A2 bio

• PAPER 5 - PLANNING, ANALYSIS, EVALUATION

a, Planning

- constructing a hypothesis (+ IV, DV, Δ affect) ⟶ sketch a graph – quantifiable, testifiable, falsifiable
- use the right apparatus
- identify variables
  - independent : changing [range]
  - dependent : measuring
  - control : standardized
    - ↳ control experiment

b, Method

- changing and measuring Independent Variable
- keep control variables constant ⟶ apparatus used
- measuring Dependent Variable ⟶ step-by-step
- how volumes / solutions are prepared

- describe control experiment → ensure IV affects DV
- sequence of steps : how use apparatus to collect results
- describe how to ensure quality of results : inspection
  - validity
  - reliability : 3 repeats
- risk assessment : identify + reduce
- recording + displaying data, drawing conclusions

c, dealing with data

- use table to identify key points in data
- sketch / draw graphs
  ↳ confidence limit error bars :
  + certain 95% of data lies in the range
- carry out calculations with data

↳ Calculations:

- mean, median, mode / modal class
  ↳ data has normal distribution → 3m's the same
- % ↑ or ↓
- range + IQR = UQ - LQ
- STATISTICS → state null hypothesis
  + standard deviation

$$s = \sqrt{\frac{\sum (x - \bar{x})^2}{n-1}}$$

  + standard error

$$S_M = \frac{s}{\sqrt{n}}$$

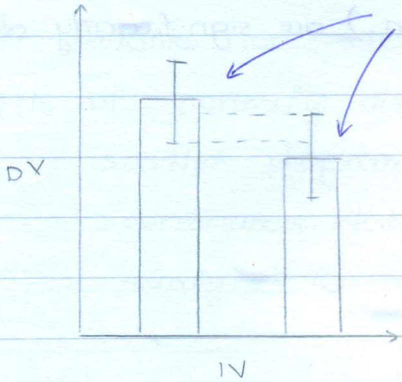
s.d

sample size

↳ tells: 95% certain that mean value is within $2 \times S_M$

null hypothesis: ~~assume there is no~~

- error bars:



$\pm 2 S_M \rightarrow$ 95% confidence limits

↳ overlap: not a significant difference between

- chi-squared (χ^2) test:

  test difference between observed and expected frequencies due to chance, wether null hypothesis can be correct

	Yellow, large	Yellow, small	White, large	White, small
observed: O				
expected: E				
$O - E$				
$(O-E)^2$				
$\frac{(O-E)^2}{E}$				

$\Sigma \frac{(O-E)^2}{E}$ e.g.: eggs

e.g.: $\chi^2 = 1.24 \rightarrow$ compare with D.o.F

degrees of freedom = # of categories − 1

↳ $\geq 0.05 \Rightarrow$ probability of null hypothesis being correct

- t-test: whether the means of 2 sets of data (with roughly normal distribution) are significantly different

$$t = \frac{\bar{x}_1 - \bar{x}_2}{\sqrt{\left(\frac{s_1^2}{n_1} + \frac{s_2^2}{n_2}\right)}}$$

→ calculate degrees of freedom $= n_1 + n_2 - 2$

$\leqslant$ 5% confidence level

⇒ accept null hypothesis: assume there is no significant difference between the 2 samples

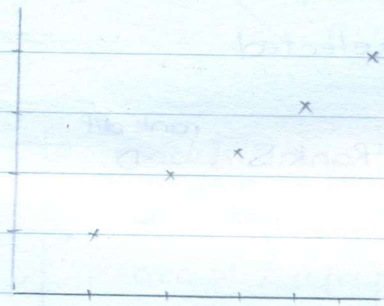
t-test suitable for:

- continuous data
- data normally distributed
- s.d. approx same
- samples $n < 30$ each

- Pearson's linear correlation

suitable for: test for correlation between 2 sets of normally distributed data

- scatter diagram indicates relationship
- continuous data
- ideally ≥ 10 paired observations



- possitive
- negative correlation
- no

	x	y	xy
1			
2			
3			
mean	$\bar{x} =$	$\bar{y} =$	$\sum xy =$
$n\bar{x}\bar{y}$			
s.d.	$S_x =$	$S_y =$	

$$r = \frac{\sum xy - n\bar{x}\bar{y}}{n S_x S_y}$$

$0 < r \leq 1$: +ve correlation

$-1 \leq r < 0$: -ve correlation

$r = 0$: no correlation

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- Spearman's rank correlation : find out if there is correlation between 2 sets of variables,
  NOT normally distributed.
  - data points independent
  - data collected can be ranked
  - scatter diagram
  - ideal $10 \leq n \leq 30$ paired observations
  - data randomly selected

Quadrat	# R	Rank R	# S	Rank S	rank dif D	D^2
1						
2						
⋮						
19						
20						

$$r_s = 1 - \left(\frac{6 \times \sum D^2}{n^3 - n} \right)$$

$0 < r_s \leq 1$: +ve correlation

$-1 \leq r_s < 0$: -ve "

$r_s = 0$: no "

d, Evaluation

- identify anomalous values — repeat / leave data out
- suggest for ↗ causes
- assess : replicated efficiently ? / range ?
- assess : method effectiveness ? / measuring IV, DV / CV controlled well ?

→ conclusion :

- all key points — explain
- accept / reject hypothesis with support from data

↳ make further predictions

how could experiment be improved ?

↳ + methods alternative
+ range

<u>Glycolysis</u> : phosphorylation of glucose and subsequent splitting of fructose 1, 6 - bisphosphate (6C) into 2 triose phosphate molecules, → further oxidised to pyruvate with a small yield of ATP and reduced NAD.

12. Energy & Respiration

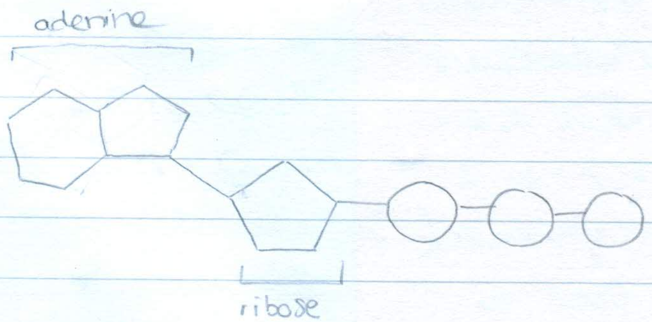
12.1. Energy

ATP is the universal energy currency as it provides the immediate source of energy for cellular purposes

a) Need for energy in living organisms : anabolic reactions
- DNA replication , protein synthesis
- active transport
- movement
- maintenance of body t°

b) ATP - universal energy currency → high turnover
↳ 'adenosine triphosphate'
- readily hydrolysed to release energy
- small } easily transported around cell
- water - soluble }
- immediate energy donor

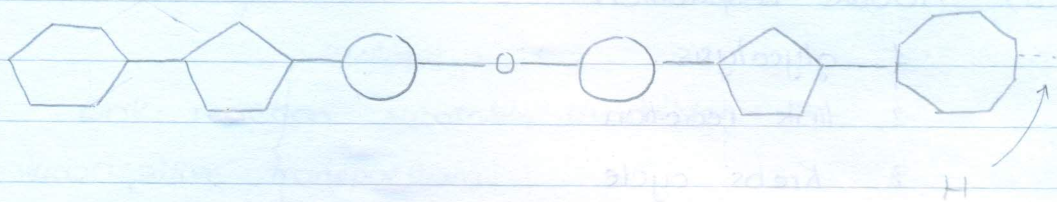
c) ATP synthesis
↳ during respiration :
→ substrate - linked reactions < glycolysis, Krebs cycle



- electron transport chain on mitochondria + chloroplasts

d) Coenzymes
- H carrier molecules
  - NAD
  - FAD
- coenzyme A

- NAD (nicotinamide adenine dinucleotide)



$NAD + 2H \rightleftharpoons$ reduced NAD

$NAD^{+} + 2H \rightleftharpoons NADH^{+} + H^{+}$

- FAD (flavin adenine dinucleotide)
  ↳ used in respiration - Krebs cycle
- coenzyme A:
  carrier of acetyl groups to the Krebs cycle

f.) Energy values

Respiratory substrate	Respiratory quotients (RQ)	Energy density / kJg^{-1}
carbohydrate	1.0	15.8
lipid	0.7	39.4
protein	0.9	17.0

Lipids → energy rich
- ↑ # of H atoms in structure ⟶ ↑ energy value
- lipids: ↑ H / per molecule (fatty acid)

→ produce more reduced NAD = more ATP per gram
- fats only broken down aerobically

12.2. Respiration

↳ process whereby energy from complex organic molecules is transferred to ATP

a) Aerobic respiration

1. glycolysis → cytoplasm
2. link reaction — matrix
3. Krebs cycle — matrix
4. oxidative phosphorylation — inner membrane

(2–4: mitochondria)

b) Glycolysis

glucose (hexose) (6C)

ATP → ↓

fructose phosphate (6C)

ATP → ↓

fructose bisphosphate (6C)

(fructose phosphate and fructose bisphosphate steps: phosphorylation)

↓

2 molecules triose phosphate (3C)

↓ → 2 ATP

↓ → 2H; 2 NAD → 2 reduced NAD

↓

intermediates

↓ → 2 ATP

2 molecules pyruvate (3C)

net yield: 2 ATP
2 reduced NAD

Glycolysis - phosphorylation of glucose and the subsequent splitting of fructose 1, 6 - bisphosphate (6C) into two triose phosphate molecules, which are further oxidised to pyruvate with a small yield of ATP and reduced NAD.

c) Link reaction * O_2 available

via active transport :

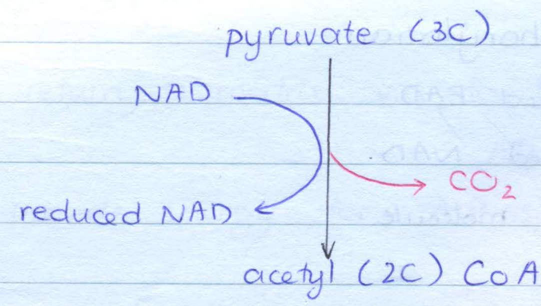
pyruvate from cytoplasm ⟶ mitochondrial matrix

pyruvate

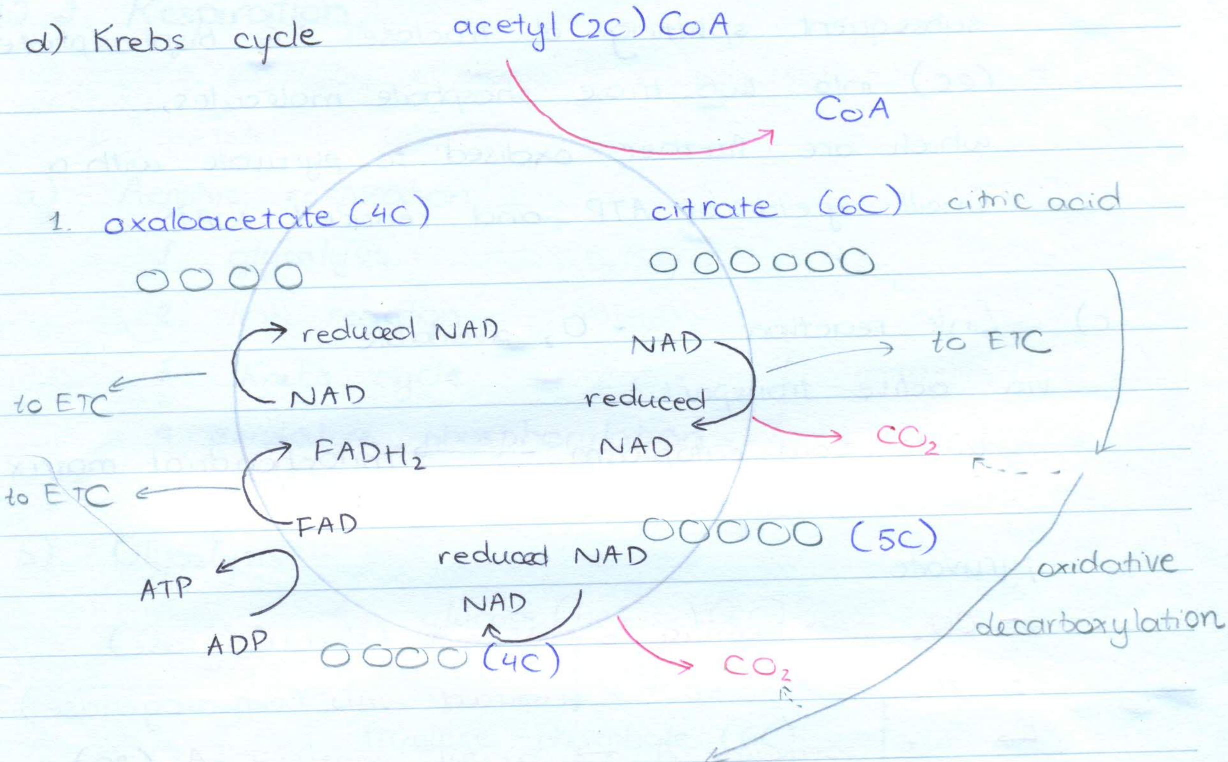
- $-CO_2$ ⟶ diffuse out of cell (decarboxylase)
- $-H$ ⟶ + NAD → reduced NAD (dehydrogenase)

↳ + coenzyme A → acetyl coenzyme A (2C)

pyruvate + CoA + NAD ⇌ acetyl CoA + CO_2 + reduced NAD



d) Krebs cycle



+ decarboxylation $(-CO_2)$

dehydrogenation $(-H)$

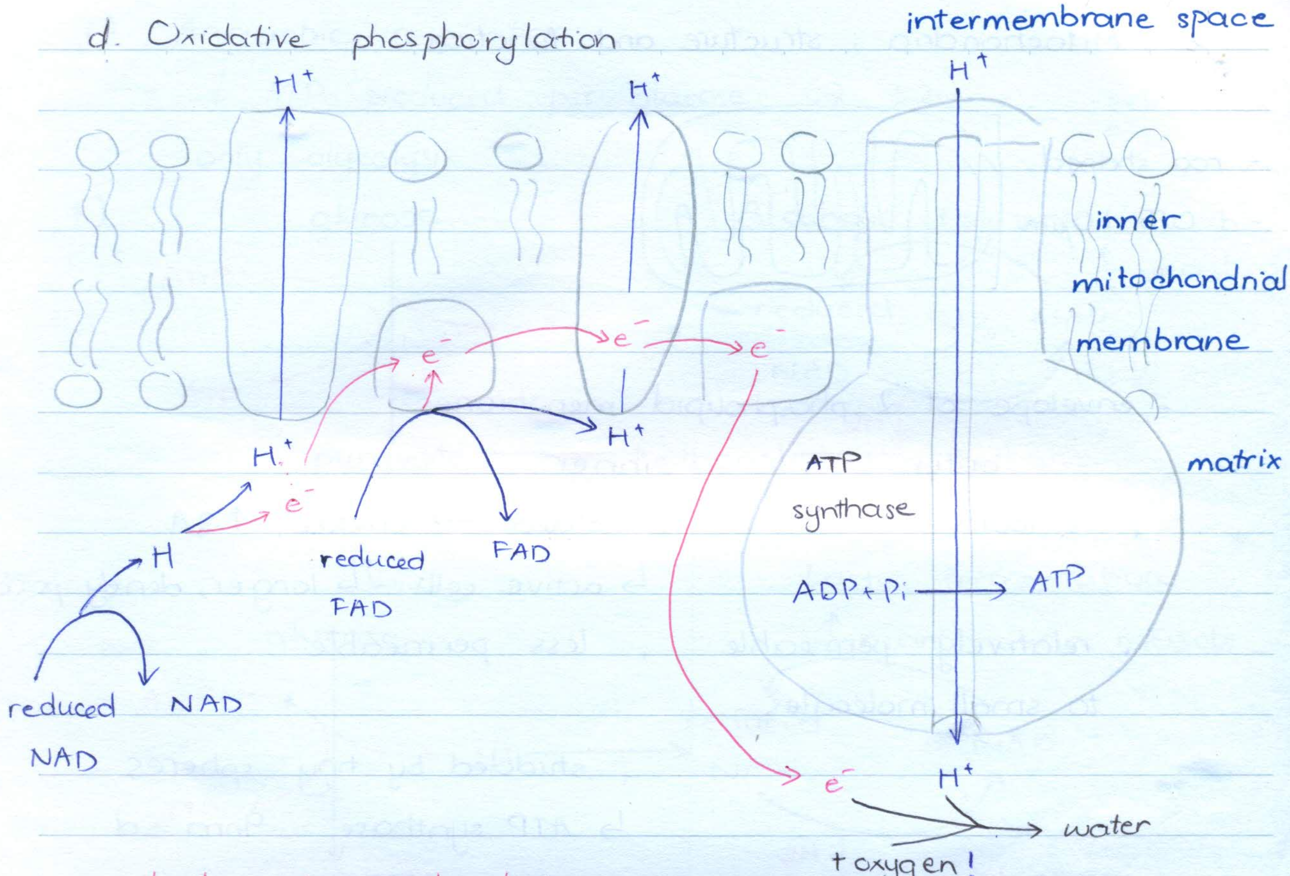
+ reduction of NAD and FAD

yield - ×2 CO_2 molecules

- ×1 reduced FAD
- ×3 reduced NAD
- ×1 ATP molecule

↳ for each repeat of glycolysis, requires 2 Krebs cycles (2 pyruvates

d. Oxidative phosphorylation



- electron transport chain

- energetic electrons release energy as they pass through ETC. from matrix → imspace

  ↓ transfer protons across membrane

  set up proton gradient

- protons return to matrix via facilitated diffusion

  ATP synthase

  ↳ provide energy for ATP synthesis

! O_2 = final electron acceptor

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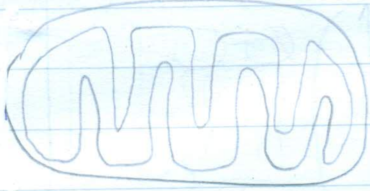
i. Mitochondria : structure and function

- rod shaped
- d: 0.5-1.0 μm



- envelope of 2 phospholipid membranes

outer	inner
+ smooth	+ folded → cristae, ↑ SA
	↳ active cells: ↑↳ longer, densely pack
+ relatively permeable to small molecules	+ less permeable
	+ studded by tiny spheres :
	↳ ATP synthase : 9nm = d
	+ site of ETC

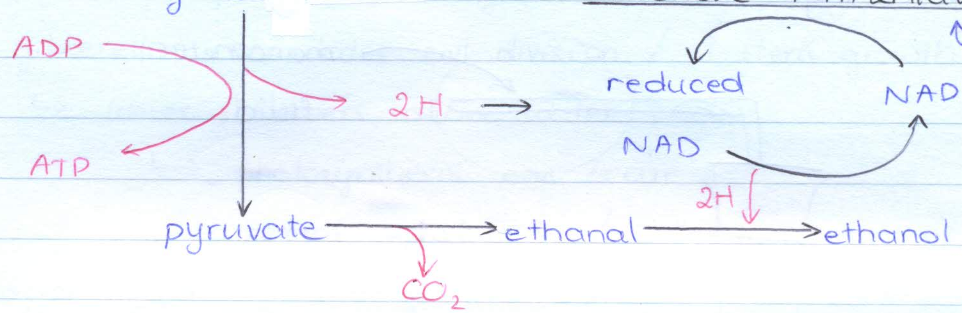
- pH intermembrane space < pH matrix
  ↳ H^+ protons released by ETC

- matrix — site of link reaction / Krebs cycle
  - 70S ribosomes
  - ↑ # looped mitochondrial DNA.

