A few days ago (December 2, 1999) Nature published a report of the almost complete sequence of the euchromatic part of one arm of human chromosome 22. In recognition of this milestone in human molecular genetics, to which Oklahoma scientists contributed significantly, this examination is built around the theme of that report.

For each item, your tasks are highlighted in **bold face**.

I. Interpretation (35 points total)

A. Genetic and molecular maps of chromosome 22 were compared graphically:

"Figure 2 The relationship between physical and genetic distance. The sex-averaged genetic distances of Dib et al.36 were obtained from ftp://ftp.genethon.fr/pub/Gmap/Nature-1995/ and the cumulative intermarker distances for unambiguously ordered markers (in cM) were plotted against the positions of the microsatellite markers in the genomic sequence. It should be stressed that the y-axis does not represent the true genetic distance between distant markers but the sum of the local intermarker distances. The positions of selected
genetic markers are labeled. Grey regions" highlight regions with slopes distinct from the predominant slope.
Assignment:
What generalization about the relationship between genetic and molecular maps does the graph support (5 points)?

What is the likely explanation for the increased slope in the grey regions? (5 points)

B. Pseudogenes
Of the potentially coding regions identified, 19% were judged to be pseudogenes because the coding regions (similar to known coding regions) contained occasional stop codons or were in more than one reading frame. Of the pseudogenes, 82% “contained single blocks of homology and lacked the characteristic intron-exon structure of the putative parent gene.”

Assignment:
Suggest a likely explanation for the origin of these pseudogenes, accounting both for their apparent lack of introns and for their inability to function (10 points).
C. Immunoglobulin genes
Chromosome 22 contains genetic information used in the synthesis of immunoglobulin lambda light chains. The sequence data revealed 36 potentially active V lambda gene segments separate from seven J lambda-C lambda gene segments. The V regions are separated by large distances from the J regions and each J region is separated from its C region by smaller distances. Light chain mRNA consists of contiguous V, J and C regions.

Assignment:
What processes (n.b.: plural) account for the ability of cells of the immune system to synthesize lambda light chain mRNA from these clusters of gene segments (10 points) ?

D. Alpha satellite DNA
At one end of the sequenced region of chromosome 22 there are multiple copies of a 171 bp satellite, alpha satellite DNA. African green monkey kidney cells were transfected with alpha satellite DNA and cells with the transfected DNA inserted into chromosomes were selected. In subsequent cell divisions, many anaphase bridges (chromosomes strung between the two daughter cells at the final phase of cell division) and many lagging chromosomes were seen.

Assignment:
What function does the observation of anomalies in cell division suggest for alpha satellite DNA (5 points) ?
II. Basis for principles (30 points)

Chromosome 22 contains genes implicated in heritable human disease, such as the schizophrenia susceptibility locus and the spinocerebellar ataxia 10 locus.

Principle:
Many diverse alterations in coding or signaling sequences of a gene can cause that gene not to function optimally.

Assignment:
List ten (10) different ways by which changes in a gene’s DNA can keep a gene from functioning properly. For each, be sure to explain how the change you mention affects gene function (i.e. what process is impaired?). (3 points each)
III. Experimentation (35 points)

A. Among the identified genes on chromosome 22 is the gene (NAGA) for N-acetyl-\(\alpha\)-D-galactosaminidase, mutations in which are associated with Schindler and Kanzaki diseases. Understanding and treatment of many diseases is often facilitated by the availability of an animal model of the disease. The following assignments have as their aim the establishment of a mouse model for the disease.

Assignment:
Describe the steps that you would take in one strategy to isolate the mouse equivalent of the NAGA gene. Be as specific as you can with respect to materials and procedures. (10 points) 

B. Assume that you have isolated the mouse NAGA gene equivalent and made some mutations in the isolated gene.

Assignment:
Describe the steps that you would take to replace the wild type NAGA-equivalent gene in mice with your mutated version (10 points).
C. Mitochondria have their own chromosomes. Indeed, the human mitochondrial chromosome was completely sequenced long before chromosome 22 was partially sequenced. Facts and observations:
   1. By density gradient centrifugation, you isolated a “satellite” DNA, a DNA whose average buoyant density is quite different from the bulk of DNA of the non-human organism you are studying.
   2. You suspect that this DNA is mitochondrial DNA rather than highly repeated nuclear DNA.

Assignment:
Devise a test of your suspicion and describe the test. (7 points)

Describe the expected results of your test and how you would interpret them. (3 points)

D. DNA repair. Facts and observations:
   1. Multiple systems for repair of damage to DNA exist in most cells.
   2. You are studying a novel modified base that could arise by reaction of DNA with a new chemical.
   3. You have made a double-stranded circular DNA containing a single unit of this novel base at a known position in the sequence of the DNA. You can also make this molecule with specific nucleotides (or their bases) labeled.
   4. When transformed into target cells, the apparent DNA damage is repaired.
   5. Extracts of these cells are known to perform reactions of nucleotide excision repair (NER) and of base excision repair (BER).

Assignment:
Describe a test of whether the modified nucleotide is removed by NER. (3 points)
Describe the expected results of your test and how you would interpret them. (2 points)

IV. Extra (no credit)

Fit the following four DNA strands together in double helices:
GGAATTTAAACCGCTAAACTTTTTTATTATT
TCCGTTTAGCACCATTTCATATAAAAAAT
AAATAATAAAAAGTTTGGTGCTAAACCGGA
ATTTTTGTATATGAATAGCGGTTTAATTCC

Have a happy Holliday!