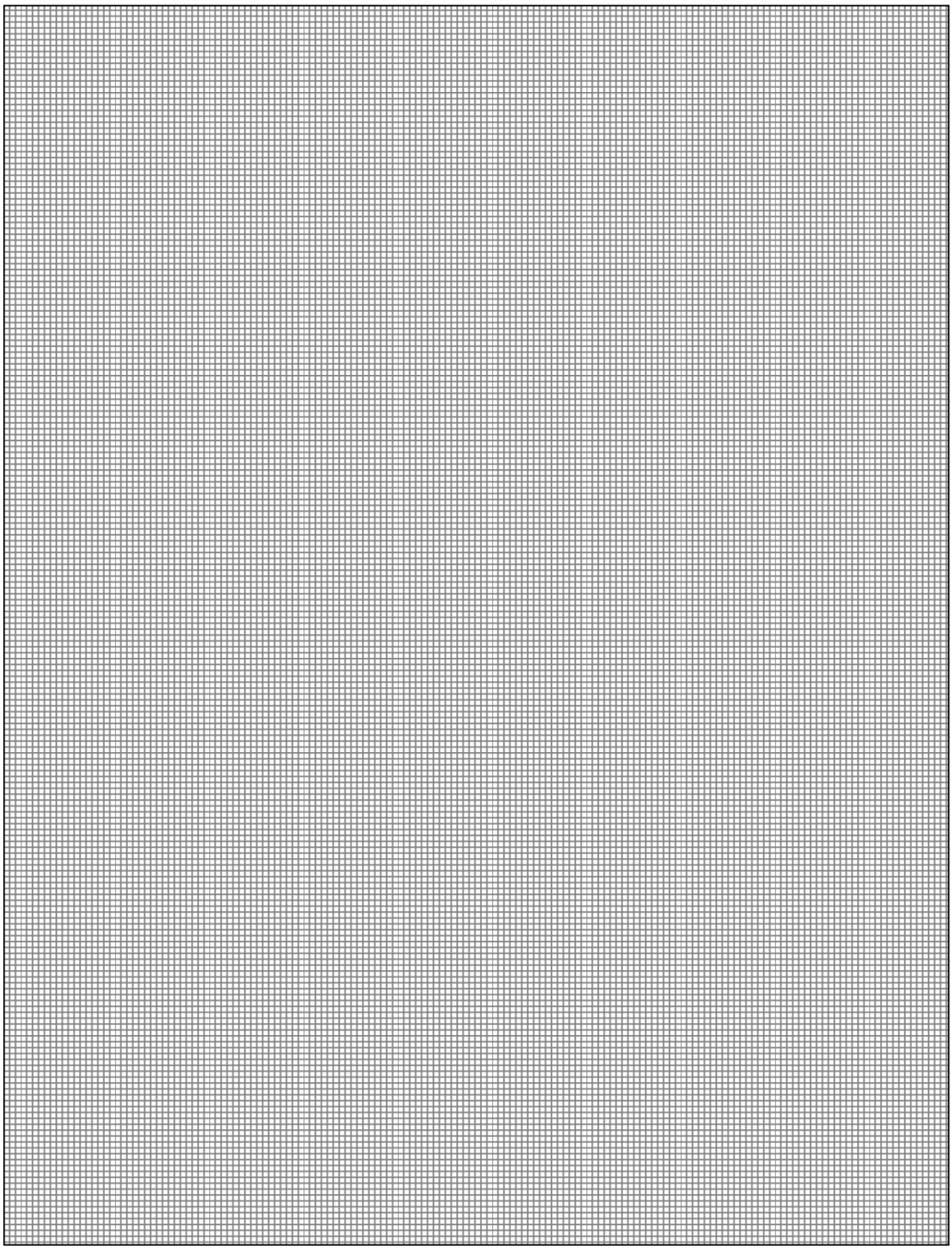
Treatment	**Enzyme Activity (units/ml)**
Drug X	1.37 ± 0.10
Drug X + Y	1.89 ± 0.25
Untreated	0.82 ± 0.05
Drug Y	0.90 ± 0.04
Control	0.03 ± 0.00

