AQA A2 Unit 5

What is a stimuli? a change in the internal or external environment

Why do organisms need to respond to stimuli? for survival (predator/prey awareness, homeostasis)

What is taxis? directional response to a stimuli (towards or away from)

What is kinesis?

- non-directional movement from an unfavourable area to a favourable
- organism moves rapidly and randomly in unfavourable area until they reach favourable area where they move slowly and less randomly
- spend more time in favourable area, less time in unfavourable area

What is tropism?

- directional growth in plants in response to a stimuli
- towards = positive, away = negative
- light = photo, water = hydro, gravity = geo
- shoot shows positive phototropism and negative geotropism
- root shows positive geotropism and positive hydrotropism
- controlled by a Plant Growth Factor = IAA

What is a plant growth factor? equivalent to animal hormones – difference: made by cells throughout the plant, only affects cells locally, just causes growth

What are the affects of IAA? promotes growth in the shoot, inhibits growth in the root

How does positive phototropism in the shoot take place?

- normally: shoot tip produces IAA, sending it down both sides causing the shoot to grow forwards
- if light is present on one side, the IAA redistributes to the opposite side (shaded side)
- this causes the opposite side to grow faster
- so the shoot bends towards the light

Evidences for tropism?

- removing or covering shoot tip prevents tropism [tip causes tropism]
- placing 'micin' across shoot inhibits tropism [prevents movement of chemical e.g. IAA]
- placing 'gelatine' across shoot does not affect tropism [prevents movement of electrical signals]
- when in light or darkness the overall levels of IAA remain the same [light does not inhibit or breakdown IAA but rather redistributes it]

Response to Stimuli in Mammals? uses Nervous System, Hormonal System, Local Mediators (nervous and hormonal systems coordinate response to stimuli)

How do Local Mediators work? when cells are damaged or infected they release Histamine and Prostaglandin which causes Inflammation (increase of tissue fluid – brings in white blood cells)

Job of Nervous System? coordinate response to certain stimuli – response is fast, short acting, localised

Pathway of Nervous System? stimuli to receptor to sensory neurone to spinal cord to brain to spinal cord to motor neurone to effector = response

What does a receptor do?

- detects stimuli
- converts stimuli energy into nerve impulse
- (acts as a transducer – converts one type of energy into another)
- uses stimuli energy to send Na+ ions into the start of the sensory neurone
- each type of stimuli has a specific receptor
- 2 examples of receptors: Pacinian Corpuscle, Eye

What does a Pacinian Corpuscle do?

- touch receptor
- found in skin, fingers and toes
- responds to pressure/touch
- structure = corpuscle (several layers of tissue) wrapped around the start of a sensory neurone
- process = pressure applied, corpuscle compressed, stretch-mediated Na+ channels opened, Na+ ions move into the start of the sensory neurone

How does the Eye work?

- detects light so the brain can generate an image
- detected by retina (located at back of eye)
- made of cone and rod cells
- cone cells detect high light intensity only, produces colour image, high visual acuity
- rod cells can detect low light intensity, produces black and white image, low visual acuity
- cone cells located in centre of retina (fovea)
- rod cells located in periphery of retina
- cone cells made of iodopsin pigment, has one cone cell to one sensory neurone (no summation = can only detect high light intensity, can distinguish each stimuli = high visual acuity)
- rod cells made of rhodopsin pigment, has several rod cells to one sensory neurone (summation = can detect low light intensity, merges the stimuli = low visual acuity)

What is the Central Nervous System (CNS)?

- made of brain and spinal cord
- brain = analyses and coordinates response to stimuli
- spinal cord = connects brain to sensory and motor neurones

What is the Peripheral Nervous System (PNS)?

- made of the sensory and motor neurone
- sensory neurone takes nerve impulse from receptor to CNS
- motor neurone takes nerve impulse from CNS to effector
- a neurone transmits a nerve impulse
- sensory neurone has its cell body in the middle and has a dendron and axon
- motor neurone has its cell body and the start and has a long axon

What are the 2 different types of Motor Neurone?

- Voluntary (Somatic) and Involuntary (Autonomic) motor neurones
- Somatic supplies skeletal muscle = under conscious control
- Autonomic supplies cardiac muscle, smooth muscle, glands = under subconscious control
- Autonomic can be divided into Sympathetic and Parasympathetic (have opposite effects)

What is a nerve impulse?