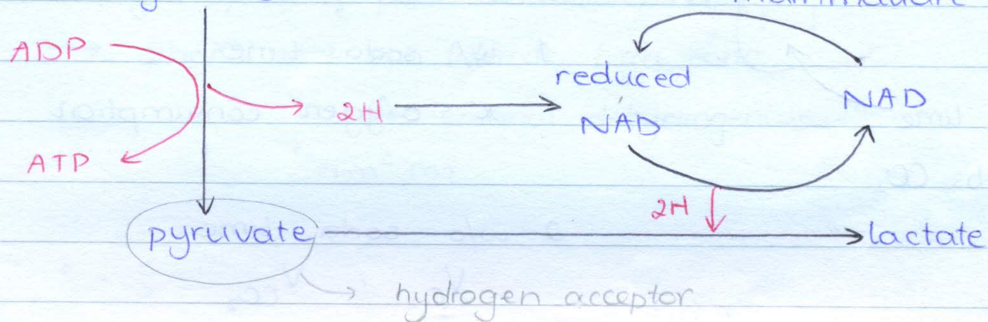
j) Anaerobic respiration

- ↓ ATP produced per glucose (2 : 32)
- only glycolysis

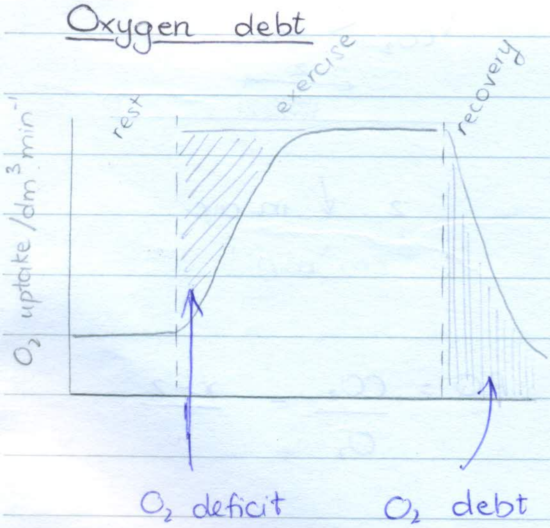
k) **Alcoholic fermentation** ↑ yeast



Lactic fermentation ↑ mammalian muscle



Oxygen debt



- strenuous exercise
→ lactic fermentation in muscles.
→ builds up O_2 deficit
→ post-exercise uptake extra O_2

V_{O_2} required after exercise to metabolise lactate from anaerobic respiration.

1820 R 530 R 550 T 740

+100 +100

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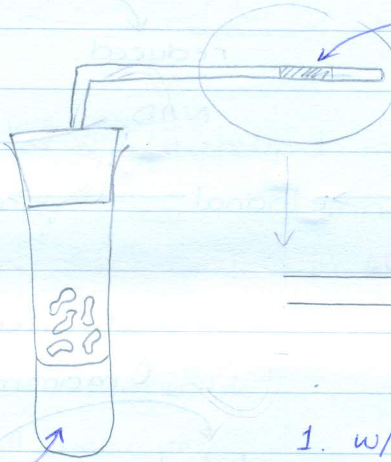
12.1. g)

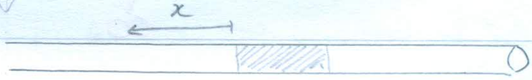
Respiratory quotient (RQ): the ratio of the volume of carbon dioxide given out in respiration to that of oxygen used.



manometer fluid

soda lime

↳ absorbs CO_2



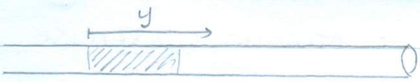
1. w/ soda lime

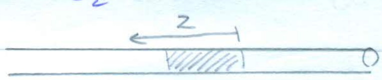
x: oxygen consumption $cm^{-3} min^{-1}$

2. w/o soda lime

V_{O_2} ? V_{CO_2} $\gtrless$ = ?

$V_{CO_2} > V_{O_2}$ | $V_{CO_2} < V_{O_2}$





y: ↑ in air $cm^3 min^{-1}$

z: ↓ in air $cm^3 min^{-1}$

$RQ = \frac{CO_2}{O_2} = \frac{x+y}{x}$ per unit time

$RQ = \frac{CO_2}{O_2} = \frac{x-z}{x}$

m) $t°$↑ = respiration rate ↑

Rice adaptation to flooded areas

- ethene
- stimulates production of gibberellin
- gibberellin stimulates cell division / ↑ stem growth
  - ↳ leaves / flowers above water
    - ↳ _ photosynthesis can occur
    - _ sexual reproduction / pollination can occur
- aerenchyma : assists gas diffusion
  air can be trapped by underwater leaves
- submerged parts of plant : anaerobic respiration
  → ethanol → can tolerate high conc
  ↑ ethanol dehydrogenase

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13. Photosynthesis

13.1 . Photosynthesis as an energy transfer process

a) - Energy from light trapped by chlorophyll (LDR)

- split bonds in H_2O → release H
- produce ATP
- reduce NADP

} produce complex organic molecules eg : glucose

oxygen ↑ light dependent reaction (LDR) —— ATP ——→ light independent reaction (LIR) → glucose ↑

in grana- Thylakoid membrane

(LDR) —— NADPH ——→ (LIR)

light energy → (LDR)

CO_2 → (LIR)

in stroma

c) chloroplast pigments
↳ dif pigment = dif λ

- chlorophylls
  - α
  - β

  } red, blue, violet
- carotenoids
  - β carotene
  - xanthophyll

  } blue-violet

d) * absorption spectrum: graph of absorbance of different λ of light by a pigment

* action spectrum: rate of photosynthesis at different λ of light (graph)

$2H_2O \xrightarrow{light} 4H^+ + 4e^- + O_2$

f). Light dependent reaction (photolysis

↳ synthesis of ATP in phosphoryl

- photolysis: split water give H^+

→ H^+ + NADP → NADPH

→ $2H_2O \xrightarrow{light} 4H^+ + 4e^- + O_2$

↓

picked up by chlorophyll in PSII.

LDR: photoactivation of chlorophyll

→ . photolysis of water (in PSII)

- transfer of energy to ATP and reduced NADP

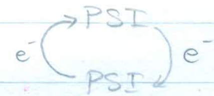
NADP → cyclic

non cyclic

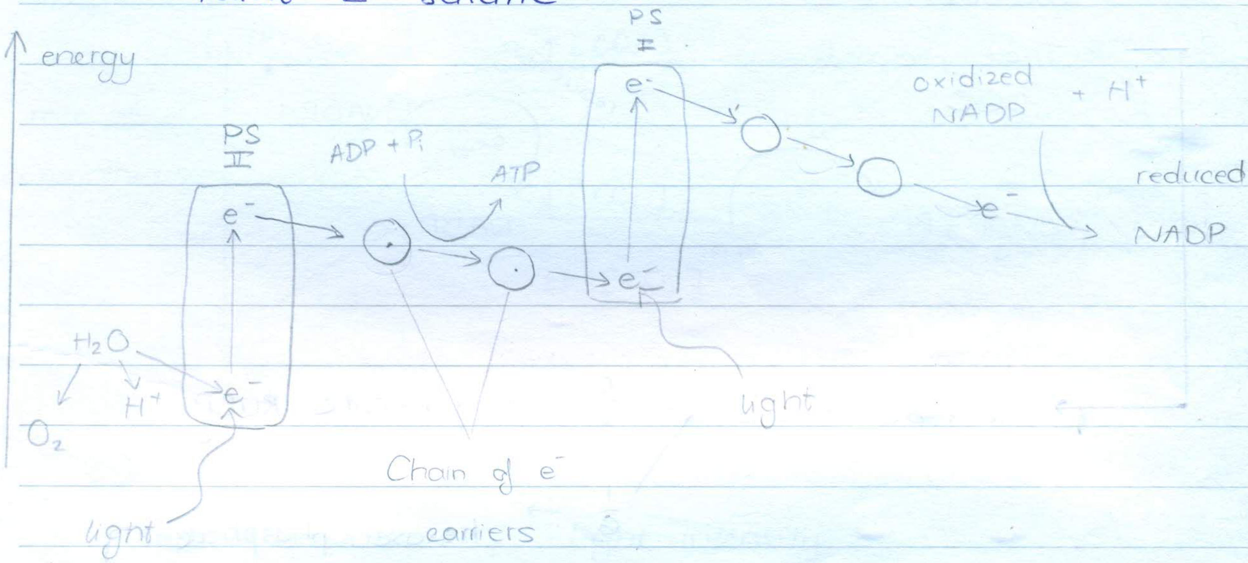
Phosphorylation

photophosphorylation ← cyclic

← non-cyclic



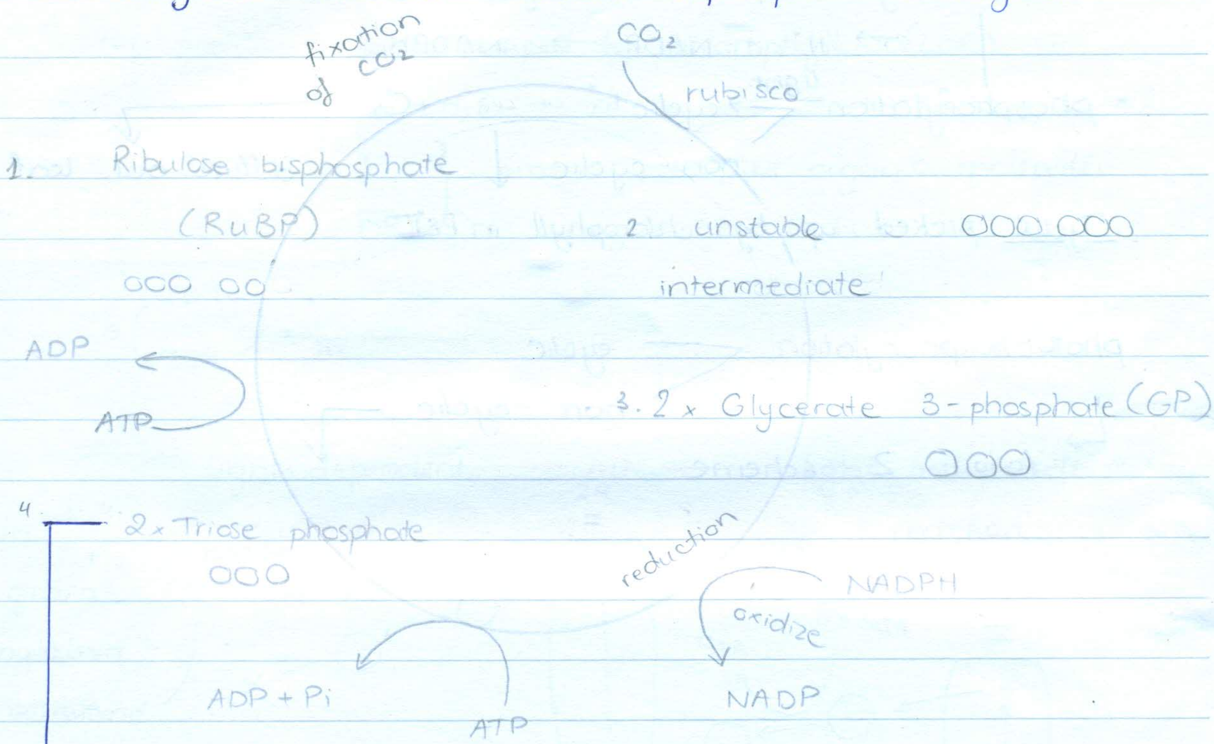
↳ follow 2 scheme



- e^- taken by PSII from photolysis of water
- e^- energy raised (chlorophyll PSII absorb light energy)
- e^- lose energy along ETC
- e^- taken by PS I
- energy ↑
- becomes part of reduced NADP molecule

g) The light-independent reaction: Calvin cycle

- site: stroma
- enzyme 'rubisco': ribulose bisphosphate carboxylase.



$2 \times 3C = 6C$ — $\frac{5}{6} \rightarrow$ generate RUBP

— $\frac{1}{6} \rightarrow$ hexose phosphate

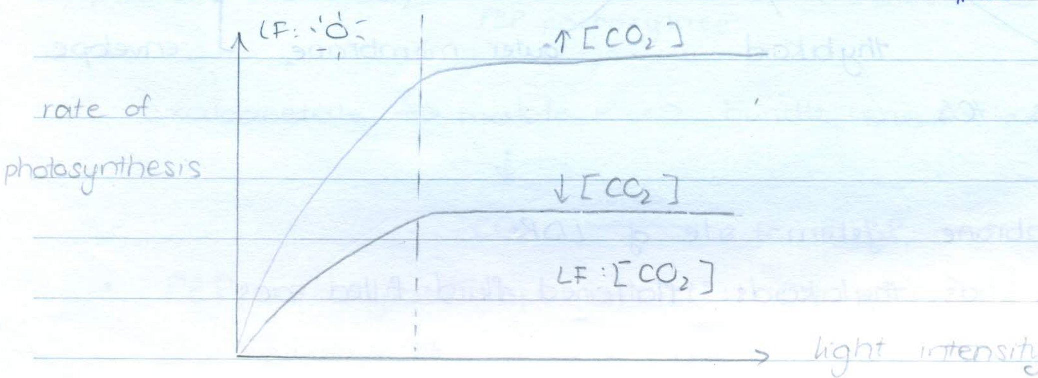
- starch for storage
- sucrose for translocation
- cellulose for cell walls
- glycerol, fatty acid → lipid
  + cellular membranes
  + acetyl coenzyme A — respiration
    — amino acid production
    ⟶ protein synthesis

13.2. Investigation of limiting factors

Environmental factors influence the rate of photosynthesis

λ + - light intensity: affect LDR → e transferred in light rays

- t°: affect LIR → KE
- conc CO_2 in atmosphere : reactant in photosynthesis
  ↳ 0.04%
- availability of water: reactant in photosynthesis
  (usually not a problem)
  ↳ indirectly : ↓ H_2O = stomata ↓ = CO_2 can't diffuse into leaf



c) Glasshouses for crops

- sensors monitor: + light intensity
  + humidity of atmosphere
  + conc CO_2 around plants
- grown hydroponically: roots → nutrient solution
  → content varied at dif stages of plant growth.
- control insect pests + fungal diseases

⟶ maximise crop yield.

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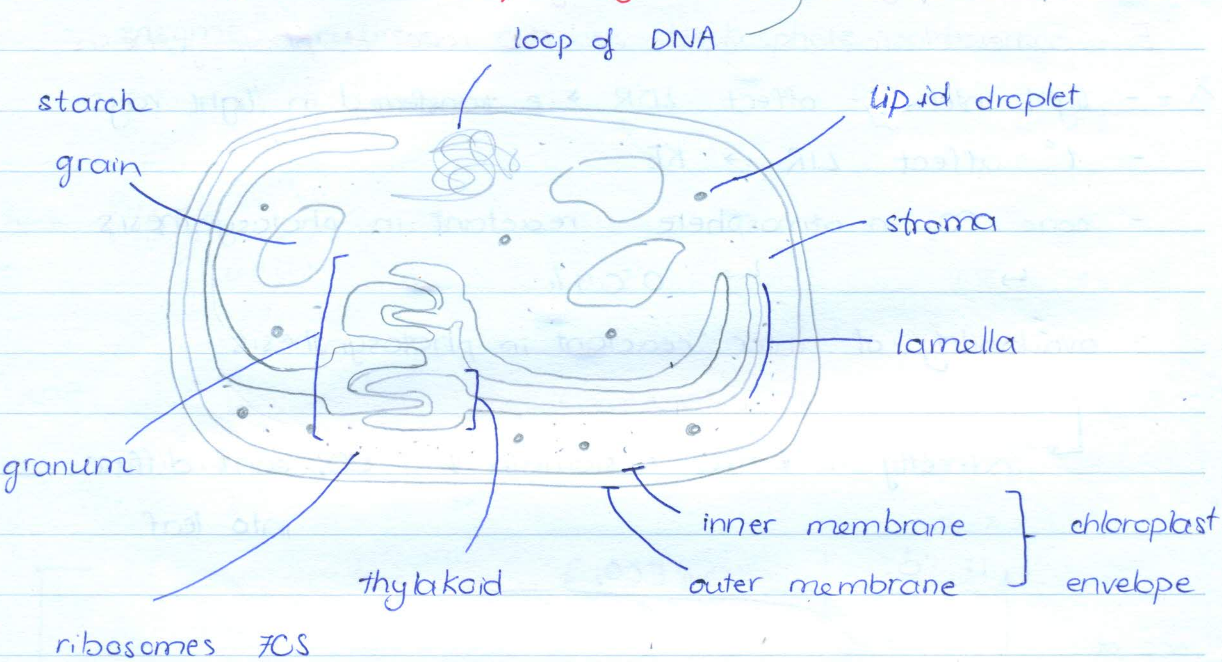
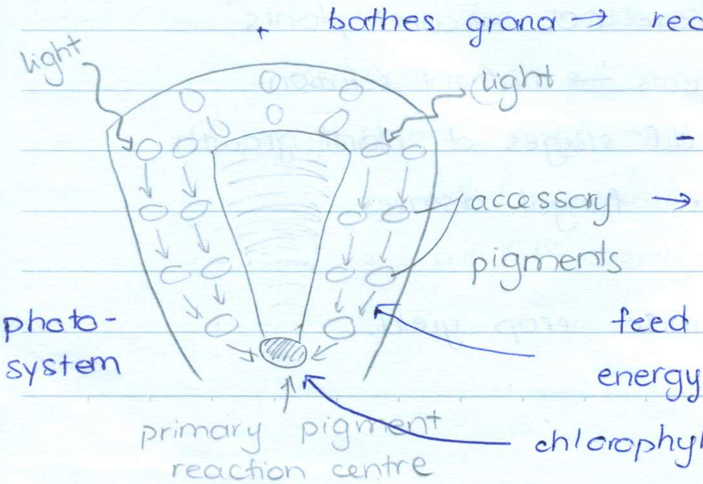
13.3. Adaptations for photosynthesis



- membrane system: site of LDR
  + has thylakoids: flattened fluid-filled sacs
    → stacks: grana
  + grana: ↑SA : holds pigments, enzymes (ATP synthase),
    electron carriers for LDR → chemiosmosis
- stroma: site of LIR
  + contains enzymes of Calvin cycle, sugar, a.acid
  + bathes grana → receive product from LDR



- arranged pigment molecules → light harvesting clusters

b) C4 plants

- rubisco catalyses
  - CO_2 + RuBP
  - O_2 + $\underbrace{\text{RuBP}}$ @ ↑t° ↳ wasted

Avoiding phosphorespiration

↳ • keep rubisco and RuBP away from O_2

↓

cells → vascular bundles : bundle sheath cells

no direct air contact

- CO_2 absorbed by mesophyll cells:

$$PEP + CO_2 \xrightarrow[\text{PEP carboxylase}]{} \text{oxaloacetate}$$

oxaloacetate → malate - - -> bundle sheath cells

↓

$$CO_2 \xrightarrow{\text{rubisco}} \text{to RuBP} \longrightarrow \text{LIR}$$

• PEP carboxylase optimum t° C4 45°C > C3 30°C

- photorespiration: wasteful reaction. RUBP combines with O_2 rather than CO_2 favoured by ↑t° + ↑☼

C4 - 1st pro compound of LIR has 4C

PEP - phosphoenol pyruvate

- Light energy absorbed by chlorophyll

ETC photophosphorylation

- e^- excited, raised to higher energy level
- e^- emitted by chlorophyll and gets passed on to e^- carriers
- e^- passes along ETC, energy produced is used to pump protons into thylakoid space (thylakoid membrane impermeable to protons)
- proton gradient forms, proton moves down gradient through ATP synthase
- enzyme rotates
- ATP produced from ADP + Pi

14. Homeostasis

14.1. Homeostasis in mammals

↳ maintaining a relatively constant environment for the cells within the body

- homeostatic mechanisms control composition of blood
  → controls composition of tissue fluid (around cells)

- core body t°
- blood pH
- metabolic waste e.g.: CO_2 and urea
- blood glucose concentration
- Ψ in blood
- conc of respiratory gases in blood: O_2, CO_2

Negative feedback control loop

- internal + external stimuli
- negative feedback: a process in which Δ parameter, such as blood glucose level, brings about processes which moves its level back towards normal again

receptors (sensors) detect Δ in stimuli/factor → Effectors receive info from receptor

factor rises above set point

factor falls back to set point

factor fluctuates around set point

factor falls below set point → " → " → " increases

effectors: muscles and glands

- homeostatic mechanisms require transfer of information
  - nervous system : ↯ impulses along nerve cells (neurones)
  - endocrine system : chemical messengers (hormones)
    travel in blood, long distance cell-signaling.

