BIOLOGY
SECTION II
Time—1 hour and 30 minutes

Directions: Answer all questions.

Answers must be in essay form. Outline form is not acceptable. Labeled diagrams may be used to supplement discussion, but in no case will a diagram alone suffice. It is important that you read each question completely before you begin to write. Write all your answers on the pages following the questions in this booklet.

1. The human genome illustrates both continuity and change.

   (a) **Describe** the essential features of **two** of the procedures/techniques below. For **each** of the procedures/techniques you describe, **explain** how its application contributes to understanding genetics.

   - The use of a bacterial plasmid to clone and sequence a human gene
   - Polymerase chain reaction (PCR)
   - Restriction fragment length polymorphism (RFLP) analysis

   (b) All humans are nearly identical genetically in coding sequences and have many proteins that are identical in structure and function. Nevertheless, each human has a unique DNA fingerprint. **Explain** this apparent contradiction.

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Question 1

1. (a) • **Maximum 4 points for this part of the question (1 point earned for each bullet below, up to 4)**

(**Maximum 3 points, 1 for each bullet**)

Describe the use of **plasmid for cloning/sequencing** a human gene

- Cut plasmid with "restriction" enzyme
- Cut/isolate human sequence with the corresponding "restriction" enzyme
- Mix/anneal/ligate
- Introduce recombinant plasmid into bacteria
- Select recombinant bacteria (e.g., antibiotic resistance, fluorescence, reporter gene, etc.)
- Bacterial reproduction used to amplify the sequence
- Describe either degradative (Maxam-Gilbert) or dideoxy (Sanger) method to generate fragments
- Electrophoresis to separate fragments
- Read the sequence (automated method is OK)

(**Maximum 3 points, 1 for each bullet**)

Explain the contribution of this procedure

- Source of the DNA is immaterial to cloning
- Used to produce transgenic organisms
- Used to make human proteins (e.g., insulin, HGH)
- Understanding gene structure/regulation
- Comparative genomics
- Development of gene therapies
- Making gene library
- Amplifying a particular sequence

• **Maximum 4 points for this part of the question (1 point earned for each bullet below, up to 4)**

(**Maximum 3 points, 1 for each bullet**)

Describe PCR

- Heat to separate strands
- Add primers
- Cool to anneal
- Add polymerase and/or nucleotides
- Specification of heat stable (Taq) polymerase
- Description of thermocycling process
- Repetition of process

Question 1 (cont'd.)

(**Maximum 3 points, 1 for each bullet**)

Explain the contribution of this procedure

- Allows amplification of very small samples
- Replicates/amplifies a defined region
- Can be automated to allow for faster expansion of knowledge
- Can be used for forensics
- Can be used for diagnosis
- Evolutionary applications
- Other

- **Maximum 4 points for this part of the question (1 point earned for each bullet below, up to 4)**

(**Maximum 3 points, 1 for each bullet**)

Describe RFLP analysis

- DNA sample cut with "restriction" enzyme(s)
- Separation of fragments (electrophoresis)
- Description/elaboration of electrophoresis (charge/size/apparatus)
- Visualize fragments (probes, dyes, blots)
- Compare fragment sizes/mobility
- Compare single and double digests (two or more restriction enzymes)
- Compare individuals/species/organisms/tissue samples

(**Maximum 3 points, one for each bullet**)

Explain the contribution of RFLP analysis

- Trace RFLPs as genetic markers in families
- Diagnose disease/carriers/prenatal samples
- Prepare fingerprints (for forensics, etc.)
- Order fragments for physical mapping
- Compare genomes of different species/evolutionary relationships
- Locate the flanking regions of the gene/sequence
- Find mutations
- Individual bands can be used for further analysis
- Can determine presence of sequence without knowing its function

Question 1 (cont'd.)

1. (b) **Maximum 4 points — Explain the contradiction**

Sources of difference in DNA fingerprint
- Variation in non-coding material (introns, spacers, minisatellites, "junk," transposable elements)
- Point mutations, small deletions, SNPs (single NT polymorphisms)
- Variable number of tandem repeats (VNTRs/STRs)

Recognition of differences
- A small percentage difference of a very large genome results in a large number of nucleotide differences
- PCR-based fingerprinting: differences found by where primers anneal
- Variation in restriction enzyme cutting sites

Similarities among proteins
- Redundancy in the code for amino acids
- Neutral/silent mutation does not alter the function of the protein

Caution: No explanation points in (a) without an attempted description of procedure
Order of procedure points is not important if they are logical and accurate
No credit for mutations leading to new phenotypes
Codons specify amino acids (not proteins)