Test Name: Immobilised Enzymes

Marking Guides

Question: 1 (299049)

Question	Expected Answer	Mark	Additional Guidance
(a) (i)	microbes **/** (living) organisms **/** cells **/** enzymes ; (make) product **/** for human benefit / (carry out) conversion / reaction / industrial process ;	2	**CREDIT** microorganisms / bacteria / prokaryotes / fungi **CREDIT** living things **CREDIT** cell components / parts of cells **CREDIT** example such as (named) food or medicine BUT **IGNORE** cheese (as stated in question) **IGNORE** process unqualified
(a) (ii)	microbes **/ AW** , killed / removed / not present ; enzymes denatured ; (so no) competitors **/** unwanted reactions **/** (human) health risk ;	2 max	**Mark the first two suggestions** **IGNORE** contamination / sterile **IGNORE** idea of preserving milk **AW** for microbes as in **(a)(i)** plus **ACCEPT** organisms **DO NOT CREDIT** microbes denatured **CREDIT** (no) competition **CREDIT** (no) food spoilage / change of flavour / loss of quality **CREDIT** (no) pathogens / harmful microbes / TB **"Kills harmful microbes"** or **"Kills pathogens"** scores 2 marks (mps 1 & 3)

Question	Expected Answer	Mark	Additional Guidance
(b) (i)			**Award mp 1 plus 2 max from the other mark points**
1	enzyme ;		*1* **ACCEPT** globular / tertiary / catalyst / catalytic (protein)
	plus any 2 of the following	1	
2	(enzyme) not, changed / used up ; **ora**		*2* **ora** = can be used again / re-used **IGNORE** enzyme recycled
3	*idea of* ESC (forms) / substrate and enzyme (bind) ;		*3* ESC = enzyme-substrate complex **ACCEPT** substrate entering active site
4	products (and enzyme) released at end ;	max 2	
(b) (ii)			**Mark the FIRST suggestion on each numbered line** **IGNORE** 'cheaper' without qualification
1	(enzyme can be removed to be) used again ;		
2	(enzyme can) to leave pure(r) product ; **ora**		*2* **ACCEPT** cheaper / easier, downstream processing
3	(enzyme) more stable / more efficient / works better ;	2	*3* **CREDIT** less susceptible to, pH / temperature, change / extremes **"enzymes work at high temperatures"** = 0 **"enzymes work at higher temperatures"** = 1 (because comparative statement made)

Question	Expected Answer	Mark	Additional Guidance
(c)	***This is a QWC question*** *Section I - Obtaining the gene*		
1	use restriction, enzyme / endonuclease ;		*1* **CREDIT** named example e.g. *Eco* R1, *Bam* H1, *Hin* dIII
2	to, cut out / get / isolate, (rennin) gene / DNA coding for rennin ***or*** to, fragment / digest, DNA ;		*2* **DO NOT CREDIT** 'cut gene' **IGNORE** 'break up DNA'
3	gene probe ; ***OR***		
4	obtain rennin mRNA ;		***NOTE***
5	(use) reverse transcriptase ;		***1-9*** **CREDIT** whichever of the three alternative "obtaining the gene" protocols yields most marks, **either award marking points** **1-3** **or 4-6** **or 7-9**
6	to make cDNA ; ***OR***		
7	sequence, rennin (protein) ;		
8	work out base code ;		
9	make this DNA sequence ;		
10	sticky ends ;		*10* can be awarded, once only, in Sections I or II
	Section II - Vector		
11	cut (open), plasmid / phage ;		*11* **DO NOT CREDIT** 'cut out plasmid' **DO NOT CREDIT** 'ring of DNA' unless it is clear that plasmid is being referred to
12	using same restriction enzyme ;		*12* **CREDIT** same named enzyme (re. mp1)
13	annealing / base pairing of sticky ends ;		*13* **CREDIT** idea of sticky end bases hydrogen bonding
14	join sugar-phosphate backbones ;		*14* **CREDIT** formation of phosphodiester bonds
15	(using DNA) ligase ;		
16	recombinant, vector / plasmid / phage / DNA ;		
	Section III - Introduction into host cell		
17	mix with bacteria ;		
18	detail of conditions ;		
19	transformation (plasmid) / transduction (phage) ;	max 7	*18* e.g. Ca^{2+} ions added / heatshock (freeze then inc to 40°C) *19* **CREDIT** transform / transformed / transduce / transduced **IGNORE** transgenic
	QWC – sequencing of steps – at least 1 mark point scored from each of the three sections, in the correct order ;	1	I. obtaining gene (mp **1 – 9**) followed by II. vector (mp **13 – 16**) followed by III. introduction to host cell (mp **17 – 19**)
	TOTAL	**17**	