- movement of an action potential along a neurone
- action potential = change in membrane potential in one section of the neurone
- changes from negative (polarised) to positive (depolarised) back to negative (repolarised)

What is resting potential?

- membrane potential of neurone at rest
- is -65mV
- polarised
- caused by having more positive ions outside compared to inside
- involves Na+/K+ pump, pumping 3 Na+ ions out, 2 K+ ions in
- K+ channel allowing K+ ions to leave

What happens during an Action Potential?

- stimuli causes Na+ ions to enter the neurone
- makes membrane potential less negative
- if it reaches threshold (-50mV), Na+ channels open
- more Na+ ions enter the neurone, membrane becomes depolarised (positive)
- the potential reaches +40mV
- then the Na+ channels close, the K+ channels open
- K+ ions move out, membrane becomes repolarised (negative)
- too many K+ ions move out, so the membrane becomes more negative than normal =

hyperpolarised
- one action potential = depolarisation, repolarisation, hyperpolarisation

How does an Action Potential move along a neurone?

- by local currents
- Na+ ions that move in during depolarisation diffuse along the neurone causing the next section to reach threshold and an AP to occur
- this continues along the neurone
- * an AP will always move along the neurone
- * why does it not move back? Previous section is in refractory period (Na+ channels cannot be opened) and is hyperpolarised (threshold cannot be reached)

How does the size of stimuli affect a nerve impulse?

- does not affect size of AP (AP is all or nothing – get AP always with same size OR no AP)
- larger stimuli increases the frequency (number) of AP

What affects speed of nerve impulse?

- temperature = higher temp, higher kinetic energy, faster rate of diffusion, faster nerve impulse
- axon diameter = wider diameter, neurone less leaky, faster nerve impulse
- myelination = schwann cells wrap around axon, insulates axon, AP occurs in gaps – node of ranvier, so AP jumps from node to node = saltatory conduction

What is a synapse?

- connection between 2 different neurones
- sends nerve impulse across the gap (synaptic cleft) using neurotransmitters (e.g. acetylcholine)
- AP arrives in presynaptic neurone
- Ca2+ channels open
- Ca2+ ions enter presynaptic neurone
- causes vesicles containing neurotransmitter to move to membrane
- vesicle binds to membrane releasing neurotransmitter into cleft
- neurotransmitter diffuses across cleft
- binds to complementary receptors on postsynaptic membrane
- Na+ channels open, Na+ ions enter
- if threshold is reached, AP occurs
- (enzyme used to breakdown neurotransmitter, e.g. acetylcholinesterase breaksdown acetylcholine into ethanoic acid and choline, diffuses back into presynaptic neurone, ATP used to reform vesicle and actively transport Ca2+ ions out)

What are the properties of synapses?

- unidirectionality = AP/nerve impulse travels in one direction, from pre to post, pre has the neurotransmitter, post has the receptors
- filters out low level stimuli = low level stimuli does not release enough neurotransmitter for threshold to be reached on postsynaptic neurone
- summation = low level stimuli add together to produce an AP in postsynaptic neurone, can be temporal or spatial, temporal = low level stimuli present for extended period of time, spatial = a few different low level stimuli add together
- inhibitory = normal synapses are excitatory (cause AP), some can be inhibitory – prevent action potential from occurring by making postsynaptic neurone hyperpolarised

What is a reflex?

- a rapid involuntary response to a stimuli
- does not use the brain
- the sensory neurone connects directly to motor neurone
- ensures less damage done and does not require learning

How is heart rate controlled?

- the heart is myogenic, its heart beat is initiated by the SAN
- the Medulla Oblongata in the brain can increase or decrease heart rate
- receives nerve impulse from chemoreceptors (respond to blood pH) in the carotid arteries and pressure receptors (respond to blood pressure) in the carotid arteries and aorta
- sends impulse in sympathetic to SAN to increase HR and sends impulse in parasympathetic to SAN to decrease HR

What is the job of the skeletal muscle?

- moves the body skeleton
- when the muscle contracts (shortens) the tendon pulls on joints causing movement

Structure of skeletal muscle?