  ↳ thermoregulation , osmoregulation , control blood glucose conc.

Excretion

∴ removal of unwanted products of metabolism

→ deamination : liver removes amino group from protein ($-NH_2$)

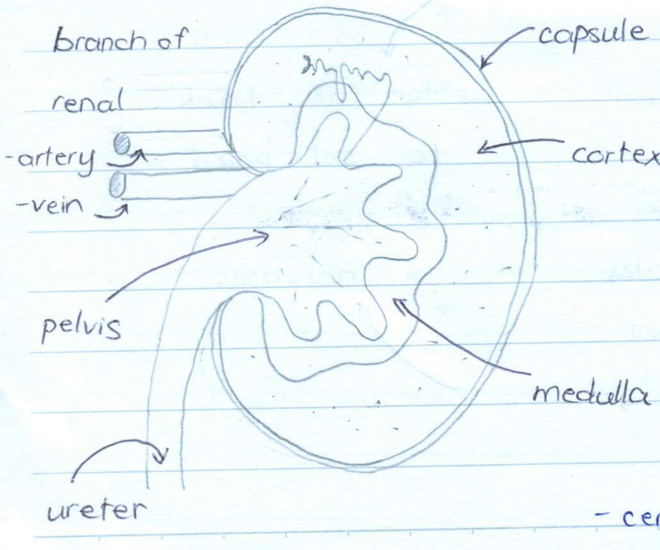
$$\text{amino acid} \xrightarrow[NH_3]{H_2O} \text{keto acid} \text{ — respired / converted into glucose / fat} \nearrow \text{Krebs cycle}$$

Urea cycle

$$2NH_3 + CO_2 \longrightarrow \underbrace{CO(NH_2)_2}_{\text{urea}} + H_2O$$

Urea : main nitrogenous excretory products

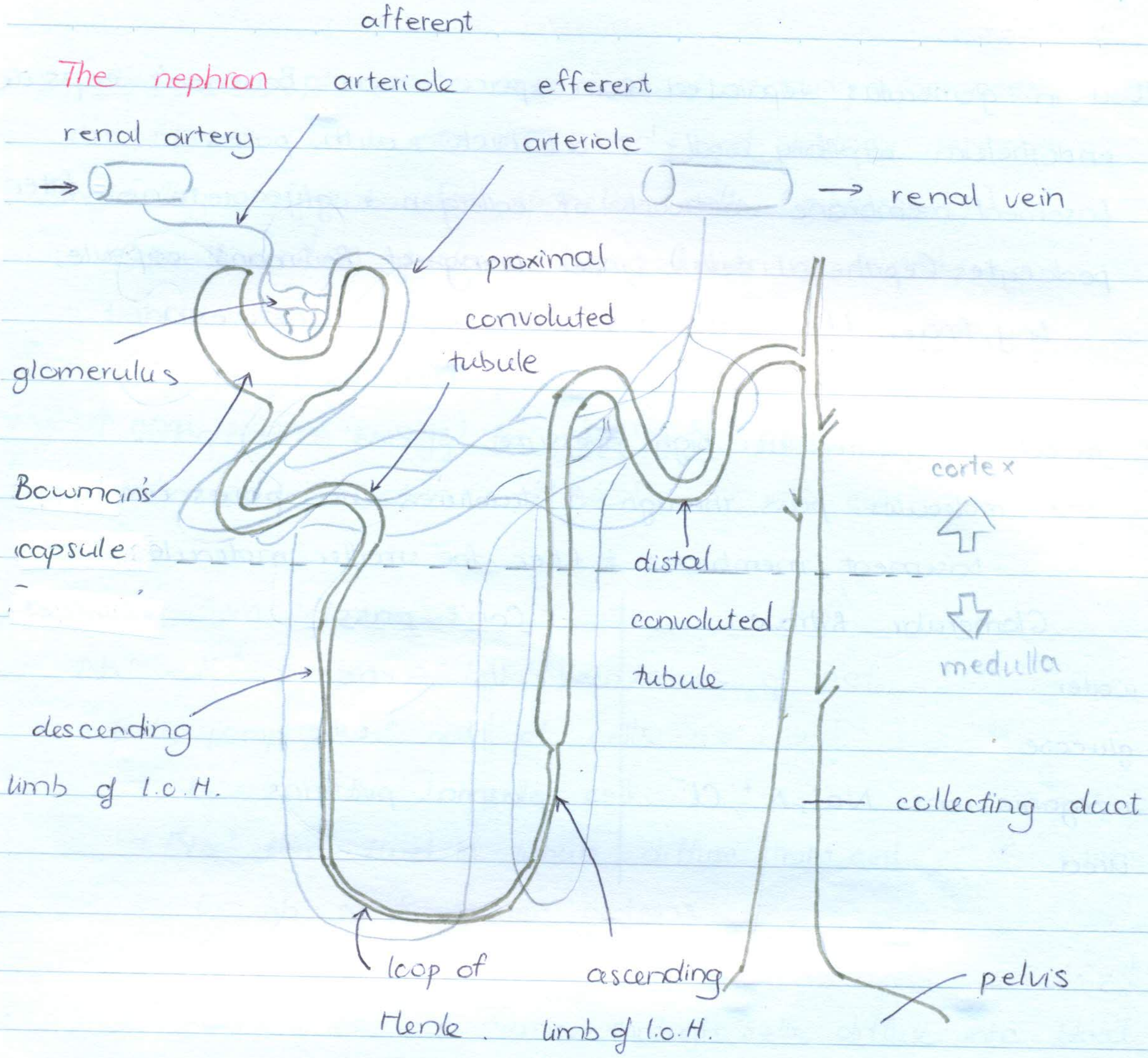
↳ diffuse from liver ⟶ blood plasma ⟶ kidney filters urea ⟶ excreted



Kidney

- receives blood : renal artery
  returns " : " vein
- ureter : narrow tube carries urine from kidney to bladder
  → urethra : urine to outside body
- kidney covered by tough "capsule"
- beneath is the "cortex".
- central area : "medulla"
- pelvis : where ureter joins with medulla

The nephron



Each nephron has a network of blood vessels associated withit

- blood from renal artery → afferent arteride → glomerulus in the cup of Bowman's capsule → efferent arteriole → network of capillaries running alongside the nephron → renal vein
- Bowman's capsule → proximal convoluted tubule → loop of Henle → distal convoluted tubule → collecting duct → pelvis
- kidney makes urine in 2-stage process
  1. ultrafiltration
  2. selective reabsorption

1. Ultrafiltration

Blood in glomerulus separated from space inside Bowman's capsule:

- endothelium: capillary wall - 1-cell thick, with pores
- basement membrane: network of collagen + glycoproteins = filter
- podocytes (epithelial cells): inner lining of Bowman's capsule; tiny, finger-like projections with gaps in between

- blood in glomerulus: high pressure (efferent smaller than afferent)
  ↳ - molecules pass through 3 structures via pores and gaps
  - basement membrane = filter for smaller molecules

Glomerular filtrate	Can't pass
- water	- cells: rbc, wbc
- glucose	- plasma proteins
- inorganic ions: Na^+, K^+, Cl^-	
- urea	

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2. Reabsorption in the proximal convoluted tubule

selective reabsorption: The movement of certain substances from the filtrate back into the blood and kidney nephron

Single layer of cuboidal epithelial cells (pct lining)

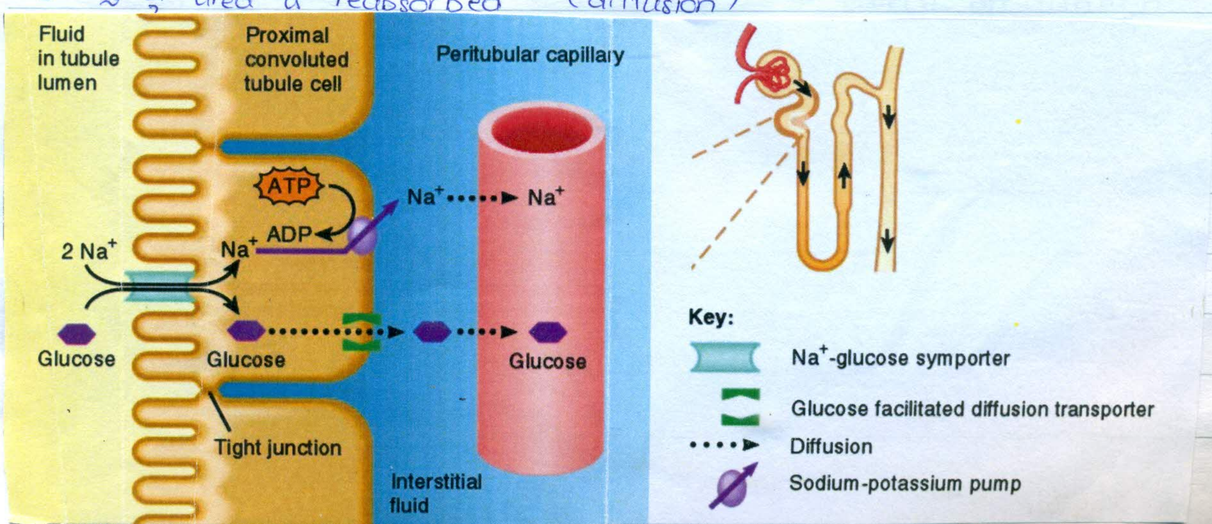
- microvilli : ↑ SA surface facing lumen , folded
- tight junctions: hold adjacent cells together
  ↳ fluid can't pass between cells
- ↑ mitochondria: energy for Na^+ + K^+ ~~pump~~ pump proteins
- cotransporter protein ↳ out of → into cell

Blood from glomerulus

- Na^+ - K^+ pumps in basal membranes of pct
  actively pump Na^+ out of cells ⟷ blood
  → lowers $[Na^+]$ inside cell
  → $[Na^+]$ from fluid in tubule diffuse into cell
  through co-transporter proteins
  ↳ carry glucose with Na^+
  ↳ move through cells, diffuse into blood

amino acids
vitamins
Na^+, Cl^-
reabsorbed

into blood
- movement Na^+ + glucose = ↓ Ψ in blood
  water (via osmosis): from tubule → cell → blood into circulation
- ≈ ½ urea is reabsorbed (diffusion)

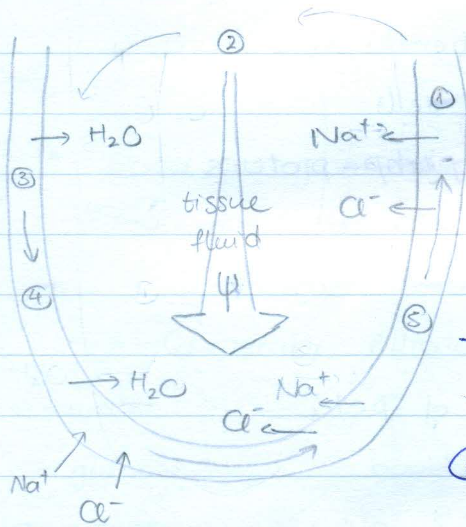


3. Reabsorption in the loop of Henle & collecting duct

Loop of Henle: build high conc of Na^+ and Cl^- in medulla

↳ allows highly concentrated urine to be produced

→ preserve water → prevent dehydration



descending: permeable to Na^+, H_2O

ascending: " " "

impermeable to H_2O

① Na^+, Cl^- actively transported out ascending limb → tissue fluid

→ ② ↑ [Na^+, Cl^-] in tissue fluid = ↓ψ

↑ψ in descending limb

③ H_2O in filtrate → osmosis → tissue fluid from descending limb

④ • Na^+, Cl^- diffuse into loop

→ fluid more concentrated towards bottom of loop

⑤. so conc → Na^+ Cl^- diffuse out of ↑limb → tissue fluid

↳ lost as go up

Osmoregulation

↳ control of Ψ of body fluids

- involves : hypothalamus, posterior pituitary glands, kidney

$\Psi\downarrow$ in blood

osmoreceptors → ϟ along neurone → posterior pituitary gland → stimulate ADH release (antidiurectic hormone)

detect

↳ specialized sensory neurones in hypothalamus

- peptide hormone
- $\downarrow$ H_2O loss in urine

[stimulating H_2O reabsorption in kidneys

ADH : acts on cell-surface membrane of collecting ducts → more permeable to water

Thermoregulation - control of body t° (core t°)

- central thermoreceptors in hypothalamus and spinal cord detect Δ blood t° → send impulses

In the cold		In the heat
- Vasoconstriction	physio-	- Vasodidation
- Shivering	logical	- lowering body hairs
- Raising body hairs	responses	- ↑ sweat production
- ↓ sweat production		
- ↑ adrenaline secretion		

+ behavioural responses ← curling up / spreading out; air con / thick clothes

- hypothalamus release hormone → stimulate anterior pituitary gland to secrete thyroid stimulating hormone (TSH)

→ stimulate thyroid gland to secrete thyroxine

- thyroxine : ↑ metabolic rate - ↑ heat production esp. in liver

t° ↑ ⇒ ↓ TSH production

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Controlling blood glucose

glucose ⟷ glycogen

↳ is controlled by hormones secreted by endocrine tissue in pancreas
↓
islets of Langerhans

- α cells : secrete glucagon
- β cells : secrete insulin

⇒ - receptors
- central control

⇒ coordinate actions of effectors

[negative feedback control mechanism]

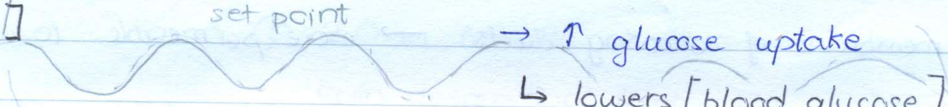
- less glucagon
- more insulin

<u>Receptors:</u>
α + β cells
detect rise

<u>Effectors:</u>
- liver responds less glucagon
→ no glycogen breakdown
- liver, muscle, fat cells : ↑ insulin
→ ↑ glucose uptake
↳ lowers [blood glucose]

high
[blood glucose]

set point



- cyclic AMP: second messenger ; response to glucagon + adrenaline
- cell signaling in the control of blood glucose
  - hormone - receptor interaction at cell surface
  - formation of cyclic AMP → binds to kinase proteins
  - an enzyme cascade involving activation of enzymes by phosphorylation to amplify the signal

- hormone - receptor interactions at cell surface membrane

glucagon binds to receptor molecules at ↑

→ activates G protein → activates enzyme

$$ATP \xrightarrow{\text{catalyse}} \text{cyclic AMP} \text{ (2nd messenger)}$$

- cyclic AMP + binds with kinase enzymes (in cytoplasm)

  → phosphorylates other enzymes

↳ enzyme cascade : amplifies original signal of glucagon

Urine Analysis

- presence of glucose and ketones in urine ≈ diabetes

  ↳ [ ] ↑ above renal threshold

  = not all glucose was reabsorbed from filtrate in PCT

- protein in urine for long periods of time

  ≈ kidney damage / infection ≈ high blood pressure

Glucose Analysis

1. Dip sticks : test for glucose, pH, ketones, proteins

- urine analysis
- 2 immobilized enzymes: glucose oxidase, peroxidase

$$\text{glucose} \xrightarrow{\text{glucose oxidase}} \text{gluconolactone} + H_2O_2$$

$$H_2O_2 + \text{colorless chemical (chromogen)} \xrightarrow{\text{peroxidase}} \text{darker compound (brown?)}$$

* shows sugar level in urine from bladder

NOT current blood sugar level

2. Biosensors

- blood analysis - quantitative data
- a pad impregnated with glucose oxidase catalyses reaction to form gluconolactone

⟶ generates tiny electric current

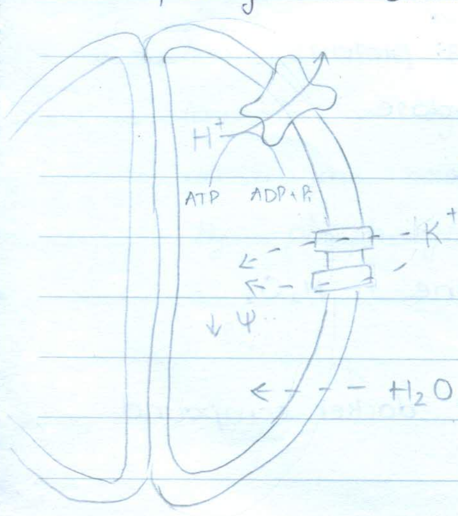
detected by electrode ⟶ read by meter

14.2. Homeostasis in plants

* stomata have daily rhythms of opening and closing and also respond to Δ environmental conditions to:
  - allow diffusion of CO_2
  - regulate water loss by transpiration

open stoma	closed stoma
- ↑ light intensity	- darkness
- ↓ $[CO_2]$ in air spaces	- ↑ $[CO_2]$ in air spaces
	- low humidity - ↑ t°
	- water stress

Opening and closing of stomata



1. ATP - powered proton pumps → actively transport H^+ out
2. low $[H^+]$ and -ve charge inside cell → K^+ channels open → diffuse in
3. high $[K^+]$ in cell ↓ Ψ
4. H_2O moves in via osmosis
5. water entry ↑ $V_{guard\ cell}$ ⟶ expand

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Structure of stomata

- each stomatal pore surrounded by 2 guard cells
- guard cells: turgid: → open (gain H_2O)
  flaccid: → close (lose H_2O)

Absaisic acid & stomatal closure @ (v) water stress

↳ ABA = stress hormone (e.g.: - drought
- high t°)
- closes stomata

↳ reduce transpiration : ↓ water loss

ABA: - binds to surface cell receptors
→ inhibits proton pumps: stop H^+ pumped out
- stimulates movement Ca^{2+}
through c.s. membrane, cytoplasm, tonoplast

Ca^{2+}: 2nd messenger
- activate channel proteins: -ve ions leave cell
→ open " " allow K^+ leave
close " " K^+ Ø enter

net movement: K^+ leaves

loss of ions = ↑ Ψ inside cell · H_2O passes out by osmosis = guard cells → flaccid = stomata close

15. Control and co-ordination

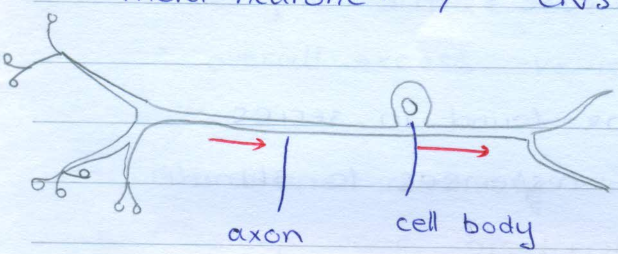
15.1. Control and coordination in mammals

a) Communication systems that co-ordinate responses to changes in the internal + external environment

