KENYATTA UNIVERSITY

UNIVERSITY EXAMINATIONS 2010/2011

INSTITUTIONAL BASED PROGRAMME (IBP) DECEMBER SESSION

EXAMINATION FOR THE DEGREE OF MASTER OF SCIENCE

SBT 535: PRINCIPLES OF GENETIC ANALYSIS

DATE: Thursday, 28th April, 2011 **TIME:** 9.00a.m. – 12.00noon

---

INSTRUCTIONS: Answer any **four** questions

1. Plasmid pNov345 if 7.0 kb in length and has single *Bgl*II, *Hind*lII, and *Bam*HI sites. You have cut the plasmid with *Bgl*II and inserted a 4.0 kb fragment into the site. From the data below, determine the restriction map of the resulting plasmid.

*Bg*lII	7.0	4.0		
*Hind*lII	6.0	5.0		
*Bam*HI	8.9	2.1		
*Bgl*II + *Hindl*II,	4.3	3.3	2.7	0.7
*Bgl*II + *Bam*HI	6.1	2.8	1.2	0.9
*Hind*lII + *Bam*HI	5.0	2.1	2.1	1.8

NB. Assume $\pi = 22/7$

2. Below are partial DNA sequences of four Cystic Fibrosis (CF) alleles. Answer the questions below based on these sequences.

GGG TTC TTT GTG GTG TTT TTA TCT GTG CTT CCC TAT GCA

GGG TTC TTG TGG TGT TTT TAT CTG TGC TTC CCT ATG CAC

GGG TTC TTT GTG GTG TTT GTA TCT GTG CTT CCC TAT GCA

GGG TTC TTT GTG GTG TTT TAA TCT GTG CTT CCC TAT GCA

(i) Below each sequence, write the mRNA sequence that would be produced during transcription.

(ii) Assuming the top allele is from an unaffected individual and the others are from affected individuals, what type of maturation is found in each of the lower three alleles? What major type of point mutation is **NOT** found in these sequences?

(iii) Under the mRNA sequence, write the protein produced during translation. Refer to the translation table in attached.

(iv) For each of the lower three alleles, describe how the protein produced is different from that of the unaffected individual.

(v) Do you think the differences in these protein sequences will affect the function of each of the proteins? Why or why not?

(vi) You are a scientist at an up-and-coming pharmaceutical company and have identified a region of the Cystic Fibrosis (CF) gene that you would like to target for gene therapy. First, though, you must amplify that region and test its behavior in vitro (outside of an organism). In the space below, describe a procedure that would allow you to amplify a specific 476 base pair region of your gene of interest. Include in the discussion/illustration the principle behind the process and the key enzyme involved.

3. Using illustration, describe the construction of the following

(a) Genomic library and list its advantages and disadvantages

(b) cDNA library and screening of the same.

4. Describe the following:

(a) The Sangers sequencing method

(b) The modification of the method stated in 4(a)

(c) The difference between the Sangers and the Maxam and Gilbert method.

(d) The working of an automated sequence (use an idealized representation).

5. (a) List the **four** nucleic acid blotting techniques and with illustration, describe a typical blotting technique.

(b) After blotting, it is common for one to purify the DNA, sequence and with the new sequence, do a BLAST to determine if a complementary sequence exist, get a related protein sequence, get a primer, search of alignment to the sequences in the NCBI data base. In relation to this define the following terms found in the NCBI data base

(i) BLASTp

(ii) BLASTx

(iii) BLASTn

(iv) TBLASTn