- basic structure = sarcomeres (made up of actin and myosin, actin is thin and has tropomysosin wrapped around it, myosin is thick and has heads), when the sarcomere contracts the whole muscle contracts, contracts/shortens by the sliding filament mechanism
- many sarcomeres = myofibril
- many myofibrils = muscle fibre (surronded by a membrane called sarcomere, contains fluid called sarcoplasm, contains tubes called sarcoplasmic reticulum)
- many muscle fibres = bundle
- many bundles = whole muscle

Locations in a sarcomere?

- A band = location of myosin [no change in contraction]
- I band = location between the myosin [shortens in contraction]
- H zone = location between the actin [shortens in contraction]
- Z line = end line of sarcomere [moves closer together in contraction]

What occurs in sliding filament mechanism?

- how the sarcomere shortens
- the myosin heads pull the actin inwards
- the somatic motor neurone connects to the skeletal muscle via a neuro-muscular junction
- one motor neurone connects to a few muscle fibres = motor unit
- releases acetylcholine that binds to complementary receptors on the muscle fibre membrane (sarcomere)
- Na+ channels open, Na+ ions enter the muscle fibre causing depolarisation
- wave of depolarisation travels thru sarcoplasmic reticulum
- causes release of Ca2+ ions into the sarcoplasm
- this moves the tropomyosin on the actin
- exposes binding sites on the actin
- myosin heads now bind to the actin (cross bridge)
- a power stroke occurs, the myosin pulling the actin inwards
- ATP attaches to myosin head so it detaches
- ATP brokendown by ATPase to release energy
- myosin head goes back to its original position
- so it reattaches, pulling the actin further inwards

Role of Ca2+ ions and ATP in muscle contraction?

- Ca2+ ions causes the tropomyosin to move exposing binding sites on actin
- Ca2+ ions stimulate ATPase
- ATP causes myosin head to detach
- ATP releases energy so myosin head returns to original position
- ATP actively transports Ca2+ ions into sarcoplasmic reticulum when the muscle is relaxed

What are the 2 types of muscle fibres? fast twitch and slow twitch

How does fast twitch muscle fibres work?

- provide powerful but short lasting contractions
- found in biceps and sprinters
- has thicker myosin
- contains more enzymes for anaerobic respiration
- contains phosphocreatine, provides phosphate to ADP to reform ATP

How does slow twitch muscle fibres work?

- provide less powerful but long lasting contractions
- found in thigh and marathon runners
- adapted for aerobic respiration
- has a rich blood supply
- contains many mitochondria
- contains glycogen
- contains myoglobin (stores oxygen)

What is homeostasis?

- maintenance of a constant internal environment (the blood and tissue fluid)
- control body temperature, glucose levels, water levels, salt levels, pH, oxygen levels, carbon dioxide levels

Homeostasis and negative feedback? the response to the change is to oppose the change

What is positive feedback? the response to the change is to continue the change (e.g. Na+ ions entering a neurone stimulating more to enter in depolarisation)

Why do organisms need to maintain a constant body temperature? maintain optimum temperature for enzyme activity

What are endotherms and ectotherms?

- endotherms = maintain a constant internal body temperature irrespective of environmental temperature
- ectotherms = internal body temperature varies with changes in environmental temperature

How is internal body temperature controlled? anatomical, behavioural, physiological changes

Anatomical adaptations in organisms in warm areas?

- small body size = large surface area to volume ration (lose heat)
- less fur
- less fat
- large extremities (lose heat)

Anatomical adaptations in organisms in cold areas?

- large body size = small surface area to volume ratio
- more fur
- more fat
- small extremities

Behavioural/Physiological changes in ectotherms?

- warming up = expose to sun, press on warm surface, darker skin colouration, more respiration in liver, less breathing
- cooling down, shade from sun, press on cold surface, lighter skin colouration, less respiration in liver, more breathing

Control of body temperature in endotherms?

- controlled by Hypothalamus in the brain
- receives nerve impulse from peripheral thermoreceptors in the skin and central thermoreceptors in the hypothalamus
- peripheral thermoreceptors monitor changes in environmental temperature
- central thermoreceptors monitor changes in core body temperature

How a endotherm warms itself up?

