(a) 1. Cut (DNA) at same (base) sequence / (recognition) sequence;

## Accept: cut DNA at same place

2. (So) get (fragments with gene) $\mathbf{R} /$ required gene.

Accept: 'allele' for 'gene'/ same gene
(c) 1. Fragments L from parent rr, because all longer fragments / 195 base pair fragments;

Ignore: references to fragments that move further / less, require identification of longer / shorter or 195 / 135 Accept: (homozygous) recessive
2. Fragments $N$ from parent RR, because all shorter fragments / 135 base pairfragments;

1 and 2 Accept: A3 for 195 and A4 for 135
2. Accept: (homozygous) dominant
3. (M from) offspring heterozygous / Rr / have both 195 and 135 base pairfragments.
Accept: have both bands / strips
Reject: primer longer / shorter
(d) 1. (Cells in mitosis) chromosomes visible;
2. (So) can see which chromosome DNA probe attached to.
(e) (i) 1. For comparison with resistant flies / other (two) experiments / groups;
Ignore: compare results / data / no other factors
2. To see death rate (in non-resistant) / to see effect of insecticide in non-resistant / normal flies. Accept: 'pesticide' as 'insecticide' Accept to see that insecticide worked / to see effect of enzyme
(ii) (PM must be involved because)

1. Few resistant flies die (without inhibitor);
2. More inhibited flies die than resistant flies;
3. (PM) inhibited flies die faster (than resistant flies);
(Other factors must be involved because)
4. Some resistant flies die;
5. But (with inhibitor) still have greater resistance / die slower thannon-resistant flies.

Accept: (with inhibitor) die slower than non-resistant flies
4 max
[15] (a) Reverse transcriptase;
(b) 1. Probe (base sequence) complementary (to DNA of allele $A$ / where $A$ is (and) binds by forming base pairs / hydrogen bonds; Accept gene A
2. So (only) this DNA labelled / has green dye / gives out (green) light; Accept glows for green light
(c) (i) 1. More probe binding / more cDNA / mRNA / more allele / gene A means more light;
2. DNA (with A) doubles each (PCR) cycle;
3. So light (approximately) doubles / curve steepens more and more (eachcycle) / curve goes up exponentially / increases even faster;
(ii) (G because)

1. (Heterozygous) only has half the amount of probe for $\mathbf{A}$ attaching / only half the amount of DNA / allele A (to bind to); Accept only one A to bind to
2. (So,) only produced (about) half the light / glow / intensity (of H) (per cycle of PCR);
If reference to 'half' for point 1, allow 'less light' in 2.

## Essay Using DNA in science and technology

## DNA and classification

2.2 Structure of DNA
2.3 Differences in DNA lead to genetic diversity
2.9 Comparison of DNA base sequences

## Genetic engineering and making useful substances

2.5 Plasmids
5.8 The use of recombinant DNA to produce transformed organisms that benefit humans

## Other uses of DNA

2.5 Cell cycle and treatment of cancer
5.8 Gene therapy;

Medical diagnosis and the treatment of human disease;
The use of DNA probes to screen patients for clinically important genes.
(a) (i) 1. Negative correlation;

Accept: description for 'negative correlation'
Neutral: 'correlation'
Reject: positive correlation
2. Wide range;
3. Overlap;
4. (Graph suggests that) other factors may be involved (in age of onset);

2 / 3 Accept the use of figures from the graph
2 / 3 Can refer to age of onset or number of CAG repeats
Ignore references to methodology
3 max
(ii) 1. Age of onset can be high / symptoms appear later in life;

Accept: 'gene' for 'allele'
2. (So) individuals have already had children / allele has been passed on;

## OR

3. Individuals have passed on the allele / already had children;
4. Before symptoms occur;
(b) (i) 1. Person K;
5. (As has) high(est) band / band that travelled a short(est) distance / (er) so has large(st) fragment / number of CAG repeats; Must correctly link distance moved and fragment size
(ii) Run fragments of known length / CAG repeats (at the same time);

Accept: references to a DNA ladder / DNA markers
Do not accept DNA sequencing
(iii) Homozygous / (CAG) fragments are the same length / size / mass;

5
Reject breakdown, cutting out
1
(ii) To separate the (pieces of) DNA;
(c) Complimentary base sequence / complementary DNA; binds to both (haplotypes);

Label would show up in both;
Idea of complimentarity required
2
(d) (i) Y chromosome inherited / comes from male parents / only found in males;
(ii) Mitochondria in egg / female gamete / no mitochondria come from sperm / malegamete;

1
(e) (i) Allows comparison;

Different (sized) areas covered;
(ii) Wolves do not eat all of prey animal / do not eat (large) bones / skin; Inedible parts make up different proportions / wolf eats different proportions;
(f) Limited by food / prey; as prey increases so do wolf numbers / positive correlation;

Large range so other factors involved;
[12] (a) a length of DNA;
6 that codes for a single protein / polypeptide;
(b) by heating;to break the H -bonds (between complementary bases);
(c) (i) to allow the DNA polymerase to attach / start addition ofnucleotides / mark start and end of sequence to be copied / prevents strands re-joining;
(ii) because the sequences at the ends of the target sequenceare different / one is at the beginning and one at the end;
(d) 8 ;
accept 7

1
[7] (a) 1 DNA is cut;

2 Using restriction enzyme;
3 Use electrophoresis;
4 Separates according to length / mass;
5 Southern blotting / transfer to (nylon) membrane;
6 Make single-stranded;
7 Apply probe;
8 Radioactive / fluorescent;
9 Reference to tandem repeats / VNTRs / minisatellites;
10 Autoradiography / eq;8 and 10 should be consistent
$\max 6$
(c) (i) All bands in cub which don't come from mother;