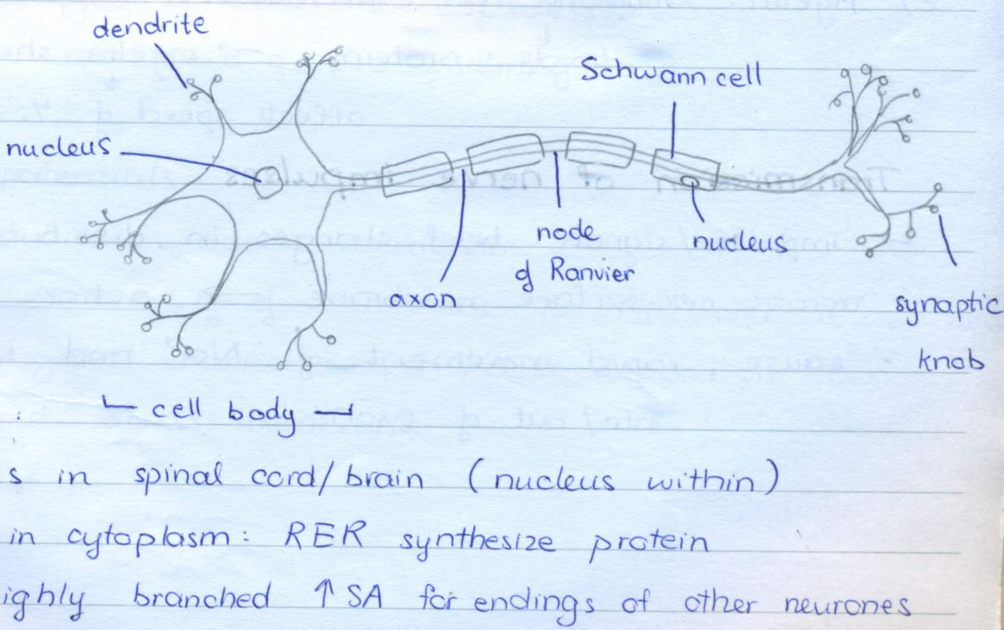
- endocrine systems : glands secrete hormones
  → chemical messengers travel in blood
- nervous systems : nerves transmit info : ϟ impulses

b) Structure of sensory & motor neurones

- sensory neurone : transmit ϟ : receptors → CNS
- intermediate / relay : ϟ sensory neurone ⟶ motor neurone
- motor neurone : ϟ : CNS → effectors



Sensory neurone :
- one long axon with cell body
- near source of stimuli / swelling



Motor neurone :

- cell body lies in spinal cord / brain (nucleus within)
- dark specks in cytoplasm : RER synthesize protein
- dendrites : highly branched ↑ SA for endings of other neurones
- axon : conduct impulse over long distances , ↑ mitochondria + vesicles + transmission substances

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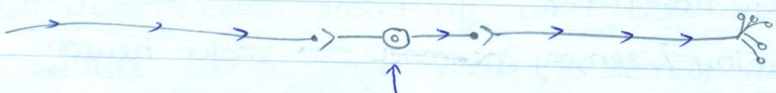
c) Sensory receptor cells: detect stimuli → stimulating transmission of nerve impulses in sensory neurones

d) Reflex arc : pathway along which impulses are transmitted from a receptor → an effector without involving 'concious' regions of the brain (heat / light / pressure / pain)

receptor $\xrightarrow[\text{neurone}]{\text{sensory}}$ CNS $\xrightarrow[\text{neurone}]{\text{motor}}$ effector



relay neurone

- sensory, relay, motor neurons found in series
  ↳ controls fast, automatic responses to stimuli

e) Myelin: Schwann cells (specialized) wrapped along axon
↓ lipids + proteins → myelin sheath
↓ affects speed of $\frac{4}{i}$ conduction

Transmission of nerve impulses
↳ impulses/signals: brief changes in distribution of $\frac{+}{-}$ charge across cell surface membrane → action potentials
- cause: rapid movement of Na^+ and K^+ into/out of axon.

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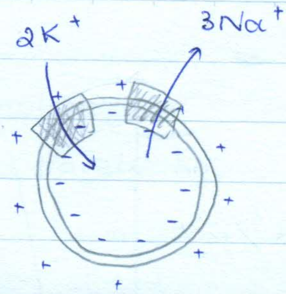
① Resting potential



- inside of axon: slightly -ve
- potential dif: -60mV to -70mV
  = resting potential: less than outer

- potential produced + maintained by $Na^+ - K^+$ pumps
  - membrane proteins
  - use energy from hydrolysis of ATP : active transport
- • more channels for K^+
  - large, -ve molecules inside cell attract K^+
  → less K^+ diffuse out
  → overall excess -ve ions inside membrane

- membrane relatively impermeable to Na^+
- • steep conc gradient } electrochemical
  - -ve charged inside } gradient
  → inward movement of Na^+ during a.p.

② Action potentials
↳ rapid Δ p.d. across membrane
cause: Δ permeability of membrane to Na^+ and K^+
- voltage-gated channels: open / close depend on Δ p.d. across membrane

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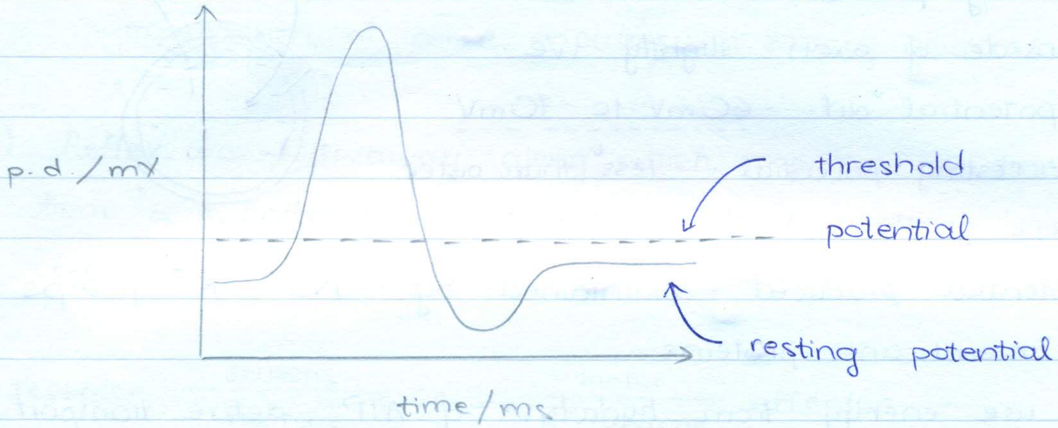
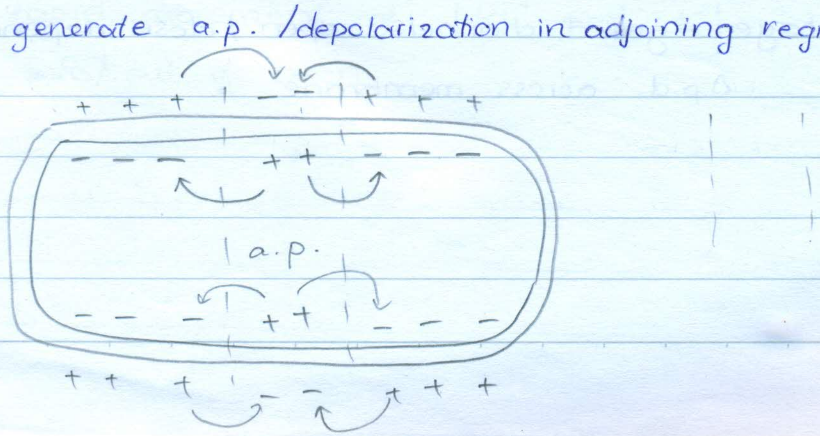


a.p.

1. depolarization: Na channels open → Na^+ enter
   → p.d. less negative on the inside
   (+ve feedback : small depol lead to larger depol)
2. p.d. reach threshold potential → generate action potential
3. repolarization: Na^+ channels close, K^+ channels open
   → outward movement of K^+ removes +ve charge inside axon.
4. refractory period: axon is unresponsive : recovering from an a.p. → another a.p. cannot be generated

transmission of a.p. : a.p. in 1 cell triggers a.p in another
↳ temporary delocalisation of membrane sets up a 'local circuit' between depolarized + resting regions
→ generate a.p. /depolarization in adjoining regions



How a.p. carry info:

- a.p. have same — size (same amplitude)
  - speed which a.p. travels by
- dif: frequency
  - # of neurones carrying a.p.

→ get info of strength of stimulus

- nature of stimulus: deduced from position of sensory neurone

Initiation of an a.p.:

- receptor cell: responds to stimulus → generate a.p.
  - ↳ transducers: transform energy of one form (light, heat ...) → ϟ impulse in neurone

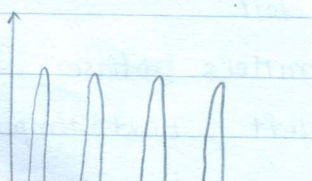
depolarized

→ receptors stimulated : receptor potential above threshold

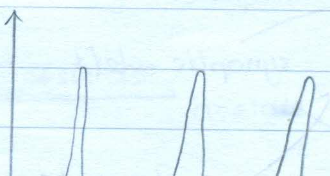
→ a.p. initiated → stimulates sensory neurones send impulses to CNS

- all-or-nothing law: neurones either do/do not transmit ϟ

* threshold levels rarely stay constant

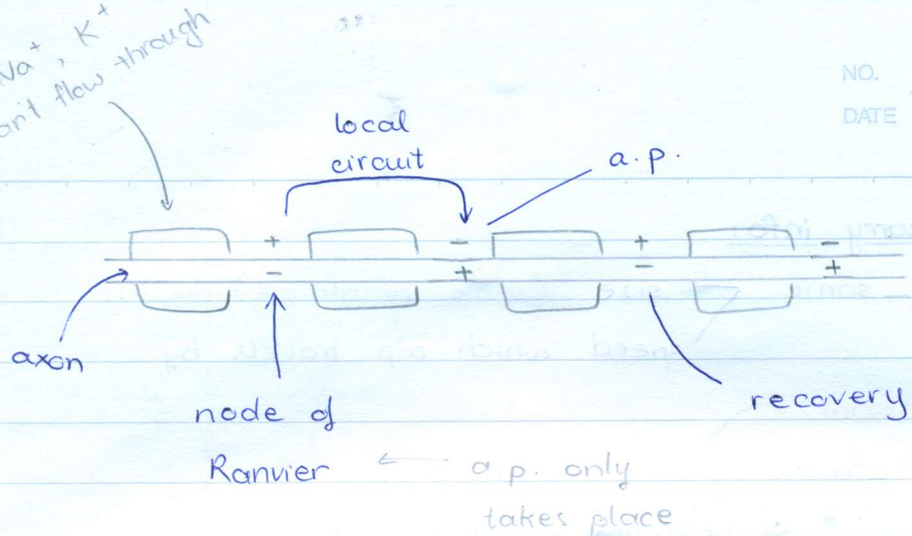


strong stimulus



weak stimulus

f)



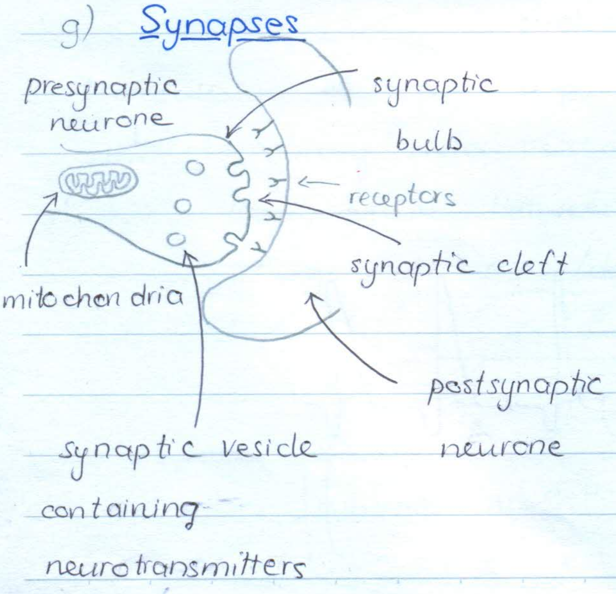
Speed of conduction:

- Myelin insulates axon membrane → speed up rate that a.p. travel
- "local circuits" exist from one node to the next
  ↳ saltatory conduction: a.p.: jump from one node to the next
- with myelin: speed of transmission x 50

- diameter ↑ = ↓ resistance = faster transmission

* refractory period determines max. frequency of impulses

g) Synapses



- AP occur at membrane of presynaptic neurone
- ↳ release neurotransmitters into synaptic cleft
- ↳ neurotransmitters diffuse across cleft, bind temporarily to receptors on postsynaptic neurone
- ↳ neurone response: depolarising; reach threshold → send impulses

* cholinergic synapses: synapses that use ACh as neurotransmitters

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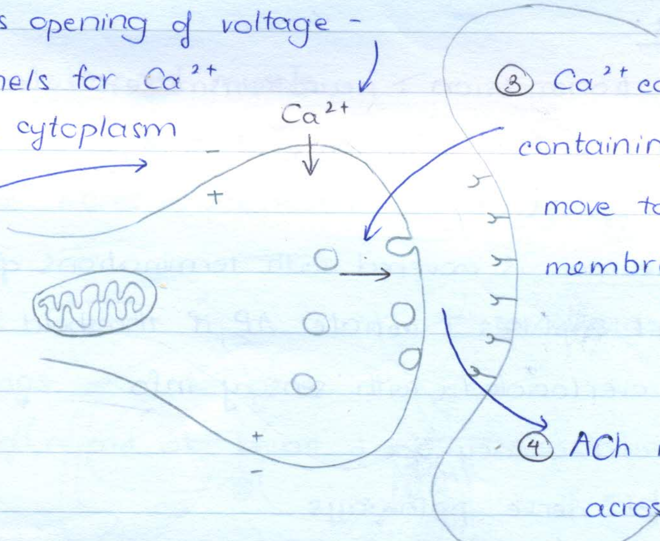
Synapse: point where 2 neurones meet but do not touch
↳ end of presynaptic neurone + synaptic cleft + end of postsynaptic neurone

Mechanism of synaptic transmission

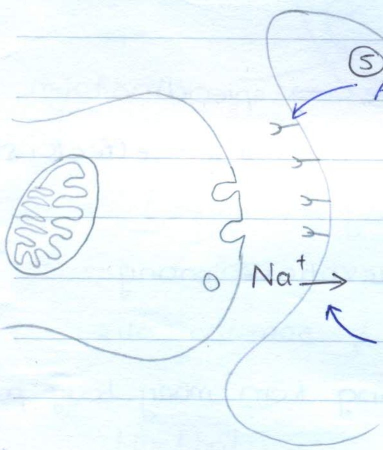
② stimulates opening of voltage-gated channels for Ca^{2+}
→ diffuse → cytoplasm

① AP arrives



③ Ca^{2+} cause vesicles containing acetylcholine move towards presynaptic membrane → fuse

④ ACh released, diffuses across cleft



⑤ ACh binds to receptor proteins on postsynaptic membrane
→ chemically-gated ion channels for Na^{+} open

⑥ Na^{+} diffuse through
→ depolarizes membrane
↳ AP generated (threshold reached)

⑦ recycling

ACh: ACh $\xrightarrow{\text{acetylcholinesterase}}$ acetate + choline
→ stops continuous production of APs

⑧ choline moves back into presynaptic neurone
choline + acetyl coenzyme A → ACh
ACh → transported to presynaptic vesicles
↳ ①

* ↑ chances AP generated and impulses sent :
  - > 1 presynaptic neurone releases ACh at the same time
  - " " " " " over short time period

Role of synapses :

1. ensure one-way transmission : neurotransmitters released from one side; receptors on the other side of synapses
2. integration of impulses

   Body of motor neurone is covered with terminations of relay neurones
   ↓
   only transmit impulses + initiate AP if threshold is reached

   → brain not overloaded with sensory info

   (impulses of ↓ frequency don't travel to brain)
3. allow connection of nerve pathways

   → wider range of behaviour
   - in dangerous situations : info from 1 neurone → spread throughout body → reach many relay neurones + effectors

   (axons branch out → form ↑ synapses with many neurones)
   - decision-making in brain : motor neurones have many dendrites → ↑ SA for ↑ synapses

     ↳ neurone can integrate info coming from many body parts
4. involved in memory and learning

   e.g : brain receives info about 2 things at the same time

   → new synapses form

i) # Muscle contraction

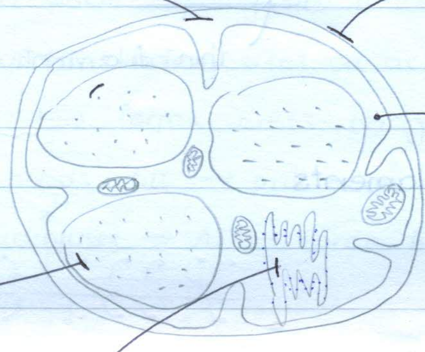
- striated muscle: attached to skeleton
  ↳ are **neurogenic**: contracts when stimulated to do so by impulses that arrive via motor neurones.

Structure of striated muscle

- a muscle contains many muscle fibres
  - muscle fibres = specialized "cell" = syncytium (multinucleate)

transverse section

T-tubules
- deep infoldings into interior of muscle fibre



sarcolemma (cell surface membrane)

sarcoplasm (cytoplasm)
- ↑ # of mitochondria packed between myofibrils
  → ATP; aerobic respiration

myofibril

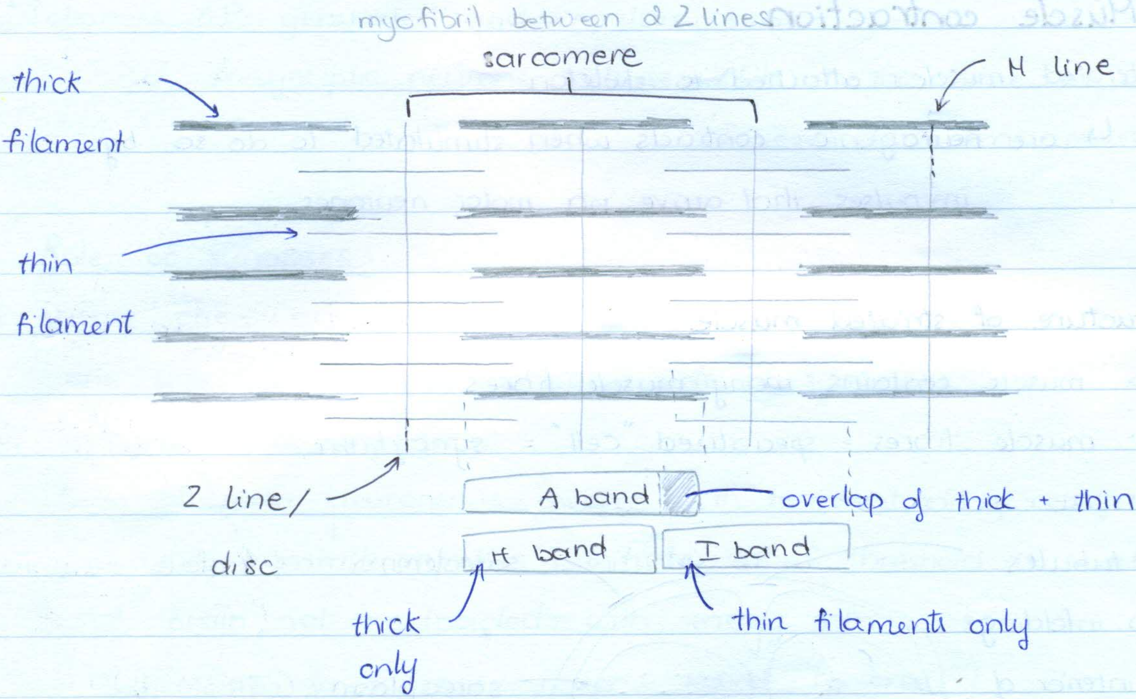
sarcoplasmic reticulum (SR)
- cell surface has ↑ # of protein pumps to transport Ca^{2+} into cisternae of SR

<u>Myofibril</u>

thick filament — myosin

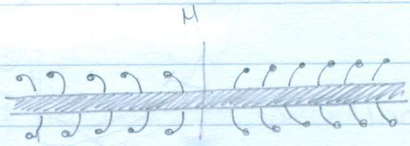
thin filament — actin

Striations: stripes on muscle fibre
- parallel groups of thick + thin filaments
  → myofibrils regular arrangement, striped in the same way
  → muscle fibre

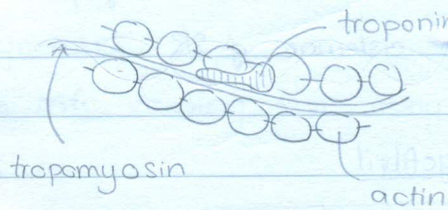


Structure of thick and thin filaments

* Thick filaments:
  - myosin : fibrous protein with globular head
    ↳ point away from M-line.