- reduce blood flow to the skin surface = vasoconstriction, smooth muscle in arterioles to the skin contract, lumen narrows, less heat lost from blood
- hair on skin stands up = hair erector muscles contract, hairs stand up, traps in air particles, forms an insulating layer, reduces heat loss
- shivering = involuntary contraction of muscles – friction in sliding filament mechanism generates heat and respiration generates heat
- increase respiration in liver = generates heat

How an endotherm cools itself down?

- increase blood supply to skin surface = vasodilation, smooth muscle in arterioles to the skin relax, lumen widens, more heat lost from blood
- sweating = evaporation of water particles from the skin surface using the heat in the blood

Control of blood glucose levels?

- if high = should be in cells for respiration, also lowers blood water potential
- if low = not enough to supply cells of the brain, also increases blood water potential
- controlled by the Pancreas
- contains the Islets of Langerhans
- made of alpha and beta cells
- alpha cells produce glucagon
- beta cells produce insulin

High blood glucose levels?

- occurs after a meal
- insulin is released
- most cells in the body have complementary receptors (particularly muscle, liver, brain cells)
- causes increase in glucose channels and carriers
- glucose taken up and used in respiration
- in muscle and liver cells, glucose converted into glycogen (glycogenesis)

- in liver cells, glucose converted into fat

Low blood glucose levels?

- occurs after starvation or exercise
- glucagon is released
- only liver cells have complementary receptors
- convert glycogen into glucose (glycogenolysis)
- convert fats and amino acids into glucose (gluconeogenesis)

Diabetes?

- person loses control of blood glucose levels
- normally high (hyperglycaemia)
- 2 types: type 1 and type 2
- type 1 starts at young age, person does not make insulin, beta cells damaged by an autoimmune disorder (treatment = insulin injections)
- type 2 starts at middle age, person makes insulin but cells are less sensitive, caused by obesity and diet high in simple sugars (treatment = diet and exercise, drugs, insulin injection)
- symptoms = tiredness, increase urination, thirst
- diagnosis = high blood glucose levels

Control of water levels?

- controlled by Hypothalamus
- contains osmoreceptors
- dehydrated = ADH released by pituitary gland, more water reabsorbed by kidneys
- overhydrated = less ADH released, less water reabsorbed, more lost in urine

What is the oestrous cycle? Monthly cycle (28 days) in female mammals that prepares individual for pregnancy = development of uterus lining and release of egg cells

Hormones in oestrous cycle?

- FSH = released by pituitary gland, stimulates development of follicle (contains the egg)
- LH = released by pituitary gland, stimulates ovulation (release of egg from follicle)
- Oestrogen = released by ovaries, causes development of the uterus lining
- Progesterone = released by ovaries, causes maintenance of uterus lining

The oestrous cycle?

- Day 1-5: uterus lining breaksdown, FSH is released, follicle develops
- Day 6-12: developing follicle releases oestrogen, uterus lining develops
- Day 13: high levels of oestrogen cause a LH surge, causing ovulation
- Day 14-28: corpus luteum forms (after follicle has released egg), releases progesterone, maintains the uterus lining
- cycle repeats = corpus luteum breaksdown, progesterone lost, lining no longer maintained

Positive and Negative feedback in oestrous cycle?

- FSH stimulates release of oestrogen (FSH causes follicle to develop which releases oestrogen)
- Low levels of oestrogen inhibit FSH/LH
- High levels of oestrogen stimulate FSH/LH (=LH surge)
- Progesterone inhibits release of FSH/LH

How is fertility increased? individual given FSH and LH

How do female contraceptives work? individual given Oestrogen or Progesterone

Job of the Hormonal system?

- coordinates the response to certain stimuli
- involves chemical messengers released by endocrine glands (exocrine glands release substance into open spaces e.g. salivary gland) into the blood, travels to target cells causing changes
- protein hormones bind to complementary receptors on target cells, activates enzymes that convert ATP into Cyclic AMP, the cAMP then makes changes in the cell (=2nd messenger system)
- lipid hormones enter cells by simple diffusion and cause direct changes e.g. oestrogen

How does a gene code for a protein?