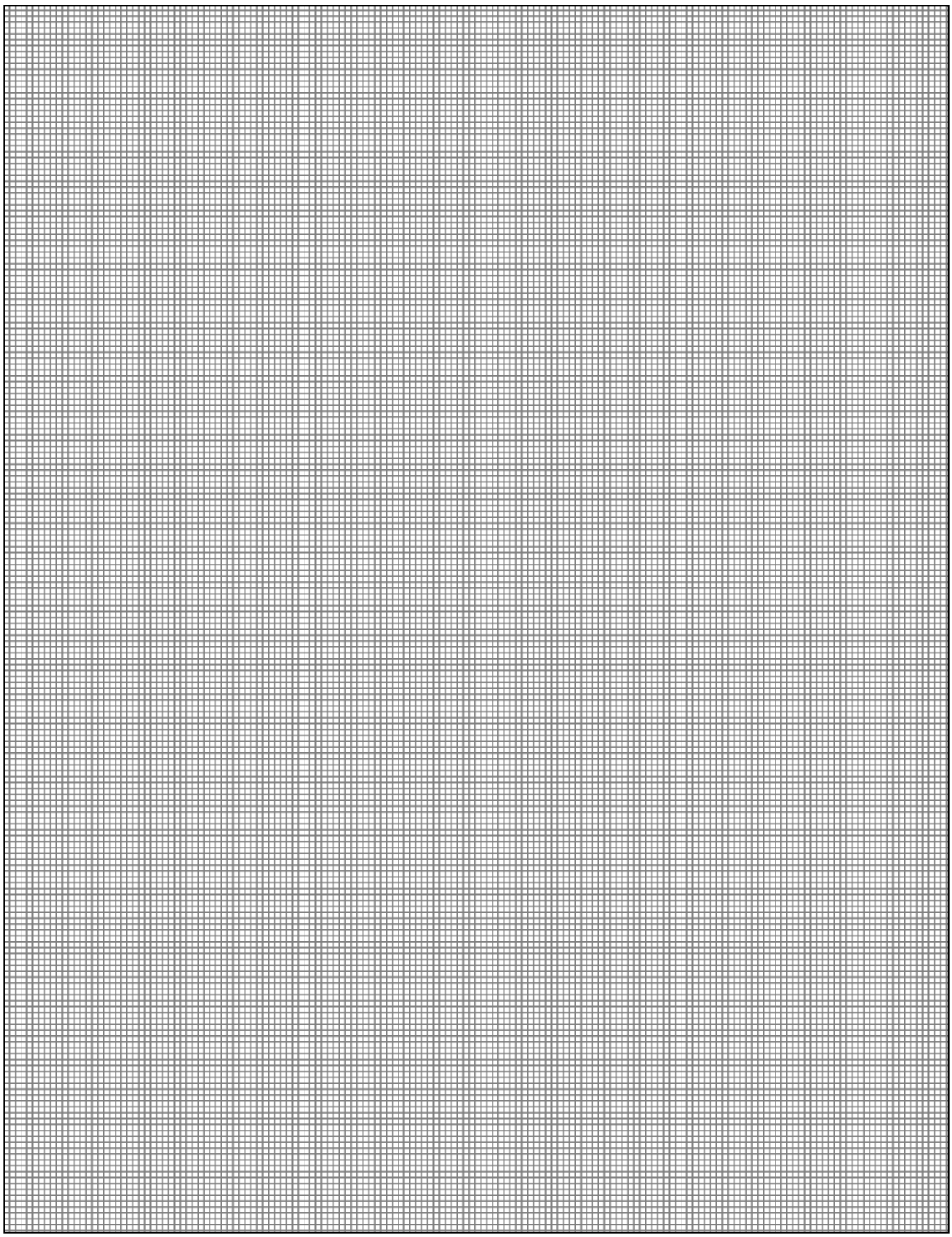
Data Set 2

In another experiment the cultured cells were treated with different amounts of drug X (data below). Display the data graphically and comment on the effects seen