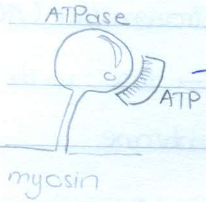


* Thin filaments:
  - actin: globular protein
    ⟶ #↑ ⇒ form chains
    * 2 chains = thin filament



  - tropomyosin (fibrous protein) twisted around 2 chains
  - troponin : attached to actin chain at regular intervals

How muscles contract - The sliding filament model of muscle contraction



- Z discs pull closer ⇒ sarcomeres in myofibrils get shorter

→ energy for movement

(ATP bind to ATPase on myosin head)

① Muscle contracts → Ca^{2+} released from stores in SR
↳ bind to troponin

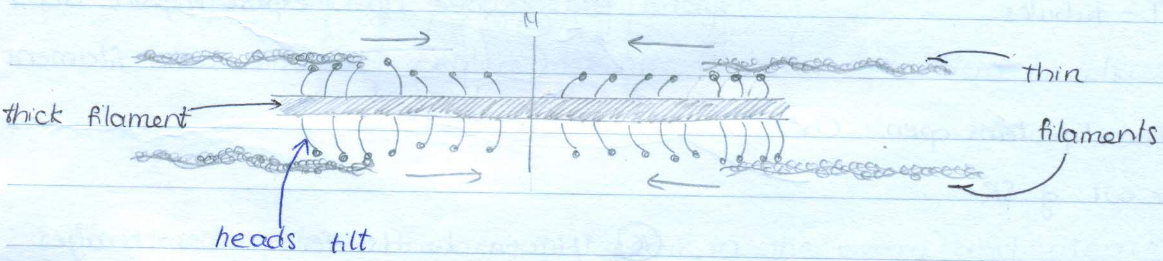
② Troponin molecules change shape

③ Troponin and tropomyosin move to dif. positions on thin filaments
→ expose binding sites on actin chain for myosin
↳ forms cross-bridges between 2 types of filaments

④ Myosin heads tilt → pull actin filaments towards sarcomere centre

⑤ ATP hydrolysis → ATP - force heads to let go of actin

⑥ Heads spring back → repeat process as long as:
- troponin + tropomyosin molecules don't block binding site
- muscle has a supply of ATP

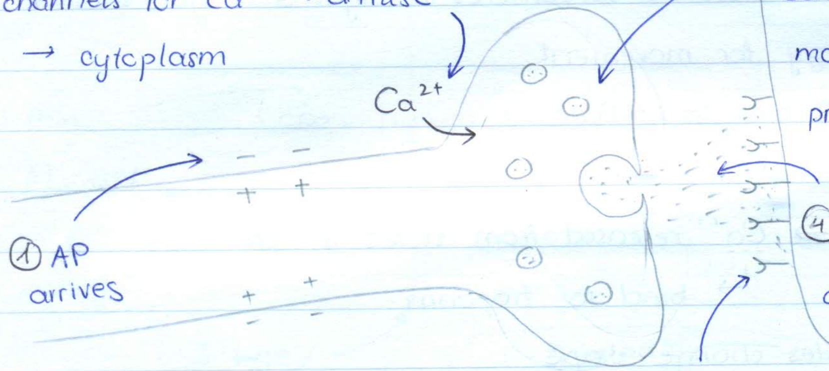


* muscles contract by ≈ 10nm

Stimulating muscles to contract

(2) stimulates opening of voltage-gated channels for Ca^{2+} → diffuse → cytoplasm

(3) Ca^{2+} causes vesicles containing neurotransmitter (ACh) moves towards → fuse with presynaptic membrane



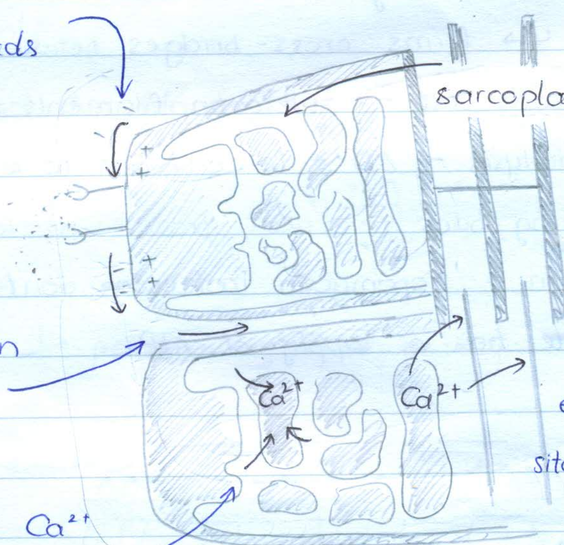
(1) AP arrives

(4) ACh released → diffuse across neuromuscular junction

(5) ACh binds to receptors on sarcolemma → Na^+ channels open

(6) Na^+ diffuse into sarcolemma → depolarize membrane → initiate AP → spreads along membrane

sarcoplasmic reticulum (SR)



(7) depolarisation of sarcolemma spreads down to T - tubules

(9) Ca^{2+} bind to troponin.
Tropomyosin moves → expose myosin - binding sites on actin filament.

(8) Channel proteins open : Ca^{2+} diffuse out of SR

(10) Myosin heads form cross - bridges with thin filaments
→ sarcomere shortens

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Providing ATP for muscle contraction

- aerobic respiration in mitochondria
- lactic fermentation in sarcoplasm
- creatine phosphate
  - stored in sarcoplasm
  - immediate source of energy when ATP in sarcoplasm runs out

  creatine phosphate + ADP ⟶ creatine + ATP
  - demand for energy is slowed / stopped
    ↳ ATP molecules "recharge" creatine

  creatine + ATP ⟶ creatine phosphate + ADP
  - demand for energy is high, but no ATP spare to regenerate
    creatine ⟶ creatinine excreted in urine

e) Hormonal communication

cell-signalling molecules

Endocrine glands make hormones

group of cells that secrete (produce + release) ≥ 1 substances

↳ steroid hormones: lipid soluble =
- pass through phospholipid bilayer
  → bind to receptors molecules inside cytoplasm / nucleus
    ↳ activate processes

Menstrual cycle: changes that occur in the ovary and uterus
≈ 28 days involving ovulation
- ovulation
- menstruation: breakdown + loss of uterus lining

- coordinated by glycoprotein hormones secreted by anterior pituitary gland + ovaries

* uterine cycle and ovarian cycle are synchronized

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① During menstruation (4-8 days), anterior pituitary gland secrete
- follicle stimulating hormone (FSH) } over next few days
- luteinising hormone (LH) } [FSH + LH] ↑

↳ control activity of ovaries (responsible for ovulation)

② In the ovary, one follicle becomes 'dominant' one
- presence of FSH + LH ⟶ stimulate oestrogen secretion from cells surrounding follicle
  ↓ production and [LH + FSH] ↓ ← negative feedback
- oestrogen: stimulate endometrium to
  - grow, thicken
  - develop numerous blood capillaries

③ Surge of LH and a bit ↑ FSH secretion
↳ dominant follicle burst ⟶ shed gamete into oviduct
  ↓
  collapse ⟶ form corpus luteum (yellow body)

- corpus luteum secretes:
  - progesterone } maintain uterus lining
  - some oestrogen } ⟶ ready to receive embryo if fertilisation occurs

↳ inhibits a.pit. gland secreting FSH + LH = no more follicles develop

④ ↳ less stimulation of corpus luteum
  ↳ degenerates

= less oestrogen + progesterone secreted

[ ]↓ = endometrium not maintained ⟶ menstruation begins

↳ release a.p. gland inhibition
  ↳ FSH secreted
    ↳ begin another cycle!

Birth control - preventing pregnancy

1. The birth control pill

↳ contains steroid hormones that suppress ovulation

- synthetic hormones: breaks down more slowly in body → act longer
- type (1) progesterone only ; (2) progesterone + oestrogen

synthetic   combined oral contraceptives

- 21 days pills active; 7 days pills inactive

• oestrogen + progesterone suppress secretion of FSH and LH from anterior pituitary gland (negative feedback)

→ prevents [FSH + LH] reaching levels that would stimulate ovulation

• after 21 days, [O + E] fall

→ uterine lining no longer maintained → menstruation occurs

* progesterone: may allow ovulation to occur BUT contraceptive bc:

- ↓ ability of sperm to fertilise egg
- making mucus in cervix more viscous

→ less easily penetrated by sperm

2. The morning - after pill

* taken after woman has had unprotected sex (up to 72hrs)

• synthetic progesterone - like hormone

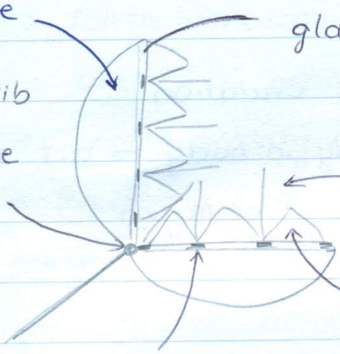
- ↓ chances of sperm reaching + fertilising egg
- prevent pregnancy: stopping embryo implanting into uterus.

15.2. Control and coordination in plants

Venus fly trap

lobe

Midrib = hinge



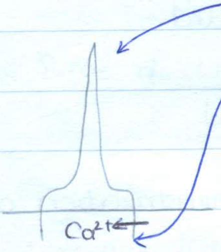
glands secrete digestive enzymes

Insect: N_2 source

each lobe × 3 sensory hairs - responds when deflected

nectar-secreting glands → attract insects

stiff outer-edges (interlock) → trap insects



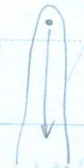
1. Sensory hair is deflected
2. Ca^{2+} channels at base opens. Ca^{2+} flows in
   → generate receptor potential
3. Within 20 - 35s : 2 hairs stimulated / 1 hair stimulated twice

↳ AP spreads across lobe = trap closes

- further stimulation: deflection of hairs
  - ↳ edges of lobes forced to seal
  - ↳ ↑ Ca^{2+} enter cells → exocytosis of vesicles containing digestive enzymes
- trap closes for a week, opens when cells on outer surface ↑ length. ← due to auxin

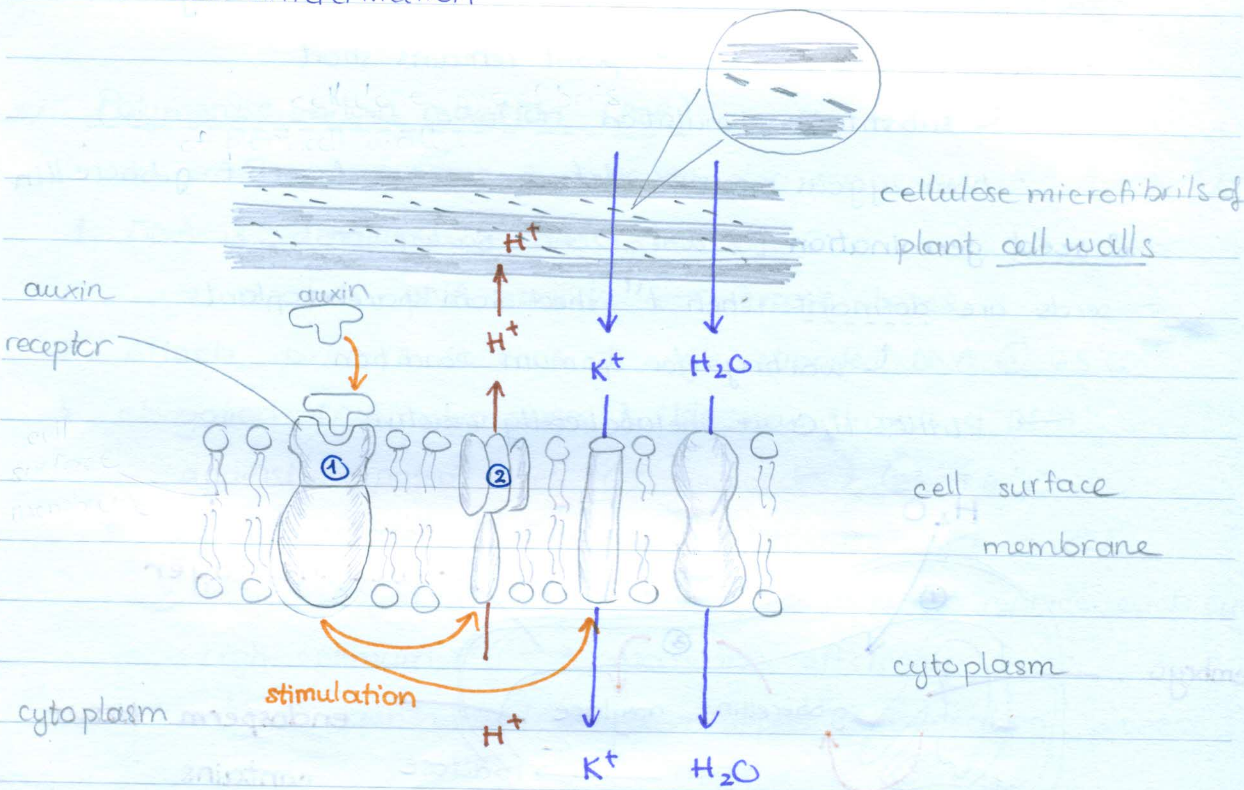
Adaptations to reduce waste of energy

- 1 hair stimulated will not close trap e.g: wind, raindrop
- gap between stiff outer edges allow small insects to escape
  not worth it



Plant hormones - 1. Auxin (main type = IAA)

- synthesized in meristems (tip of shoots + roots; @ cell division occurs)
  - → auxin actively transported away from meristem (cell to cell).
- function : growth
  1. cell division: mitosis
  2. cell elongation — absorbing H_2O, controlled by auxin
  3. cell differentiation



1. Auxin binds to receptor
2. Stimulates ATPase pump H^+ across cell surface membrane (from cytoplasm to cell wall → ↓pH)
3. Expansins (proteins) activated ↳ loosen linkage between cellulose microfibrils
4. disruption occurs briefly → cell expands without losing overall strength

- K^+ in = ↓Ψ ⇒ H_2O in

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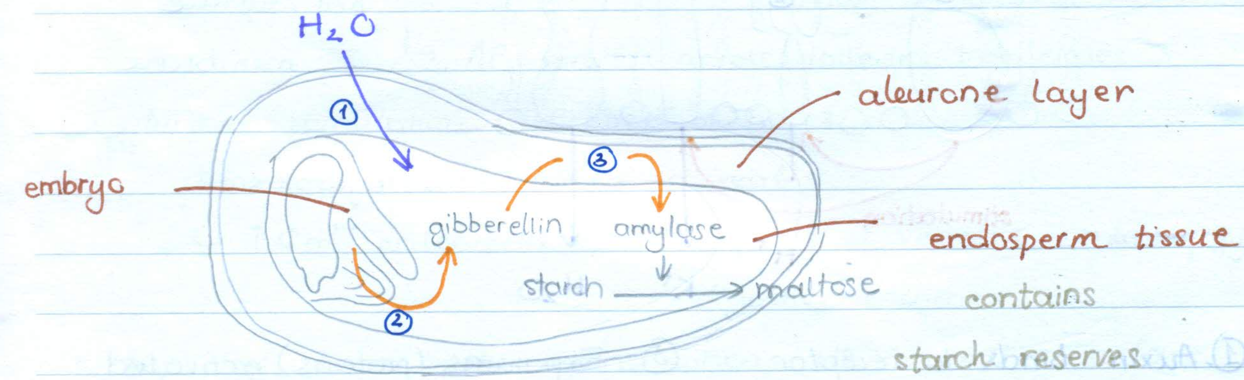
2. **Gibberellins** - growth regulator ∴ / seed germination
controlling stem elongation.