- gene is made up of a sequence of bases
- contains exon and introns (exon = coding DNA, intron = non-coding DNA)
- the exon codes for a protein
- each 3 bases code for an AA (codon)
- the sequence of bases determined the sequence of codons which determines the sequence of AAs (= primary structure of protein)
- the codon is degenerate (each AA has more than one codon)
- the codon is non-overlapping (each base is read only once)
- there are stop codons (at the end of the sequence, do not code for an AA)

How is a protein assembled? by transcription and translation

What is transciption?

- producing a complementary single stranded RNA copy of a gene
- produces mRNA
- involves DNA Helicase breaking the hydrogen bonds between complementary bases in the gene
- the double strand of the gene unwinds
- leaves 2 separate strands
- RNA nucleotides bind to complementary bases on one of the strands (sense strand)
- RNA Polymerase joins the sugar-phosphate backbone of the RNA strand
- leaves pre-mRNA (contains introns and exons)
- the copies of the introns are removed by splicing
- leaves mRNA

What is translation?

- using the sequence of codons on the mRNA to assemble the protein
- mRNA attaches to a ribosome
- ribosome covers the first 6 bases (first 2 codons)
- complementary tRNA bind to the codons via their anticodon
- the AAs on the tRNA are joined by peptide bonds
- the ribosome moves along by one codon
- a complementary tRNA binds to this
- its AA is attached to the first 2
- this continues along the whole mRNA
- until the last codon is reached
- this is a stop codon (the complementary tRNA does not carry an AA)
- leaves the complete polypeptide chain

DNA vs RNA?

- deoxyribose sugar vs ribose sugar
- thymine vs uracil
- double stranded vs single stranded
- one type vs two types (mRNA and tRNA)

Structure of tRNA?

- single stranded RNA folded over into a 'clover leaf' shape (held by hydrogen bonds between the bases)
- has an AA attachment site on the top
- has 3 specific bases on the bottom (anticodon)

What is a Gene Mutation?

- a change in the base sequence of DNA
- 2 types = substitution and deletion
- substitution = replace one base for another, changes one triplet code
- substitution can be silent (new triplet code codes for same AA), mis-sense (codes for a different AA), non-sense (codes for a stop codon)
- deletion = loss of a base, causes frameshift, all the triplet codes after the mutation changes

What is cancer?

- uncontrolled cell division
- leads to formation of a tumour
- caused by mutation of genes that control cell division
- causes of mutation = random or mutagens (chemicals/radiation)
- genes that control cell division = proto-oncogene and tumour-suppressor gene
- proto-oncogene stimulates cell division (produces growth factor and receptor proteins, when growth factors bind to receptor proteins on cells activates dna replication and cell division)
- tumour-suppressor gene inhibits cell division

- mutation of proto-oncogene leads to formation of a oncogene = over production of growth factor or receptor proteins permanently active = uncontrolled cell division
- mutation of tumour-suppressor gene = loss of inhibition = uncontrolled cell division

What is a stem cell?

- a unspecialised/undifferentiated cell
- pluripotent/totipotent (can form any type of cell)
- found in zygote (embryonic stem cells) of plants and animals
- found in adult plants (supports asexual reproduction e.g. by cuttings)

How does a stem cell be come a specialised cell? certain genes are activated and other genes are inhibited, so certain proteins are made, so it becomes different

Example of activating genes?

- using oestrogen
- normally transcriptional factors (promote transcription of a gene) are blocked by an inhibitor
- oestrogen can enter a cell and bind to receptors on the transcriptional factor
- causes transcriptional factor to change shape
- releases inhibitor
- now enters nucleus binding to promoters on the DNA to activate transcription
- = activated genes

Example of inhibiting genes?

- using siRNA (small interfering RNA)
- making siRNA = double stranded RNA cut down into small sections, made single stranded then attaches to an enzyme
- siRNA will bind to complementary sections on mRNA = the enzyme will cut the mRNA so translation cannot occur = gene inhibited

What is genetic engineering? changing the genetic makeup of an organism's DNA by adding or removing a gene

Why do we genetically engineer bacteria? so they can make useful proteins

Why do we genetically engineer plants/animals?

- they can make useful proteins
- give them additional characteristics

Genetically engineering bacteria?