Must be in father's DNA fingerprint;
Principle that all bands in cub must come from mother and father $=$ 1
(ii) Select pairs with dissimilar DNA fingerprints;
(d) (i) Cells (from panda) in faeces / gut cells / blood cells;
(ii) To increase amount of DNA / only small amount present;
(iii) DNA / primer has specific base-sequence; Reference to specific / complementary base-pairing;
(e) Taking samples from animals causes stress / injury to animal;

Difficult to find animals;

Pandas are dangerous / threat to human;
[15] (a) 1. DNA is cut;

8
2. using restriction enzyme;
3. electrophoresis;
4. separates according to length / mass / size;
5. DNA made single-stranded;
6. transfer to membrane / Southern blotting;
7. apply probe;
8. radioactive / single stranded / detected on film / fluorescent;
9. reference to tandem repeats / VNTRs / minisatellites;
10. pattern unique to every individual;
$6 \max$
(b) cells on toothbrush;

DNA present in cell;
(c) (i) toothbrush gives small sample of DNA / need more DNAfor analysis; PCR gives many copies;
(ii) uses heat;to separate strands;

OR
PCR replicates pieces of DNA;
because DNA has been cut;
OR
primer added in PCR;
to initiate replication
2 max
(d) (i) PCR / amplification needed;
(ii) other DNA present; need to identify 'required' DNA from rest;
[15] (a) Mother and father both heterozygotes / Tt / carriers;

Probability of thalassaemia $1 / 4$ and female $1 / 2$;
Probability of both $1 / 8$;
(b) (i) Cut at same base sequence as same enzyme used;

Fragments are same length / size / have same charge;
(ii) Single base occurs many times;

Sequence of 20 unlikely to occur elsewhere;

Allow one mark for establishing the principle where neither marking point clearly made.
[7] (a) (i) Different genes / characteristics / features;

Reference to mutations;
Or
Base sequence determines protein;
Different species have different protein sequences;
$\max 2$
(ii) Primer has different DNA sequence;

DNA specific / complementary base-pairing;
(c) 1 Heat DNA;

2 Breaks hydrogen bonds / separates strands;
3 Add primers;
4 Add nucleotides;
5 Cool;
6 (to allow) binding of nucleotides / primers;
7 DNA polymerase;
8 Role of (DNA) polymerase;9 Repeat cycle many times;
$\max 6$
[15] (a) Endonuclease / restriction enzyme;
(b) DNA made of base pairs;

Each base pair is same length / occupies same distance along backbone;
(c) (i) Second blank box from left labelled 6;
(ii) Distance moved depends on length / number of base pairs /second longest fragment / second shortest distance identified;
(d) 5 ;
[6] (a) only small amounts obtained / PCR increases the amount / mass of DNA;
12
(b) (i) to separate the two strands of the DNA /to break the hydrogen bonds;
(Reject "unzip")
(ii) short lengths / fragments of DNA / nucleotides /single stranded DNA;
(iii) to mark beginning and / or ends of the part of DNA needed /for attachment of enzymes or nucleotides / initiator / keeps strands apart;
(iv) would not be denatured;must be heated to $95^{\circ} \mathrm{C} /$ must withstand high temps;
(c) 1 DNA extracted from sample;

2 DNA cut / hydrolysed into segments using restriction endonucleases;
3 must leave minisatellites / required core sequences intact;
4 DNA fragments separated using electrophoresis;
5 detail of process e.g. mixture put into wells on gel and electric current passed through;
6 immerse gel in alkaline solution / two strands of DNA separated;
7 Southern blotting / cover with nylon / absorbent paper (to absorb DNA);
8 DNA fixed to nylon / membrane using uv light
9 radioactive marker / probe added (which is picked up by required fragments) / complementary to minisatellites;
10 (areas with probe) identified using X-ray film / autoradiography;
(d) adult 3;this is only one which, (with number 1), can provide (all) the DNA fragments which children have / all bars match;
(Reject 'genes')

1 DNA heated to 90 to $95^{\circ} \mathrm{C}$;

## 13

2 strands separate;
3 cooled / to temperature below $70^{\circ} \mathrm{C}$
4 primers bind;
5 nucleotides attach;
6 by complementary base pairing;
7 temperature $70-75^{\circ} \mathrm{C}$;
8 DNA polymerase joins nucleotides together;
9 cycle repeated;
6 max
[6] (a) to separate the two strands / break hydrogen bonds;
(b) (i) enables replication / sequencing to start (allow keeps strands separate);
(ii) joins DNA nucleotides (not complementary bases);
(c) (i) 64;
(ii) replication of DNA from crime scene / tissue sample /for DNA sequencing / gene cloning;
(d) (transcription uses) RNA polymerase;RNA nucleotides / uracil; one (template) strand / PCR both strands; start / stop codons;
(accept enzyme separates strands)
[7] (a) (i) to separate polynucleotide strands / form single strands;

## 15

(ii) not denatured (at $\left.95^{\circ} \mathrm{C}\right)$;
(iii) for binding of primers / nucleotides (to DNA strands);
(b) (i) doubling (of DNA) each cycle; but very low numbers to start with, so appears flat then exponential growth;
(ii) suggestion; with explanation e.g.:
nucleotides being used up; so less / nothing to make complementary chains;
primers used up; so cannot start
complementary chains;
enzymes losing activity / denatured; so no polymerisation of complementary strands;