- synthesized in most parts of plants (mainly in young leaves + seeds)
- function: 1. stem elongation
  - dominant allele Le → plant grows tall
    ↳ codes for functional enzyme of active form of gibberellin GA1 - stimulates cell division / elongation

  e.g.: cabbage
  - recessive allele le → plant remains short
    ↳ substitution mutation (alanine → theorine a.a.)
    - homozygous recessive lele : no active form of gibberellin

  2. seed germination of wheat and barley
  - seeds are dormant when 1st shed from parent plant
    ↳ waiting for optimum condition
  → little H_2O + metabolically inactive



1. Absorption of H_2O stimulates germination

2. Embryo synthesizes gibberellin in response to water uptake

3. Aleurone layer synthesizes amylase in response to gibberellin

4. amylase mobilizes energy reserves
   - hydrolyses starch
   ↳ maltose < glucose / glucose

16. Inherited Change

16.1. Passage of information from parent to offspring

a) Homologous chromosomes : in diploid cell
- same structure, gene, loci
- pair together (bivalent) during first division of meiosis

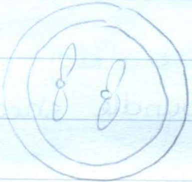
b) haploid cell : possesses 1 complete set of chromosomes : n

diploid cell : " 2 " " " " : 2n

- meiosis (reduction division)
  - \# of chromosomes would double w/o
  - introduces genetic variation → mutation

d. Meiosis

Parent cell



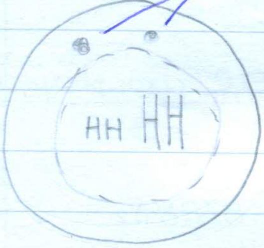
2n = chromosomes

→ interphase × 2 chromosomes

= 4n

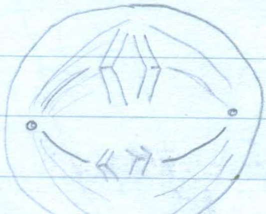
Meiosis I

Prophase I



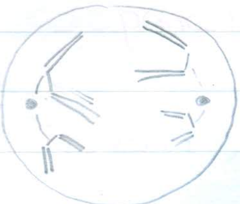
- centrioles divide → opposite poles
  → spindle formation
- chromosomes condense (crossing over)
  ↳ form bivalents (pair up)
- nucleolus disappears
- nuclear envelope breaks down

Metaphase I



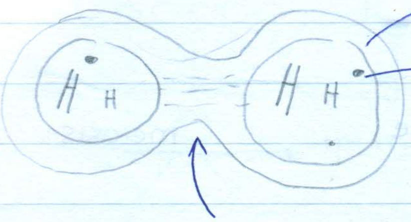
- bivalent form at equator
- spindle attach to centromere

Anaphase I



- whole chromosomes move to opposite end/poles of spindle (pulled by microtubules)

Telophase I



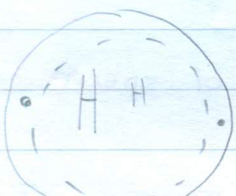
nuclear envelope reforms

nucleus reform

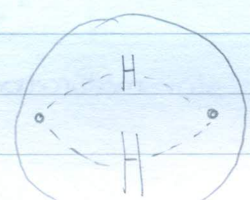
cytokinesis

Meiosis II

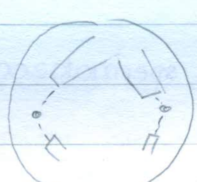
Prophase II



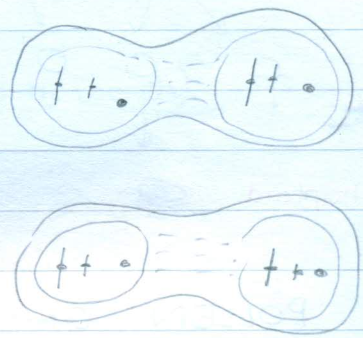
Metaphase II



Anaphase II



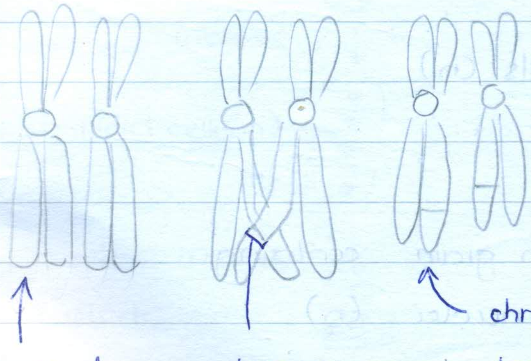
Telophase II



↳ 4 haploid daughter cells

How meiosis causes variation ← crossing over / independent assortment

independent assortment ↓ dif alleles on dif chromosomes end up in any combo in gametes (from random alignment of bivalent on equator in Mei I).



pair of homologous chromosomes

chiasmata: point where crossing occurs

chromosomes have different combination of alleles

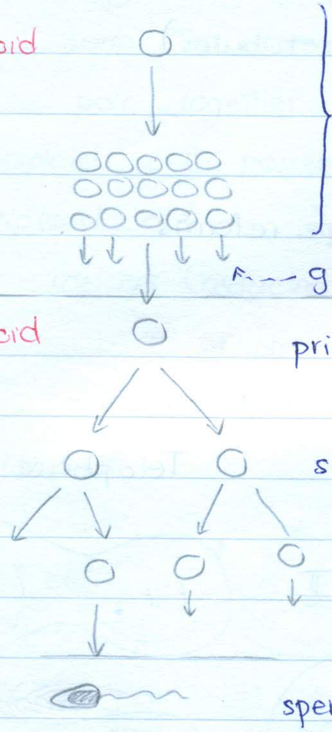
* ≥1 chiasma possible

* chromatid break away

→ reconnect to another chromatid

c.) gametogenesis

SPERMATOGENESIS



diploid — spermatogonia — mitosis

growth

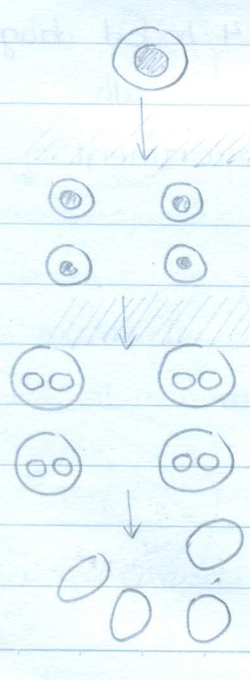
diploid — primary spermatocyte — meiosis

haploid — secondary spermatocyte

spermatid

haploid — spermatozoan — maturation of sperm

POLLEN GRAIN



pollen mother cell (2n)

meiosis

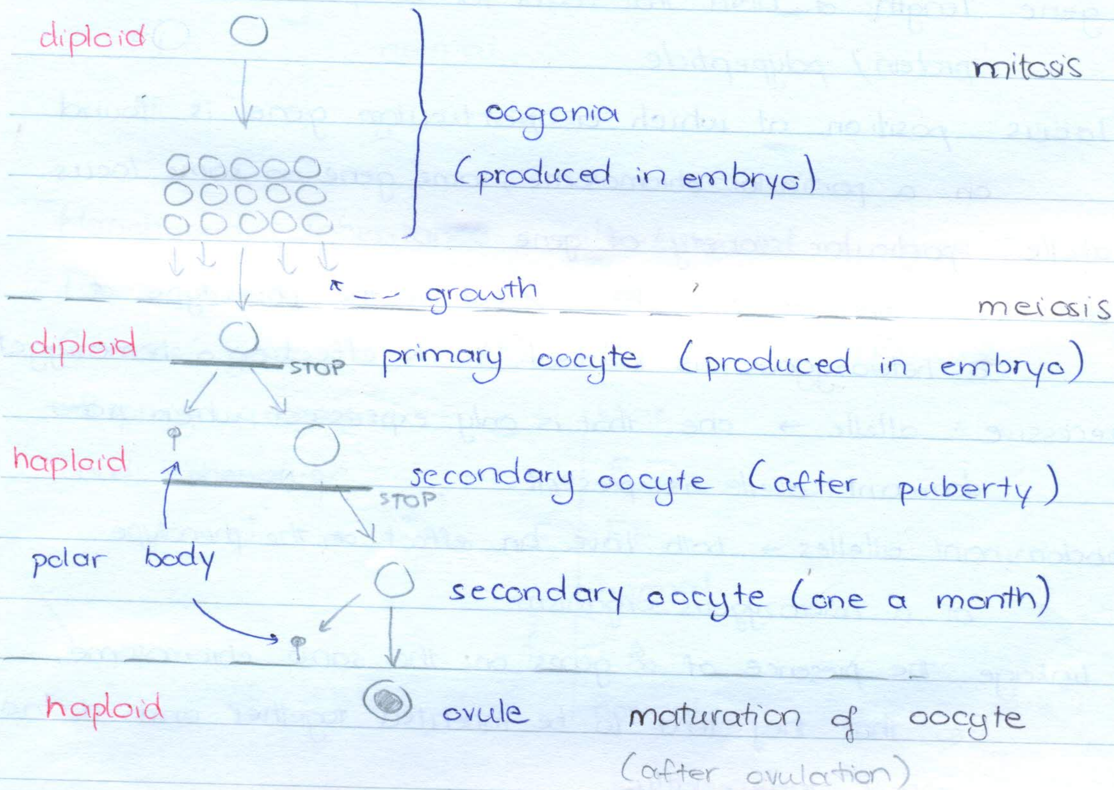
4 haploid cells (n)

mitosis

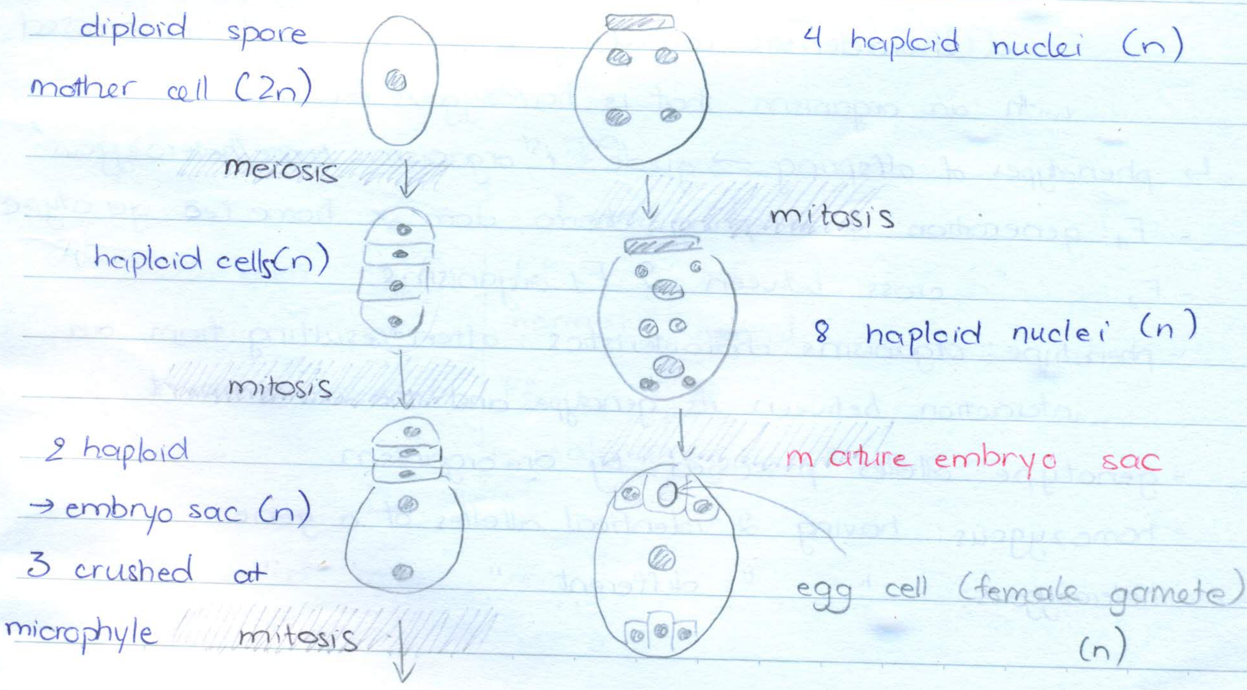
young pollen grain containg 2 haploid nuclei (n)

mature pollen grains (n)

OOGENESIS



Development of embryo sac



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16.2. The roles of genes in determining phenotype

a) - gene: length of DNA that codes for a particular protein/ polypeptide.
- locus: position at which a particular gene is found on a particular chromosome; same gene on same locus
- allele: particular variety of gene
- dominant: allelle → one whose effect on the phenotype of a heterozygous is identical to its effect on a homozygote
- recessive: allelle → one that is only expressed when no dominant allelle is present
- codominant: allelles → both have an effect on the phenotype of a heterozygous organism.
- linkage: the presence of 2 genes on the same chromosome, so that they tend to be inherited together and do not assort independtly.
- test cross: a genetic cross in which an organism showing a characteristic caused by a dominant allelle is crossed with an organism that is homozygous recessive;

↳ phenotypes of offspring → guide: 1st organism homo/heterozygous?
- F_1: generation of offspring: homo-dom x homo rec genotype
- F_2: " cross between 2 F_1 organisms
- phenotype: organisms' characteristics; often resulting from an interaction between its genotype and the environment
- genotype: allelles possessed by an organism.
- homozygous: having 2 identical allelles of a gene
- heterozygous: " " different " " " "

b)

Genotype	Phenotype
FF	normal
Ff	normal
ff	cystic fibrosis

Monohybrid Inheritance : inheritance of 1 gene

Parent genotypes Ff × FF

Gametes' genotype F f F

Offspring genotype and phenotype

		egg F
sperm	F	FF normal
	f	Ff normal

Parent genotype : Ff Ff

F f F f

		egg F	f
sperm	F	FF normal	Ff normal
	f	Ff normal	ff cystic fibrosis

OR : expect ratio : normal : cf

3 : 1

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Codominance

Genotype	Phenotype
C^R C^R	red coat
C^R C^W	pink coat
C^W C^W	white coat

↳ Multiple allele

Blood type: I^o, I^A, I^B

Parent phenotype: blood group A × blood group B

$I^A I^o$ $I^B I^o$

I^A or I^o I^B or I^o

	I^A	I^o
I^B	$I^A I^B$ b.g. AB	$I^B I^o$ b.g. B
I^o	$I^A I^o$ b.g. A	$I^o I^o$ b.g. O

Sex linkage: genes present on X chromosome, not on Y

→ sex-linked gene e.g.: color blindness

Parent phenotype: ♀ normal vision × ♂ normal vision

genotype: X^A X^a X^A Y

X^A or X^a X^A or Y

	X^A	X^a
X^A	$X^A X^A$ ♀ norm	$X^A X^a$ ♀ norm
Y	$X^A Y$ ♂ norm	$X^a Y$ ♂ color blind

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Dihybrid inheritance : inheritance of 2 genes

Genotype	Phenotype
A	brown hair ⟵ allelle for
a	black hair
L	long legs
l	short legs

Parents' phenotypes : brown, long × brown, long

AaLl AaLl

AL or Al or aL or al   AL or Al or aL or al

	AL	Al	aL	al
AL	AALL brown, long	AALl brown, long	AaLL brown, long	AaLl brown, long
Al	AALl brown, long	AAll brown, short	AaLl brown, long	Aall brown, short
aL	AaLL brown, long	AaLl brown, long	aaLL black, long	aaLl black, long
al	AaLl brown, long	Aall brown, short	aaLl black, long	aall black short

- ratio 9 : 3 : 3 : 1 of dihybrid cross between 2 heterozygotes, - allelles of both genes show dominance
  - genes on different chromosomes

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Interactions between loci : different loci interact to affect 1 phenotypic character

e.g. : $\underset{\substack{\uparrow \\ \text{loci 1}}}{\underline{I / i}}$; $\underset{\substack{\uparrow \\ \text{loci 2}}}{\underline{C / c}}$

Autosomal linkage : ≥ 2 gene loci on same chromosome

→ ∅ assort independently in meiosis

e.g. : genotype : (EA) (EA)

↳ not on same chromosome

Parental phenotypes : ♂ _ _ _ _ ♀ _ _ _ _

(EA) (ea) (ea) (ea)

EA or ea ea

		♀ ea
♂	EA	(EA) (ea)
	ea	(ea) (ea)

e) Mutation : unpredictable change in genetic material of an organism

- Δ structure of DNA molecule → dif allelle of gene = gene mutation

sources :

- random
- environmental factors (mutagen) ← ionising radiation : α, β, γ
  - UV "
  - chemicals

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Gene mutations:

Type :   Effect

- base substitution   silent : same amino acid, no effect
- base addition   missense : dif " ", effect on organism
- base deletion   nonsense : stop codon
                                          frame shift → protein made is useless

↳ x structure + function

Human conditions:

1. Sickle cell anaemia → base sub. on β-globin polypeptide

allelle Hb^A allelle Hb^S

CTT → CAT

glutamic acid → valine

↳ Hb more soluble   Hb less soluble

- Effect: Hb molecules stick together → form long fibres in RBC
  → RBC pulled out of shape (half moon / sickle)
  → distorted, can't transport O_2
  - stuck in small capillaries, block other RBC

↳ ∧ can die from lack of O_2

2. Albinism → mutation in gene for tyrosinase
   ↳ absence of tyrosinase
   ↳ presence of inactive tyrosinase

tyrosine $\xrightarrow{\text{tyrosinase}}$ DOPA $\xrightarrow{\text{tyrosinase}}$ dopaquinone → melanin

Effect: - dark pigment melanin partially / totally missing from eyes, skin, hair
- poor vision; rapid, jerky eye movement; avoid bright light
- classic form : homozygous autosomal recessive
  other form : sex-linked

3. Huntington's disease : dominant allele

normal: sufferer:

CAG CAG CAG   CAG CAG CAG CAG CAG ... = "stutter"

gene code for 'huntingtin'

∆. Effect: a neurological disorder

↳ - involuntary movements (chorea)

- progressive mental deterioration

- age of onset: usually at middle-age but varies.

4. Haemophilia : sex-linked; on X chromosome

- gene code for production of protein for blood-clotting

↳ "factor VIII"

H: dominant allele

h: recessive → blood fail to clot properly

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16.3. Gene control ↓ in prokaryotes

a) genes:
- structural: code for proteins required by cell
- regulatory: " " " that regulate expression of other genes

enzymes:
- repressible: synthesis of enzyme can be prevented by binding repressor protein to a specific site (operator) on bacterium's DNA
- inducible: - requires substrate → inducer

inducer interact w/ regulatory enzyme → transcription occurs

The lac operon

- cluster of 3 structural genes + a length of DNA
  - lacZ : code for β-galactosidase : hydrolyse lactose → glucose, galactose
  - lacY : " " permease
  - lacA : " " transacetylase

NO LACTOSE	LACTOSE PRESENT
• regulatory gene codes for repressor protein*	• lactose taken up by bacterium
• ↳ bind to operator region close to β-galactosidase gene	• ↳ bind to repressor protein → distort DNA binding site → can't bind to operator site
• RNA cannot bind to promoter region of DNA	• transcription no longer inhibited
• → no transcription of 3 structural gene	• ! avoid waste of energy and materials

* protein is allosteric: 2 binding sites

Gene control ↓ in eukaryotes

Transcription factors in gene expression
protein

↳ bind to promoter region of gene

→ make sure gene expressed in correct cell, , extent

Effects:

- form part of protein complex that binds to promoter region of gene
- activate appropriate genes in sequence
- determinate of sex in mammals
- allow responses to environmental stimuli
- regulate cell cycle, growth, apoptosis
  product of proto-oncogenes, tumour suppressor genes
- hormones have their effect through transcription factors

GIBBERELLIN : controls seed germination by stimulating amylase synthesis

- DELLA proteins inhibit binding of transcription factor PIF to a gene promoter

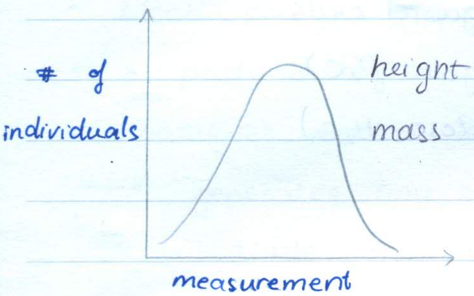
→ gibberellin causes the breakdown of DELLA

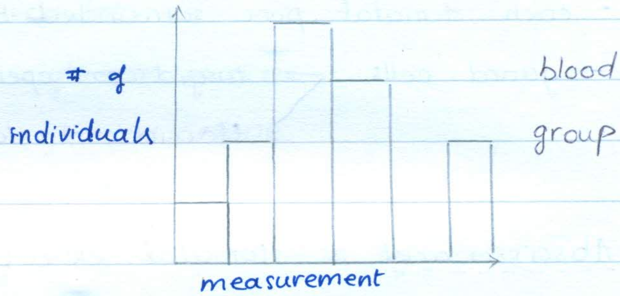
→ PIF can bind to target promoter

→ ↑ amylase production

17. Selection & Variation

17.1. Variation





continuous

- quantitative differences
- genetic basis:
  + dif allelles @ 1 gene locus

  → small effect on phenotype
  + dif genes have same/additive effect on phenotype
- + large # of genes (polygenes)

  → combined effect on a phenotypic trait

discontinuous

- qualitative differences
- genetic basis:
  + dif allelles @ 1 gene locus

  → large effect on phenotype
  + dif genes have quite a dif effect on phenotype

Environmental affect on phenotype

e.g.: hair color in Siamese cats, Himalayan

- dark extremities: tips to ears, nose, paws, tail

  ↳ allelle allows formation of dark pigment only @ ↓t°

e.g: in plants

- different growth

  ↑ lower light intensity, fewer nutrients

- use t-test to compare variation of 2 dif populations
- importance of genetic variation in selection
  basis for natural and artificial selection to act upon

Causes of genetic variation
- independent assortment of chromosomes during meiosis
- crossing over between chromatids of homologous chromosomes during meiosis
- random mating
- random fertilisation of gametes
- mutation

