A Question of Taste – Demonstrator Notes

Section	Learning outcome/ things that need to be covered	Demonstrator/ tech activity	Timing
Introduction – outside lab	• Lab rules – use as questioning opportunity to warm up group		0-10 min
Introduction	• What will we be doing today – get ideas from the group • Summary of the day		10-15 min

Recapping DNA background knowledge	• Finding out about how much group knows about DNA and genes
	• Ask questions -
	  ○ What is DNA?
	  ○ What is a gene?
	  ○ Where is our DNA?
	  ○ What are the units of DNA called?
			15-20 min

Tasting the PTC	**Students taste PTC and control strips** Discussion of results • Ask what the students could taste – is it sweet, sour, bitter etc? • Distinction between bitter and sour • Ask who rated taste as a 5 • Ask who rated taste as a 0	• Make sure students have washed hands • Ensure each student has a control (corner cut off) and PTC strip • Hand out record streets • Tidy away strips and paper towels	20-25 min

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Finding out about PTC	• Explain that the taste strips were impregnated with the chemical phenylthiocarbamide (PTC) • Not everyone can taste this chemical • PTC tasting has a genetic basis – ask students what we call the visible expression of our genes? – phenotype • We are now going to investigate our genetics – what do we call the makeup of our genes? – genotype		25 – 30 min
Harvesting cells	• Explain to students that we are going to collect a cell sample by swilling mouth with saline solution (suggest that students gently chew the inside of their cheeks to improve their sample – we don't want any blood) – ask students why we use saline • Advise students not look at each other during swilling **Students carry out mouthwash**	• Make sure each student has saline sample • Give each student a number • Make sure students label 1.5ml microfuge tube with their number • Demo and supervise pipetting 1ml of sample into 1.5ml microfuge tube, make sure students take sample from bottom of cup • Collect tubes for centrifuging	30 – 35 min
Separating out the cells	• Ask students why we centrifuge the samples • Explain what will happen when samples are returned or if they have small/ fluffy pellets • Demonstrate tipping off most of the sample – point out level on the screen • Explain how to re-suspend sample • Ask students to place samples in tube rack for later	• Place tubes in centrifuge – place in order to speed returning • Check pellets before returning tubes to students – explain what to do if pellets are small • Supervise tipping off the supernatant • Supervise vortex mixing to re-suspend pellet	35 – 45 min

<table>
<tr><th>Section</th><th>Learning outcome/ things that need to be covered</th><th>Demonstrator/ tech activity</th><th>Timing</th></tr>
<tr><td>Micropipettes</td><td>Introduce idea of the precision needed to work with tiny volumes – ask students if they know what a micro litre is
Explain how to use the micropipettes – things to highlight –
They are delicate and expensive!
How to hold them correctly
There are two stopping points that have different uses
How to alter volume
How to put on and remove a tip
Avoiding contamination
Students practice pipetting</td><td>Supervise pipette practice
Encourage students to measure a variety of volumes
Make sure students don’t go outside the ranges of their pipettes – explain why this is</td><td>45–55 min</td></tr>
<tr><td>Extracting DNA</td><td>Explain that 1.5ml microfuge tubes containing chelex have been placed in the trays
Demonstrate how to take 30µl of the cell suspension and transfer this to the chelex
Students transfer sample
Ask students to collect samples in the rack and pass to the presenter ready to go in the heat block</td><td>Hand out chelex – 1 sample per student
Supervise transfer of cell suspension
Collect samples from group and place in 100°C heat block – set timer for 10 minutes</td><td>55 min – 1 hr</td></tr>
<tr><td>While samples are in heat blocks</td><td>Explain why we use chelex – use PowerPoint slide 20
Ask students why we are heating the samples to 100°C
Ask why we will centrifuge the samples again</td><td>Remove samples from heat block after 10 minutes
Spin samples in centrifuge for 1 minute</td><td>1hr – 1hr 10</td></tr>
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Section	Learning outcome/ things that need to be covered	Demonstrator/ tech activity	Timing
Taste – how it works and the *TAS2R38* gene (during 10 minute heat block)	• Explain that taste receptors are proteins • PTC taste receptor coded for by *TAS2R28* on chromosome 7	• During this time samples will finish in heat block/ centrifuge so return samples to students	
PCR set up	• Ask students if they remember what the process we are doing today is called • Explain that we will think about how it all works once we have set up the reaction • Explain the different ingredients needed – see if the students can name any of them • Ask students to label PCR tubes with numbers • Demonstrate how to take DNA sample - *use PowerPoint slide 26 to show where to concentrate* • Explain how much to add – *see PowerPoint slide 27*	• Hand out tubes with primers (one per pair) and PCR beads (one each) • Supervise students taking their DNA samples • Make sure students take samples from the top of the liquid avoiding beads etc • Supervise mixing of ingredients • Collect PCR tubes and place in PCR machine	1 hr 10 – 1hr 20
PCR background	• Ask students if they can explain what PCR does • Explain that it can be used to copy specific areas of DNA • Show flash animation – describe each step		1hr 20 – 1hr 35

PCR activity	• Explain how the activity works
	
	Students carry out activity
		• Hand out the following to groups of 4 students
		  ○ 1 piece of double stranded DNA
		  ○ Some primers
		  ○ Some nucleotides
		  ○ Scissors
		  ○ tape
		• Supervise students as they carry out the activity
			1hr 35 – 1hr 50