- to make useful proteins like insulin

- normally used animal sources (problems = limited supply, infection risk, immunorejection)
- involves adding human insulin gene to a plasmid, then inserting this into a bacteria = the bacteria now has the code to produce the insulin protein
- involves 5 steps = 1. Isolation, 2. Insertion, 3. Transformation, 4. Identification, 5. Growth/Cloning
- 1. Isolation
- either by Reverse Transcriptase or Restriction Enzyme
- RT = found in virus, converts RNA into DNA, obtain mRNA for insulin, the RT will convert it into cDNA (single stranded complementary DNA), nucleotides and DNA Polymerase added to make it double stranded
- RE = found in bacteria, cuts DNA at certain base sequences (called recognition sites), can cut straight or staggered, staggered used in GE as it leaves exposed bases called 'sticky ends'
- 2. Insertion
- cut plasmid using the same RE
- leaves complementary sticky ends
- join gene with plasmid via the sticky ends
- use DNA Ligase to join the sugar-phosphate backbone
- = recombinant plasmid
- 3. Transformation
- mix recombinant plasmid with bacteria
- add Ca2+ ions and vary temperature
- bacteria will become permeable and take up the recombinant plasmid
- 4. Identification
- identify which bacteria have taken up the recombinant plasmid and of these which ones have accepted the new gene (the insulin gene)
- step 1 = choose a plasmid that carries an Ampicillin Resistant Gene, so when Ampicillin is added only the bacteria that have taken up the recombinant plasmid will survive
- step 2 = use gene markers (antibiotic resistant, fluorescent, enzyme) to identify which of the remaining bacteria have accepted the new/insulin gene, if they have they will no longer have these properties
  - antibiotic resistant = tetracycline resistant gene lost if insulin gene is accepted, so no longer resistant to tetracycline, add tetracycline by replica plating (on another plate that carries a few of the bacteria in their same position), the ones that die are the ones that we want, obtain from original plate
  - fluorescent = fluorescent lost in bacteria that have accepted insulin gene, so identify bacteria showing no fluorescence
  - enzyme = enzyme not made in bacteria that have accepted insulin gene, add colourless substrate, where there is no colour change select those bacteria
- 5. Growth/Cloning
- grow recombinant bacteria
- they will produce the protein (e.g. insulin)

Examples of genetic engineering in plants?

- for disease resistance

- for pest resistance
- for herbicide resistance
- produce genetically modified tomatoes = prevented from softening, remain hardened (easy for storage and transport), involves preventing formation of softening enzyme, a gene is added that is complementary to the the softening enzyme gene, so its mRNA will bind to the mRNA of the softening enzyme preventing translation

Examples of genetic engineering in animals?

- for disease resistance
- add gene for growth hormone for growth
- use to produce anti-thrombin = protein used to make blood clot (people with certain genetic disease may not produce), use milk producing animal to produce, add gene for anti-thrombin next to milk producing gene in animal, protein will be made in the milk (easily extracted)

What is PCR?

- polymerase chain reaction
- used to replicate DNA for forensic testing
- step 1: heat to 95oC, hydrogen bonds break, double strand separates, left with 2 template strands
- step 2: cool to 55oC, primers bind (short single stranded sections of DNA) to start of each strand, prevent the templates from rejoining and allows DNA Polymerase to bind
- step 3: heat to 72oC, DNA nucleotides attach to complementary bases, DNA Polymerase joins sugar-phosphate backbone of the new strands

In-vivo vs In-vitro method of DNA Replication?

- in-vivo = using bacteria to replicate DNA (add DNA to the plasmid by restriction enzymes), then replicate the bacteria
- in-vitro = PCR
- benefits of in-vivo = more accurate (less mutations), less chance of contamination
- benefits of in-vitro = more rapid, less complicated

How to work out the sequence of a gene?