Question: 2 (6815870)

Question Number	Answer	Max Mark
(a)(i)	*max 1 for meaning of term* attached to an insoluble material / AW; *max 2 for description* (micro)encapsulation / (trapped) in alginate beads; adsorption / stuck onto, collagen / clays / resin / (porous) glass; cross linkage / covalent / chemical, bonding to, cellulose / collagen fibres; gel entrapment / trapped inside gel e.g. silica (lattice / matrix); partially permeable membrane (polymer) microspheres;	[3]
(ii)	*any three from the following:* urine can be processed / no problem of removing urine / AW; pure / drinkable / useable, water produced; **A** water recycled space saving / less water needs to be taken into space; payload limit / weight reduction / AW; no problem in separating enzyme from products / product not contaminated; ref. to longer shelf-life of enzyme; no need to take more enzymes into space / enzymes reusable ; **A** enzymes recoverable AVP; e.g. larger surface area of enzyme exposed, more stable at extremes, ref. to ease of use (of bioreactor)	[3]
(b)(i)	adding / using, water to break, bond / ester bond, (in molecule); **A** breakdown into smaller molecules	[1]
(ii)	matrix, protects / stabilises, enzyme / lipase; functions, at optimal rate / more efficiently, at higher temperature / 45 °C; **A** *greater activity / AW* ref. to soluble lipase begins to denature (reducing activity); ora functions, at optimal rate / more efficiently, at lower pH; ref. to presence of fatty acids changing pH; ref. to ionic bonds breaking (in soluble lipase) ; ora AVP ; e.g. ref to industrial uses ref to effect on R groups	max[4]
	Total:	[11]

Question: 3 (2272120)

Question				Answer	Marks	Guidance
	(a)			reduce / slow, flow rate ; repeat process / run milk through again ; test for (named) sugars in milk ;	2	**ACCEPT** close tap for a time period **CREDIT** glucose, galactose, lactose, Benedict's test
	(b)	(i)		*any two from* hydrophobic / ionic bond, to (named), solid / support ; covalent bond / cross-link to, (named) substance; membrane separation ; (en)trap / encapsulate / suspend, in (named), matrix ;	2	**Mark as prose.** **IGNORE** ref to cross-linking agents **ACCEPT** 'insoluble material for solid. Suitable solids = clay, carbon, resin, glass, gold, ceramic beads. **CREDIT** adsorption (but not absorption) **CREDIT** carrier bound. **CREDIT** cross-link them together. Suitable substances = other enzymes, collagen, cellulose. **ACCEPT** microcapsules Suitable matrix materials = collagen, cellulose, silica gel, hydrogel, but **DO NOT CREDIT** entangled / alginate
		(ii)	1	(enzyme) can be re-used so reduces cost ;	4	
			2	product, pure(r) / uncontaminated ;		**2 ACCEPT** product not mixed with enzyme
			3	reduced downstream processing costs ;		3 **ACCEPT** save money on purifying product
			4	(immobilised enzyme) works at high(er) temperature ;		**4 CREDIT** enzymes not denaturing at increased temperature **CREDIT** immobilised enzymes thermostable
			5	(immobilised enzyme) works in changed pH ;		5 **CREDIT** enzymes not denaturing in changed pHs
			6	reaction, can be faster / have higher yield , because can be done at higher temperature ;		6 This explanation scores mp 4 and mp 6 (unless mp 4 already awarded).
				Total	8	