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17.2. Natural and artificial selection

a) Natural selection

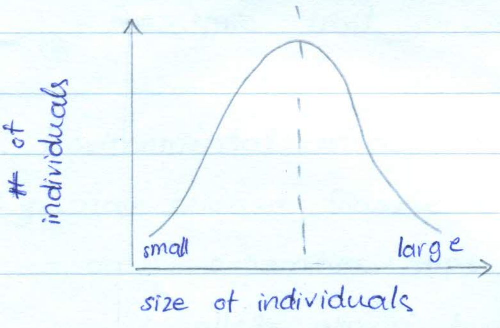
- occurs as populations have capacity to produce many offsprings ⟶ compete for resources
⟶ individuals best adapted survive
↳ breed, pass on alleles

Environmental factors

- biotic : caused by other living organisms
  - predation, food competition, infection by pathogen
- abiotic : caused by non-living components of environment
  - water supply, nutrient level of soil

↳ selection pressures : controls chances of alleles being passed on
↳ effects allele frequency in population = natural selection

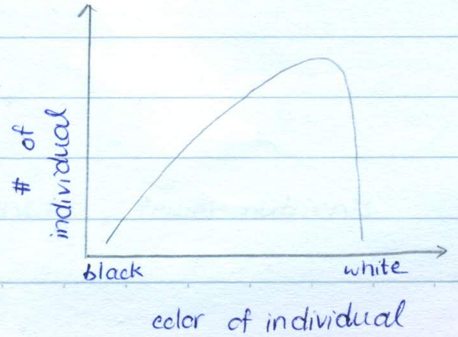
Stabilising selection



- acts against extremes
- favours the environment
- e.g.: birth weight

Directional selection

- favours variants of 1 extreme
when new allele appears
- " environmental factor
- e.g.: peppered moths



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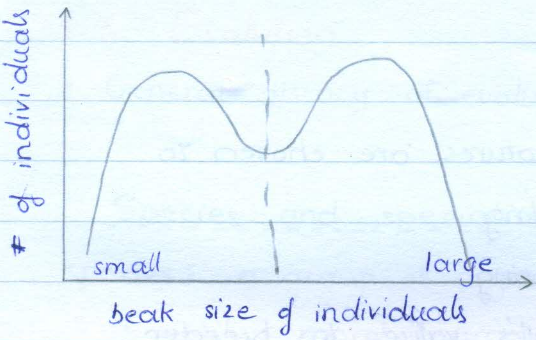
Disruptive selection



- conditions favour both extremes
→ maintains dif phenotypes in population
- e.g :: Galapagos finches

Genetic drift: - Δ gene pool of a small population
Δ in allele frequency
- due to chance [random] bc only some organisms of each generation reproduce

Founder effect: - genetic drift resulting from the colonization of a new location by small # of individuals
recently isolated
→ random Δ gene pool → affect allele frequency

Hardy - Weinberg prinicple
↳ calculate allelle frequencies and proportions of genotypes of a particular gene in a population

$$p + q = 1$$

dominant allelle frequency

recessive allele frequency

$$p^2 + 2pq + q^2 = 1$$

does not apply when: • small population
• significant selective pressure against 1 genotype
• migration into/out of a population - individuals carry 1 of 2 alleles
• non-random mating

e) Artificial selection

- selective pressure : humans
- individuals with desirable features are chosen to interbreed = selective breeding
  → alleles passed on to offspring

e.g : progeny testing : measure bull's value to breeder
- performance of milk production of the bull's offspring
  ↳ used to assess bull.

Crop improvement by selective breeding
- introduction of disease resistance to varieties of wheat + rice
- incorporate mutant alleles for gibberellin synthesis into dwarf varieties
  → ↑ proportion of energy put into grain
  ↳ ↑ yield
- inbreeding + hybridisation
  → produce vigorous, uniform varieties of maize

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17.3. Evolution

- General theory of evolution: organisms have changed over time

Species and speciation

species: a group of organisms with:
- similar morphological, physiological, biochemical and behavioural features
- can interbreed to produce fertile offspring
- reproductively isolated from other species

speciation: production of new species

1. Allopatric speciation - geographical isolation
   - population of species split → move to dif areas
   - dif. selective pressures
     ↳ features change over time
       ↳ re-introduce species → no longer able interbreed
         ↳ new species formed

2. Sympatric speciation - ecological & behavioural separation
   - polyploidy : organism ≥ 2 sets of chromosome (complete)
     ↑ meiosis goes wrong when forming gametes
   - tetraploid → sterile : 4 chromosomes try to pair up during Meiosis I
     ↳ muddle up
     ↳ grow → reproduce asexually by mitosis; usually plants
   - triploid → sterile : ∅ share evenly 3 sets of chromosomes

↳ original diploid plant + tetraploid : cannot interbreed
→ dif. species

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- autopolyploid : all sets of chromosomes from same species
- allopolyploid : dif sets of chromosomes from dif but closely related species

meiosis easier in allop. than autop.
chromosomes not identical → pair up dif species together

→ cannot interbreed with parent species
→ dif. species

Reproductive isolation : inability of 2 groups of organisms of same species to interbreed successfully
e.g : due to geographical separation / behavioural differences

1. Prezygotic isolation :
- individuals not recognizing each other as potential mates
- animals physically unable to mate
- incompatibility of pollen and stigma
- inability of ♀ and ♂ gamete fusion

2. Post zygotic isolation :
- failure of cell division in zygote
- non-viable offspring
- viable , but sterile offspring

* waste of energy and resources

Molecular comparison between species

1. comparing amino acid sequences of proteins
   - # of differences in sequence
     ↳ measure how closely related the species are

2. comparing nucleotide sequences of mitochondrial DNA

Human mtDNA – inherited through female line
↓
circular → can't undergo crossing over
↳ Δ nucleotide sequence arise by mutation

- dif. human populations = dif mtDNA sequence

→ 'molecular clock' hypothesis
↳ - constant rate of mutation over time
- ↑ dif in sequence = common ancestor longer ago
↓
estimated from fossil evidence

Extinction

- climate change:
- ↑ competition from better adapted species

Human causes:

- loss of habitat: draining wetlands, cutting down rainforests, polluting air, water, soil
- killing: for sport or for food

Mass extinctions:

- sudden Δ in environment: large asteroid colliding to Earth

11.2. Antibodies and vaccination (AS)

- Hybridoma method
  - mouse is injected w̄ antigen
  - wait for immune response to occur
  - clonal selection
  - clonal expansion
  - β-lymphocytes / plasma cells extracted from mouse spleen
  - plasma cell fuse w̄ cancer cell
    → form hybridoma cells
  - hybridoma cells producing antibodies are identified
  → culture on a large scale to secrete monoclonal antibodies

18. Biodiversity, classification, conservation

18.1. Biodiversity

a) • species: - a group of organisms with similar morphology and physiology,
- can breed together to produce fertile offspring
- are reproductively isolated from other species

• ecosystem: a relatively self-contained, interacting community of organisms, and the environment in which they live / with which they interact

• niche: role of an organism in an ecosystem

b) **Biodiversity**: degree of variation of life forms in an ecosystem

3 levels:
- • variation in ecosystems / habitats
- • # of species and their relative abundance
- • genetic variation within each species

- species richness: # of species in a community
- species diversity: species richness + measure eveness of abundance
  ↳ higher = ecosystem more stable (more resistant to changes)
- genetic diversity: diversity of alleles within the genes in the genome of a single species

obtained by
- proportion of genes with dif alleles
- # of alleles per gene

• genetic differences
- between populations
- within each population

Sampling

↳ estimate Σ # in an area

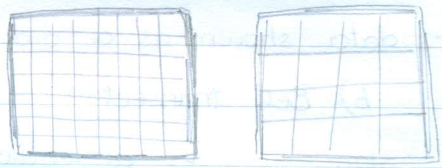
Assessing species diversity

1. Random sampling

- area looks reasonably uniform
- no clear pattern to the way species are distributed

↳ avoid bias

a) **Quadrats** - square frame that marks off an area of ground / water



↳ identify species present

measure abundance

calculate:

- species frequency: measure of chance of a particular species being found within any one quadrat.
- species density: measure # of individuals per unit area

estimate:
- percentage cover: ≈ % area in quadrat occupied by each species
- abundance scale (e.g.: Braun - Blanquet scale): # + plant cover

b) **Mark - release - recapture**: estimating # of mobile animals

1. As many individuals caught as possible
2. Individuals marked (in way that won't affect its future chances of survival)
3. Marked individuals → counted (a)
4. ↳ returned to their habitat → mix randomly w̄ population

TIME ELAPSE

5. Large sample captured
6. $\underbrace{\text{\# of } \overset{(b)}{\text{marked}} \text{ and unmarked}}_{(c)}$ individuals counted

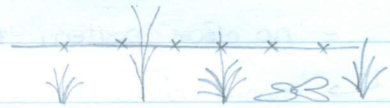
$$\text{estimated \# of population} = \frac{a \times c}{b}$$

2. Systematic sampling

- investigate species distribution where physical conditions change

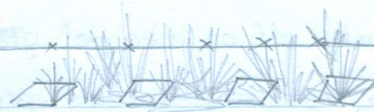
a) Line transect

• record identity of organisms that touch line at set distances



• data shown as a drawing

b) Belt transect

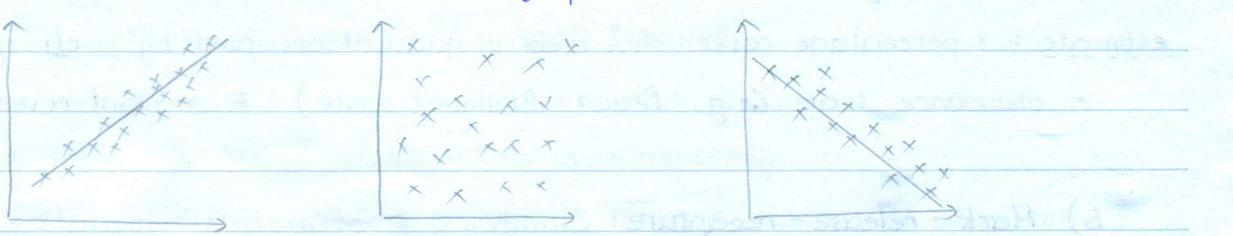


• place quadrats at regular intervals along line

→ record abundance of species within quadrat

• data plotted as bar chart or kite diagram

e) **Correlation** · plot scatter graph or calculate correlation coefficient (r)



positive linear correlation $r = 1$

no correlation $r = 0$

negative linear correlation $r = -1$

* strength = how close the points are to the straight line

Calculating correlation coefficient (r)

1. **Pearson's correlation coefficient** used when data:
   - may be a linear correlation (draw scatter graph first)
   - quantitative data collected as measurements / counts
   - must be normally distributed

$$r = \frac{\sum xy - n\bar{x}\bar{y}}{n s_x s_y}$$

2. Spearman's rank correlation

- data is correlated, but not linear (seen from graph)

→ • rank data for each variable

↳ assess difference between ranks

- make a null hypothesis

$$r_s = 1 - \left(\frac{6 \times \sum D^2}{n^3 - n} \right) \qquad -1 \leq r \leq +1$$

f) Simpson's Index of Diversity

- when you have collected info about abundance of species in an area

↳ use results → calculate value for species diversity in that area

$$D = 1 - \left(\sum \left(\frac{n}{N} \right)^2 \right)$$

18.2. Classification

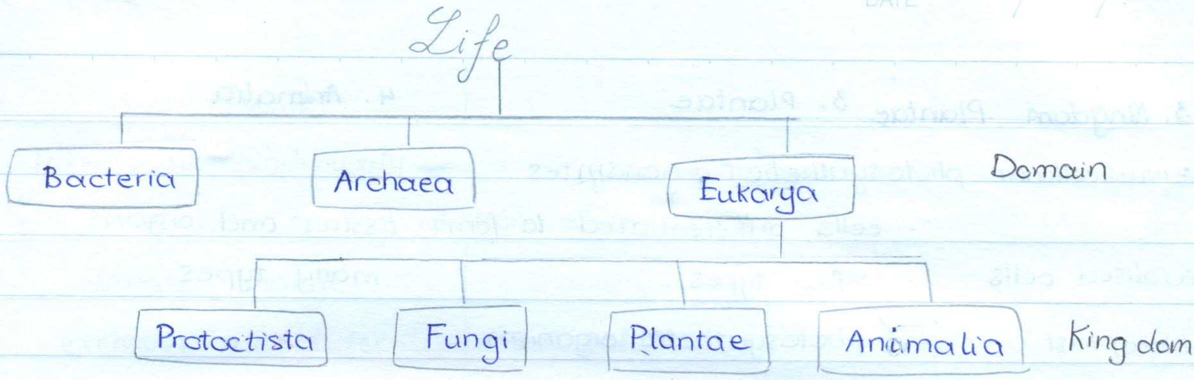
a) ↳ arranging different kinds of organisms into groups

- **Taxonomy**: the study and practice of classification → placing organisms in a series of taxonomic units (taxa)

Domain
Kingdom
Phylum
Class
Order
Family
Genus
Species

b)

	Bacteria	Archaea	Eukarya
	prokaryotic	prokaryotic	eukaryotic
nucleus	✗	✗	✓
membrane-bound organelles	✗	✗	✓
DNA	• circular "chromosome" • no histone proteins associated • smaller circular DNA molecules: plasmid		• in nucleus • arranged as linear chromosome • histone proteins ✓ • mitochondria + chloroplast ↳ circular DNA
ribosomes	(70S) < euka	(70S) < euka similar features	(80S) in cytosol > prokaryotes
cell wall	✓ peptidoglycans	✓, no peptidoglycans	
cell division reproduction	binary fission		mitosis sexually / asexually
unique		extremophiles - inhabit extreme environments	• great diversity of forms: - unicellular - colonial - multicellular organisms



1. **Kingdom Protoctista** (eukaryotic)
   - mostly single-celled or groups of similar cells
   - some are protozoa: have animal-like cells (no cell walls)
   - some are algae: have plant-like cells (cellulose c.w. + chloroplasts)

2. **Kingdom Fungi** (eukaryotic)
   - no chlorophyll → does not photosynthesise
   - heterotrophic nutrition: use organic compounds (C) made by other organisms as source of energy and molecules for metabolism
     from ← dead and decaying matter
     feeding as parasites on living organisms
   - reproduce by spores
   - simple body form: unicellular
     long threads of hyphae (w̄ or w̄/o cross walls)
   - cell walls of chitin (not cellulose)
   - no cilia or flagella

Kingdom	3. Plantae	4. Animalia
	• muticellular eukaryotes • cells differentiated to form tissues and organs	
specialised cells	few types	many types
chloroplast	✓ photosynthetic organism	✗
vacuole	• large, permanent • for support	• small, temporary
nutrition	autotrophic	heterotrophic
	cells sometimes contain flagella	cells sometimes contain cilia + flagella
unique	• complex bodies • highly branched: above + below ground	• communication by nervous system

d) Viruses → none of features traditionally used for classification

↳ acellular organisms (cellular structure ≠ bacteria + fungi)

- particles made of proteins + nucleic acid
- in free environment, viruses are infectious, but have no metabolism
- when infect cells → use biochemical machinery of host cell to copy their nucleic acid + make proteins
  - ↳ destruction of host cell
- energy provided by respiration in host cell

⟹ Taxonomic system: classified by:

- which disease the virus causes
- typee of nucleic acid (RNA or DNA)
- whether nucleic acid is single or double stranded

CH_4: cattle, rice farming, breakdown organic waste anaerobically

18.3. Conservation

Threats to biodiversity:

- habitat loss + degradation of environment
- climate change
- excessive use of fertilisers → pollution
- overexploitation + unsustainable use of resources
- alien species invasion on native species

1. Habitat loss

↑ destruction of natural environment

(land clearing for agriculture, housing, transport, ...)

⇒ habitat fragmentation (habitats become divided)

- most at risk: of extinction: endemic species on small islands

Deforestation : remove vegetation

⇒ soil erosion → severe land degradation

2. Climate change

- Air pollution : combustion of fuel with high sulfur content

  → SO_2 in atm + H_2O = acid rain

  ↳ destroy vegetation + acidification of aquatic ecosystems

  animals can't breed / survive in waters of low pH

- Industrialisation + extraction and combustion of fossil fuels

  → ↑ [CO_2 + CH_4] = greenhouse gases → climate change

  → global warming

  ↳ • Δ distribution of terrestrial ecosystems

  - acidification of oceans : destroy $CaCO_3$ mollusc shells
  - coral bleaching when t° too high (algae leaves coral)

  ↑ protection of coastlines

- rise in sea levels
- ↑ frequency of natural catastrophes (hurricane, flooding, ...)

e.g.: flooding ↑[nutrients] in coastal waters

⇒ ↑ growth of phytoplankton + food for starfish larvae → eat coral

3. **Fertilisers and pollution**

* polutant: substances animal bodies unable to metabolise/excrete

PCB
- factory wastes flow into rivers → substance persists → enter food chains

⇒ weakens immune system ; ↓ fertility

- marine pollutant: non-biodegradable plastic

e.g.: - animals (dolphins) get caught in discarded fishing nets → die

- turtles eat plastic bags (mistaken for jellyfish)

- excess fertilisers (not absorbed by crop plants)

↳ drain into river → extra nutrients → growth of producers

like algae: - produce toxic substances

\ algae growth unbalances food web

4. **Overexploitation** of resources

- overfishing : taking wild fish from their environment

↑ near extinction

- removal of valuable trees by logging companies e.g.: mahogany

→ loss of keystone species (have central role in ecosystem)

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The need to maintain biodiversity

- Ecological reasons:
  - ↑ diversity $\propto$ less likely be unbalanced by Δ condition / threats
  - ecosystems are direct value to humans
    - e.g.: + antibiotics from fungi, bacteria
      + anti-cancer drugs isolated from plants
- Aesthetic reasons:
  - ∧ gain pleasure from studying / appreciating natural world (inspiration for artists, poets, photographers, ...)
  - ecotourism: wildlife = source of income
    ↳ provides employment; contribute to economies
- Social and commercial reasons
  - wild plant species: resistant to large # of bacterial strains
    → cultivated w̄ crop plants (↓ genetic diversity, lost by selective breeding)
    → ↑ resistance
  - microorganisms = source of useful products
    e.g: Taq polymerase ⇒ used for PCR ⇒ forensic / DNA analysis
- Other services
  - forests absorb CO_2 ⇒ ↓ effect of ↑[CO_2] in atmosphere
  - organic waste added in water ∈ broken down by microorganisms
  - transpiration of plants: contribute to water cycle
    ↳ provide water for drinking + irrigation
  - termites, ants, fungi, bacteria recycle elements (C, H, O, N, ...)