Concentration of X	Enzyme activity (units/ml)
0	0.82 ± 0.05
0.1M	0.94 ± 0.10
0.2M	1.10 ± 0.20
0.3M	1.18 ± 0.10
0.4M	1.42 ± 0.10
0.5M	1.61 ± 0.20
0.6M	1.69 ± 0.20
0.7M	1.43 ± 0.30
0.8M	1.01 ± 0.07
0.9M	0.97 ± 0.10
1.0M	0.89 ± 0.08







Unit 4 Buffers and pH

Aims

The aim of this unit is for you to gain experience on the correct way to make up the most common lab solution, a buffer. Following the protocols below you will make up each buffer, measure its pH and adjust if necessary recording your actions as you go. Make sure you calibrate the pH meter before you start.

Buffer 1

Measure out approximately 90ml of dist. water into a beaker and add 1 PBS tablet and dissolve on the stirrer.

Measure the pH initial pH

Alter the pH to 8.2 What do you need to use?

How much did you need to use?

Pour the buffer into a 100ml volumetric flask and carefully top up to exactly 100ml

Now pour the completed buffer back into the beaker and check the pH again

Final pH is?

Buffer 2

Measure out approximately 90ml of dist. water into a beaker and add 1 PBS tablet and dissolve on the stirrer.

Measure the pH initial pH

Alter the pH to 6.1 What do you need to use?

How much did you need to use?

Pour the buffer into a 100ml volumetric flask and carefully top up to exactly 100ml

Now pour the completed buffer back into the beaker and check the pH again

Final pH is?

Buffer 3

Measure out approximately 90ml of dist. water into a beaker and add pot of NaCL/Tris.HCL and dissolve on the stirrer.

Measure the pH initial pH

Alter the pH to 9.3 What do you need to use?

How much did you need to use?

Pour the buffer into a 100ml volumetric flask and carefully top up to exactly 100ml

Now pour the completed buffer back into the beaker and check the pH again

Final pH is?

Take home

- an understanding of how to calibrate a pH meter
- an understanding of how to measure and alter the pH of solutions
- an understanding of how to make up a simple buffer to the correct volume and pH

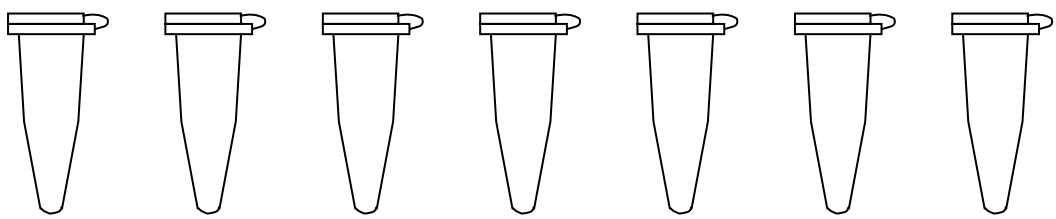
Aims

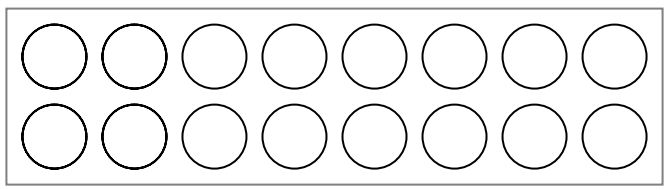
The aim of this unit is for you to make a serial dilution from a coloured solution. You will then use a spectrophotometer to find out how much colour is present in each dilution. By making each dilution up in triplicate, you can compare how accurate your preparations are. The coloured reagent we are using is a dye called coomassie blue. Be very careful with it as it will stain anything, clothes books, fingers etc!