- by Sanger Method
- make many copies of the gene to be copied
- ensure all the copies are single stranded
- set up 4 test tubes
- to each test tube add the many single stranded copies of the gene, primer, dna nucleotides and dna polymerase
- to each test tube add a different terminator nucleotide (a nucleotide that when it binds prevents any other base from joining after, is radioactively labelled)
- so terminator A in test tuble 1, terminator T in test tube 2, terminator C in test tube 3, terminator G in test tube 4
- each test tube will produce many DNA fragments of different lengths
- Gel Electrophoresis is used to separate out the DNA fragments
- involves placing a charge across the gel (negative at the start) which causes the DNA to move along the gel (is negatively charged), the smaller fragments will move further

- so the dna fragments of each test tube is placed in a different well (= 4 wells at the start), the dna fragments are separated out based on their size, each will occupy a specific position (each has a different length)
- an x-ray can be applied to make the fragments visible (terminator is radioactive), the sequence can be read from the position furthest from the start

What is gene therapy?

- adding a healthy allele to treat a genetic disorder
- can occur by replacing the defective allele (gene replacement)
- or by adding a dominant healthy allele alongside the recessive defective allele (gene supplementation)
- can target embryonic stem cells (germ line gene therapy) or adult cells (somatic cell gene therapy)
- germ line gene therapy = provides a cure, all the cells will have the healthy allele, currently illegal as it may cause mutation of embryo and unethical
- somatic cell gene therapy = treats the adult cells directly affected by the defective allele, problem is that it is a short term solution (treated cells will die and be replaced by defective cells)

Example treating Cystic Fibrosis?

- caused by inheriting a defective allele for the CFTR Gene so normal Cl- ion channels will not be produced
- therefore mucus remains thick and sticky
- normally, the Cl- ion channels allow Cl- ions to leave the cell and enter mucus, this lowers water potential of mucus, so water follows by osmosis, mucus will be thin and runny
- without the functioning Cl- ion channels, this process cannot occur, so mucus remains thick and stick
- in the lungs this causes breathing difficulties
- in the pancreas this blocks pancreatic duct, so digestive enzymes cannot reach the small intestine
- in the testis this blocks the sperm duct, causing infertility
- can be treated by Gene Therapy
- delivering the healthy allele for the CFTR Gene to the lung cells
- can be delivered by Virus or Liposome
- virus = use an adenovirus, add healthy allele to a plasmid, add plasmid to the adenovirus, attenuate adenovirus so it cannot replicate, the adenovirus will be inhaled and enter lung cells, delivering the healthy allele
- liposome = a lipid coat, placed around a plasmid carrying the healthy allele, the liposome is inhaled and enters lung cells by simple diffusion, delivering the healthy allele

Example treating SCID?

- severe combined immunodeficiency
- person does not make ADA enzyme (Adenosine Deaminase)
- ADA normally breaksdown toxins

- these toxins remain
- the toxins destroy the WBCs
- person loses their immune response
- susceptible to all infections
- can be treated by Gene Therapy
- remove t cells from the patient
- add healthy allele for ADA onto a plasmid, place plasmid in a retrovirus, attentuate retrovirus so it cannot replicate
- mix patient t cells with the retrovirus
- the virus will enter the t cells delivering the healthy allele
- the treated t cells can by inserted back into patient

What is genetic screening?

- analyse an individual's DNA for the presence of a particular gene (e.g. defective/mutated gene)
- use DNA Probes (single stranded section of DNA, complementary to a particular gene, is radioactively labelled)
- obtain individuals DNA, make it single stranded, add the specific DNA Probe for the gene to be screened for, if the gene is present the DNA Probe will bind, will show up as radioactivity on an X-ray film

What is genetic fingerprinting?

- used to produce a unique 'fingerprint' of an individual's DNA (produces a specific banding pattern)
- used in forensic and paternity testing
- involves comparing the individual's introns (non-coding DNA)
- introns contain repetitive sequences called Core Sequences
- the number and length of the core sequences are unique for each individual organism
- involves 5 steps: 1. Extraction, 2. Digestion, 3. Separation, 4. Hybridisation, 5. Development
- 1. Extraction
- extracting the individual's DNA
- 2. Digestion
- cutting the DNA down into fragments
- use Restriction Enzymes that cut just outside the core sequences (leaves the core sequences)
- 3. Seperation
- separate out the dna fragments by gel electrophoresis
- add alkali to make the separated fragments single stranded
- transfer the fragments to a nylon membrane by Southern Blotting
- add UV light so the dna fragments set
- 4. Hybridisation
- add radioactively labelled DNA Probes complementary to the DNA fragments
- 5. Development