Section	Learning outcome/ things that need to be covered	Demonstrator/ tech activity	Timing
Gel pouring	• Briefly explain that later we will be thinking about electrophoresis but now we need to start setting it up • Ask for volunteers from each table • Demonstrate how to place combs, casting gates and pour gel **Students help prepare gels**	• Hand out gels to students – make sure they take care of hot gel • Supervise gel pouring	1hr 50 – 2hr
Lunch			2hr – 2hr 30
How to work out if you have taster DNA	• Introduce the idea of SNPs – try to get students to work out what a SNP might be if they don't already know • Ask students what we call different versions of the same gene • Explain the taster and non-taster SNP • Explain how a single base change may or may not affect the protein coded for • Try to get students to work out why there seems to be a whole range of tasting abilities not just taster and non-taster *(explored in PowerPoint slide 38 if students can't answer)*		2hr 30 – 2hr 50
Homozygous and heterozygous	• Explain that we have two copies of *TAS2R38* and that the various combinations of the C145G SNP explains the range of tasting ability • Explain the terms homozygous and heterozygous		

Section	Learning outcome/ things that need to be covered	Demonstrator/ tech activity	Timing
Explaining restriction enzymes	• Ask if the students know what restriction enzymes are • Explain how the enzymes cut DNA at specific places • Explain HaeIII and its restriction site • Show students the two versions of the PTC receptor gene *(slide 42)* – can they work out which one will be cut by the HaeIII?	• Get ice beakers from the freezer	
Setting up the digest	• Ask students to label fresh tube with number and the letter D • Explain how to set up the digest – *see PowerPoint slide 43* • Explain that we will set up a negative control by heating the remaining PCR product with no HaeIII **Students set up restriction digest**	• Hand out tubes of HaeIII, water • Collect all tubes and place in 37 degree heat bath/ block for 30 min	2hr 50 – 3hr
Projected digest results	• Run through the possible digest results - *PowerPoint slide 44 (note – the diagram in this slide represents our PTC product, not our whole chromosome)*		

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<tr><td>Restriction enzyme/ electrophoresis activity</td><td>Explain how the activity works (see instructions)
Things for students to know –
The double stranded DNA will have the same restriction enzyme site with the same sequence which reads forwards on the sense strand and backwards on the antisense
Restriction enzymes only cut at specific sites
Both strands are cut
We are using just one of many restriction enzymes
Other enzymes will cut different sites
Student carry out restriction enzyme activity

Students can now simulate electrophoresis with their DNA fragments – either on the table top or wall etc

Things for students to know –
Small fragments move further than large fragments
Different DNA samples will produce different banding patterns
This technique can be used for DNA fingerprinting etc</td><td>Hand out the following
One DNA strand per group
Scissors
Velcro, blue tack etc if sticking on the wall or board</td><td>3hr – 3hr 20</td></tr>
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Section	Learning outcome/ things that need to be covered	Demonstrator/ tech activity	Timing
Humans, chimps and PTC	• Remind students that in human populations there are PTC tasters and non-tasters • Ask them if they think chimps can taste PTC, why do they think this? • Ask why humans and chimps may have evolved similar traits – useful to both species • Explain the background research looking at evolution of non-tasting in humans and chimps • Show the different SNPs and explain that these haven't been inherited from the same place, but the outcome is the same • What do we call this? Convergent evolution ○ Get students to come up with other examples of convergence on their tables		3hr 20 – 3hr 35
Gel electrophoresis	• Briefly recap how electrophoresis works • Ask for volunteers from each table to help prepare the gels – removing combs, casting gates and pouring the buffer	• Supervise gel preparation, making sure students are careful and don't damage gels • Hand out bottles of buffer • Collect up combs, casting gates	3hr 35 – 3hr 45
Loading samples	• Demonstrate how to load the samples • Explain that everyone will have a chance to practice loading • Explain that we will load both our digested and undigested samples – show lane sheet for recording **Students load their practice and real samples**	• Return samples from heat block • Supervise loading – spin samples if needed • Set up and turn on power packs (120V for 25 min)	3hr 45 – 4hr 10

Section	Learning outcome/ things that need to be covered	Demonstrator/ tech activity	Timing
Dialogue	Explain the dialogue activity – • The activity is designed to look at the possible reasons for why non tasters might have evolved in human and chimp populations • No right or wrong answer • Non-tasting may be a coincidence but there is more likely a reason – i.e. an advantage Students will – • Arrange prompt cards in order from good reason to bad (cards can go side by side if they want) • Have to be able to explain their reasoning Activity works well if started in small groups before then having a class discussion	• Hand out student cards • General tidy of lab space • Put out light boxes and filter glasses • Towards the end of the dialogue the electrophoresis will finish – remove gels and place on light boxes	4hr 10 – 4hr 40
Results	• Explain that the gels have now finished • Describe how he stain works • Run through the possible results we will see and what these tell us • Allow students to look at their gels	• Help students interpret their gels	4hr 40 – 4hr 55
Round up	• Ask students if their genotype matched their phenotype • If any don't match try and get students to explain why this might happen		
End of workshop	• Ask students/ teachers to fill in evaluation forms • Thank students for their work • Ask students to wash hands and return lab coats		4hr 55 – 5hr