Protocol

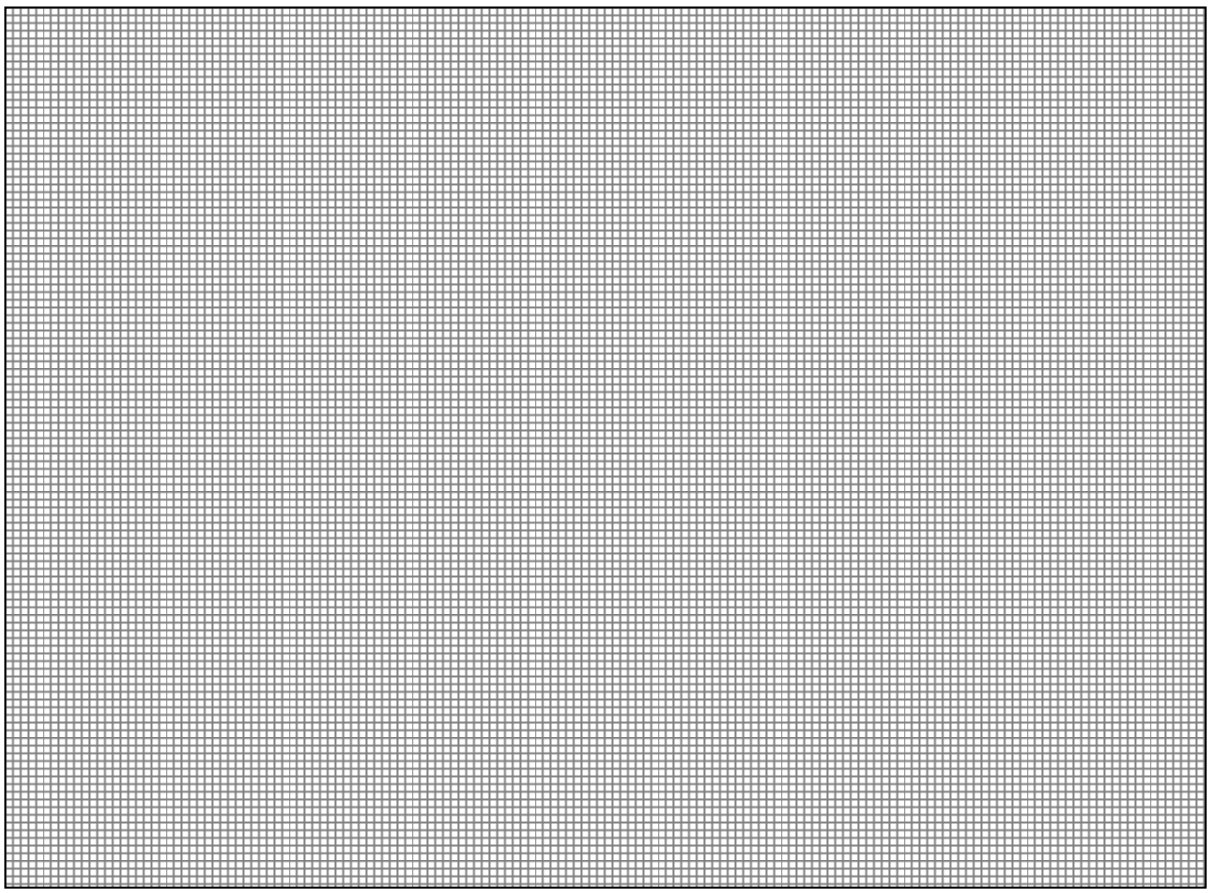
The coomassie solution you have been given is at 1% w/v. Using the microtubes provided you must make up a serial dilution of the solution from 1/2 down to 1/32 with dist. water. You must do this in triplicate and make up enough to load 100µl of each sample onto the microplate. Record below your dilution plan and then once you have the readings fill in the table with your data. Comment on the accuracy of your dilutions, and plot a small graph of your findings.

Dilution planner





Dilution	Set 1	Set 2	Set 3	Average



Unit 5 Serial Dilutions

Take home

- practical experience of making a serial dilution
- an understanding of the importance of duplication
- an understanding of how to catalogue and handle experimental data