Protecting endangered species

* endangered species: threatened with extinction

National parks

= conservation areas with strict limits to protect wildlife + environment

- alien animal species removed; invasive plants dug up and destroyed
- restriction on human activities
  - tourism: raise money + awareness
- marine parks: conserve fragile ecosystems and areas at risk of overfishing, dredging, and pollution

Zoos

= protection for endangered and vulnerable species

= provide enjoyment and interest for visitors

- captive breeding programmes to reintroduce animals to natural habitats
  - problem: inbreeding (breeding animals from small pop)
    - some captive bred animals don't know how to avoid predators, find food, rear young
    - animal refuse to breed in captivity; hard to recreate suitable habitat → animal can't be returned to the wild
- for research: understand breeding habits, habitat, how to ↑ genetic diversity

Assisted reproduction: solution to inbreeding problems

a) Sperm bank: freezing collected semen

- sperm samples collected → checked for sperm activity → diluted w̄ medium solution (buffer + albumen) → put into thin tubes (straws) → stored in liquid nitrogen @ −196 °C

b) Artificial insemination (AI)

- a straw placed in warm water → activate sperm
- placed into catheter → inserted into vagina, through cervix, into uterus

(after ♀ hormone treatment : ♀ superovulates)

- resulting embryos 'flushed out' of uterus

→ transferred to other females that had hormonal treatment

surrogate mothers to prepare for pregnancy

= embryo transfer
  - protects endangered animal from pregnancy
  - ♀ = source of many offspring

c) In vitro fertilisation (IVF)

- oocyte collected by inserting needle into ovaries
  → withdraw some mature follicles
- oocyte cultured in a medium → mix $\bar{w}$ semen
- resulting zygotes divides → form embryos → cultured many days
  → placed into mother / other females (same or dif species)

d) Frozen zoo

- holds genetic resources in form of sperm / egg* / embryo for endangered species
  + more genetic diversity
  + genetic material kept for longer periods of time

* eggs: - more difficult to freeze
- more likely damaged by freezing + thawing

Problems of successful conservation

of ecosystem

- organism from : extinction → to : ↑ # beyond sustainable capacity
- culling (aim: ↓ #)
  - transferring animals to places w̄ small populations
  - birth control : chemical contraceptives : vaccine that targets the region surrounding layer of glycoprotein around egg

(vaccine produces immune response → produce antibodies against glycoprotein)

+ antibodies attach to glycoprotein
  → block sperms from fertilising
+ 90% success rate

Botanic gardens

* seeds / cuttings collected from species in the wild
  → build up population → reintroduce to natural habitat
- sample of cells grown of agar (in sterile conditions)
  ↳ mitosis ⇒ mass of cells → transferred to medium containing plant hormones
  ↳ grow stems/ roots → transferred to soil.
- roles of botanic garden:
  - protect endangered plant species
  - research methods of reproduction + growth
  - research conservation methods
  - reintroduce species to habitats
  - educate the public — roles of plants in ecosystem, economic value

↳ Seed banks:

- seeds of same species collected from different sites
  → stored sample contain good proportion of total gene pool
  → not lose genetic diversity
- "recalcitrant seeds" cannot be dried and frozen
  ↳ e.g.: seeds of economically important tropical species
  ↳ collect seeds, grow successive generations
  keep as tissue culture

* seeds can be stored for a long time w/ little maintenance, anywhere in the world
* germinated every few years to:
  - check if seeds are still viable
  - produce new plants → collect new seeds
  - find conditions for breaking seed dormancy

Controlling alien species
↳ those moved from one ecosystem to another where they were previously unknown

causes:
- humans trading animals and plants
- introduced as biological control agents to control pests
- escapees
- animals introduced for sport

effects
- successful predator
- compete effectively w̄ native organisms of same niche → extinction
- introduce diseases → spread to organisms that have been exposed to that pathogen

e.g.: water hyacinth
- grow successfully → cover huge areas of land / water
  → block sunlight from reaching native aquatic plants
  → ↓ $[O_2]$ in water → kills fish
- habitat for mosquito larvae

Japanese knotweed
- vigorous root systems → force its way through concrete and damaged buildings, roads, walls
- outcompetes native species by reducing space where they grow

NGOs in local and global conservation

1. CITES - Conservation on International Trade in Endangered Species of Wild Fauna and Flora

- a signed agreement to control trade of e.s. and their products: furs, skin, ivory
- considers evidence presented to it about e.s
  → assigns to 1 of 3 appendices
    → given criteria & trading regulations

- sometimes CITES listings don't benefit the species:
  - species trade becomes illegal
    → price for products ↑ ⇒ ↑ trade

2. WWF - World Wide Fund for Nature
   ↳ campaining group for wildlife : #1
   - "to stop degradation of the planet's natural environments
   - build future where humans + nature live in harmony"
- funds conservation projects
- publicises environmental issues + campaigns

Restoring degraded habitats so they may support a flourishing community with high biodiversity

- degradation: human activity or natural catastrophe
- restoration:
  - e.g: small scale farmer plant trees on land that is no longer needed for food production

  - e.g: replanting mangrove forests
    - provide protection against storm damage, flooding, rising sea levels
    - important nursery grounds for young fish

planting trees in Haiti

- after deforestation, soil erosion, ...

Eden project, UK

- reclamation project
- educated people in plant biodiversity and the need for conservation

19. Genetic technology

19.1. Principles of genetic technology

a) • recombinant DNA (rDNA) - DNA made by joining pieces from ≥ 2 different sources

b) • genetic engineering :
- extraction / synthesis of genes from 1 organism
- place gene into another organism (same / dif species)

→ gene is expressed in new host

c) Polymerase chain reaction (PCR)

(rapid production of large # of copies of a particular DNA fragment)

1. DNA is denatured @ 95°C heating
   → separate DNA strands → expose bases
2. attach primers to ends of single-stranded DNA @ 65°C
3. elongation: DNA polymerase builds new strands of DNA against exposed ones (+ nucleotides) @ 72 °C

* Taq polymerase: 1st heat-stable polymerase used in PCR
- not destroyed in denaturation ∴ no need to replace each cycle
- high optimum t° → maximize efficiency
  ↳ t° doesn't need to be dropped for annealing process

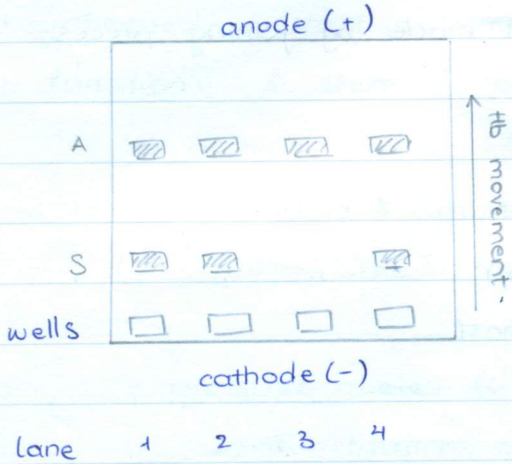
d) Gel electrophoresis

(separate dif molecules ; analysis of proteins + DNA)

- place mixture of molecules into wells cut in agarose gel + applying electric field.

Factors affecting movement speed
- • net charge of molecules
- • size of molecule
- • composition of gel : size of 'pores' within gel

Electrophoresis of proteins → separate polypeptides produced by dif alleles of many genes



- charge on protein ∝ ionisation of R groups on a.a. residues ?
  - ↳ NH_3^+ / COO^-
- charge depends on pH
  - → use buffer solution
- proteins usually −ve charged.
- polypeptides separated due to dif. net charges

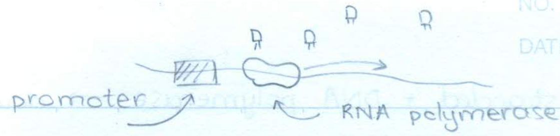
Electrophoresis of DNA

- a region of DNA is chosen
- DNA extracted
- DNA chopped to pieces using restriction endonucleases
- fragments transferred to absorbent paper
  - → placed onto gel → heat ∴ separate 2 DNA strands
- "probes" ("short sequences of single stranded DNA) added
  - ↳ radioactive P isotope → X-ray → darken film → visible

e) Plasmids as vectors in gene cloning

↳ small circles of double-stranded DNA

- small → easy to use
- exist naturally in bacteria → take up plasmids from surroundings
- can be • produced artificially → recombinant DNA
- double stranded ∴ can insert genes from prokaryotes + eukaryotes
- replicate independently in bacteria
- can be transferred between dif bacterial species



f) <u>Promoters</u> : **control expression of genes**

→ ensure high levels of gene expression

- promoter binds to DNA strand
  - allows RNA polymerase to bind to DNA
  - ensures RNA polymerase recognises template strand

↳ promoter region = transcription start point

→ enzymes producing

g) <u>Genetic markers</u> florescent substances

* antibiotic resistance gene markers → spread to other bacteria (new strains)

- GFP (green fluorescent protein) from jellyfish :
  - gene inserted into plasmid → taken up by bacteria
  - shine UV light → identify genetically modified bacteria
- GUS (β - glucuronidase) from E. coli :
  - transformed cell incubated with colorless / non-fluorescent substrate

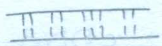
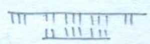
    → transform into coloured / fluorescent products

  ↳ detect activity of inserted genes

h) • **restriction endonucleases** : restrict viral infections by recognising and breaking down DNA of invading viruses

↳ binds to specific target site on DNA (sequence of bases)

→ cut sugar-phosphate backbone :

- straight across → blunt ends 
- staggered fashion → sticky ends* 

* sticky ends : short lengths of unpaired bases

↳ easily form H-bonds with complementary base sequences on other fragments of DNA cut w̄ same restriction enzyme complementary

→ single stranded + DNA polymerase → double stranded cDNA

- **reverse transcriptase**: using mRNA as template
  → produce single-stranded DNA

- **DNA ligase**: link together sugar-phosphate backbones of DNA molecules and plasmid
  → produce closed circle of double stranded DNA containing new gene : "recombinant DNA"

i) <u>Microarrays</u>

- identify genes present in an organism's genome
- find out which genes are expressed within cells

↳ microarrays contain thousands of gene probes

1. Genome analysis compare 2 types of DNA

- DNA collected, cut to fragments, denatured
  → labelled with fluorescent tags
- DNA <u>samples</u> mixed together
  → hybridise with DNA probes on microarray → inspected with UV
  - colour : DNA hybridised with probe
  - no colour: DNA not hybridised, gene not present

2. Gene expression - detecting mRNA

↳ to identify genes that are being transcribed to mRNA

- mRNA collected → reverse transcriptase → cDNA
- cDNA labelled with flucrescent tags, denatured, hybridise with probes on microarray
- <u>spots</u> that fluoresce on microarray = transcribed genes

↳ intensity of light emitted from spots
  = level of activity of gene

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19.2. Genetic technology applied to medicine

a) Bioinformatics : collecting, processing and analysis of biological info and data using computer software

b) Bioinformatics build databases which hold gene sequences, sequences of complete genomes
↳ can be matched, degrees of similarity calculated
close similarities indicate recent common ancestry

- human genes may be found in other organisms
↓
model for investigating the way such genes have their effects
- Plasmodium genome : used to find new methods to control parasites
↳ read gene sequence : develop vaccines for malaria

c) Advantages of using human proteins produced from recombinant DNA

- insulin : reliable supply available for increasing demand into bacteria
not dependent on factors e.g. meat trade
↳ act faster than animal insulin or slower over long time period
- factor VIII : genetically modified hamster cells produce factor VIII
→ extracted, purified → treat ^ w̄ haemophilia
– avoid risk of infection (e.g.: HIV from donated blood)
- adenosine deaminase (ADA) : treat severe combined immunodeficiency disease (SCID)
- genetically modified insect larvae : cabbage looper moth caterpillar
↳ administered to patients when :
- waiting for gene therapy
- gene therapy not possible

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d) Genetic screening : analysis of a person's DNA to check for the presence of a particular **allele**.

* available for: adults, fetus, embryo ...

- BRCA 1 and BRCA 2 : faulty alleles → breast cancer
  → elective vasectomy
- pre-implantation genetic diagnosis (PGD):
  - IVF procedure (sperm + egg → into dish)
    ↳ eight-cell stage → remove 1 cell → analyse DNA for genetic diseases alleles
    - no : embryo chosen for implantation
    - yes : embryo discarded

  ↳ avoid pregnancies with haemophilia, sickle cell anaemia, Huntington's disease, cystic fibrosis ...

- provides info about ↑ risk of ∧ having genetic conditions
- ∧ prepare for late onset of genetic conditions : Huntington's disease
- identify whether embryos from IVF will develop genetic c.
- " fetus that needs early treatment
- helps provide early diagnosis

e) Ethics of genetic screening

- fetus screening for genetic disease :
  - amniocentesis : look for chromosomal mutations
  - chorionic villus sampling : • minor defect cases → termination

  → risk of miscarriage ↑
- sex preselection : terminate if wrong sex → use PGD to select
- therapeutic abortions : terminating pregnancies for medical reasons

Gene therapy: treatment of a genetic disorder by altering a person's genotype (insert normal alleles of genes into cells)

- Common vectors:
  - viruses : retrovirus, lentivirus, HIV, adeno-associated virus (AAV)
  - liposomes : (small spheres of phospholipids)
  - naked DNA

- **Severe combined immunodeficiency (SCID)**

↳ crippled immune system

↳ sufferers die at infancy due to normal infections

--- * SCID: inability to make adenosine deaminase (ADA)

- alleles of ADA gene introduced into T-lymphocytes via virus vector

↳ taken out

↳ NOT PERMANENT

- vector retrovirus: insert genes randomly into host's genome

if → insert into another gene / regulatory sequence of a gene

activate nearby gene → cause cancer

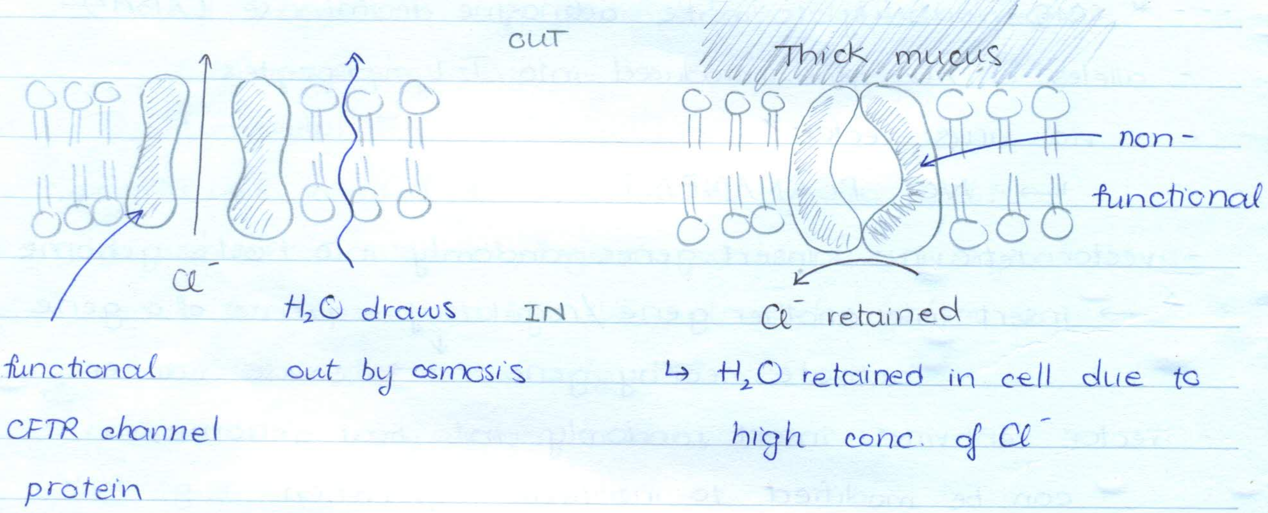
- vector lentivirus: insert randomly into host genome, can be modified to inactivate replication e.g: HIV
- vector adeno-associated virus (AAV): does <u>not</u> insert gene into host genome → ∅ passed to daughter cells (successful with long-living cells)

↳ successful gene therapies:

- cystic fibrosis
- SCID
- hereditary blindness (Leber congenital amaurosis)

- **Cystic fibrosis** : genetic disorder : abnormally thick mucus produced in lungs + other body parts
  ↳ recessive (mutated) allele of gene of transporter protein CFTR for Cl^- ions

- deletion mutation of 3 bases (AAA) in CFTR gene
- role of CFTR : transport Cl^- across epithelial cell membranes (pancreas, lungs - alveoli)
  → water follows via osmosis → membranes moist + runny
- sufferers of cystic fibrosis : CFTR non-functional
  → H_2O retained → dry membranes + sticky mucus



- Symptoms
- mucus accumulates in lungs → breathing difficulties + infections
- mucus blocks pancreatic duct → oral enzymes help digestion
- male infertility : thick secretion blocks ducts

cells involved in sexual reproduction:

- Somatic + germ cell therapy → gametes/early embryo
  - placing alleles into body cells
  - allele in 'germ line' → passed on through generations

g) Applications

- Electrophoresis of DNA: → genetic profiling (fingerprinting) in forensic science
- PCR → forensic science: amplify DNA from small tissue samples → solve crimes

19.3. Genetically modified organisms in agriculture

Social implications of using GMO in food production

- modified crop plants — agricultural weeds invade crop habitats
- introduced gene(s) may be transferred by pollen:
  - to wild relatives → more invasive hybrid offspring
  - to unmodified plants on farms with organic certification
- modified plants → toxic, produce allergies → humans + animals
- herbicides will leave toxic residues on crops
- GM seeds are $$ = $$ herbicides → no advantage
- growers need to buy new seeds each season
- lose traditional varieties

Herbicide - resistant crops

↳ fields sprayed with herbicide

kills weeds that compete for space, light, water, ions

→ ↑ crop yield

Oil seed rape.

- source of vegetable oil ; biodiesel fuel
- modified → resistant to herbicide glyphosphate.
  (inhibits synthesis of 3 a a : phenylamine,
  tyrosine, trytophan)
  - glyphosphate absorbed through leaves → growing tips
- GM : gene transferred from bacterium

Tobacco.

- resistant to herbicides : sulfonylurea + dinitroaniline
- genes taken from other plant species

Effects on environment:

- GM plant becomes agricultural weeds
- pollen will transfer gene to wild relatives
  → hybrid offspring → invasive weeds
- herbicide - resistant weeds evolve because so much of the same herbicide is used.

Genetic engineering

- improve quality (nutrition) + yield of crop plants + livestock
  ↳ solve demand for food in the world

1. Golden Rice (pro - vitamin A enhanced)

- g.m.ed → produce large quantities of β - carotene in endosperm
  ↳ human cells convert to Vitamin A.
- same yield, pest resistance, eating qualities as original varieties
- normally :
  - deficiency of Vitamin A → blindness + mortality
    ↳ immune deficiency syndrome
  - Vitamin A = fat soluble (oily - fish, dairy, liver)
  - Pro- vA present in aleurone layer, not endosperm in rice
- genes for carotene production taken from → inserted into rice
  - daffodils
  - common soil bacterium Panteea ananatis

★ ethical implications ?

- some organizations condemn Golden Rice : wrong way to solve → need to solve poverty, political, cultural, economic issues → ↓ poverty = more varied diet

2. GM Atlantic salmon

growth hormone regulating gene (Pacific Chinook salmon) + promoter (ocean pout) → injected into fertilised egg of salmon
↳ salmon reach market size in 1/2 time (18 months)

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Insect-resistant crops

↳ protect against insect pests → ↑ yield

Effects on environment:

- evolution of resistance by insect pests
- damaging effects on other insect species
- transfer of added gene to other plant species

Cotton - protected against e.g.: boll weevil

Bt maize - protected against corn borers

- Bt toxin : - lethal to insects that eat it
  - harmless to other animals

  taken from bacterium Agrobacterium tumefaciens
- GM crop plants with Bt toxin gene → produce own insecticide
- Bt resistance in corn borers: recessive allele

Adult corn borers in refuges* supply dominant allele to counteract resistance when mate with borers from fields

* non GM maize

Gene code for insulin obtained → insert into plasmid

- extract mRNA
- reverse transcriptase → cDNA $\xrightarrow{\text{DNA polymerase}}$ double-stranded
- cut with restriction enzyme with cDNA and insulin gene
  ↳ complementary sticky ends ↓ mixed
- DNA ligase → join sugar-phosphate backbone

How bacteria can be GMed → identified using antibiotic resistance genes

- recombinat plasmids mixed with bacteria
- some bacteria take up plasmids → transformed
- heat shock, Ca^{2+} solution

identify bacteria containing plasmid

- grow on agar containing antibiotic
- plasmid contains antibiotic resistant genes → survive

identify recombinant bacteria

- replica plate → onto agar with 2nd antibiotic
- if recombinant → resistance gene deactivated
- colonies on 1st plate do not grow on 2nd plate