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MANIPAL INSTITUTE OF TECHNOLOGY

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VII SEMESTER B.TECH. BIOTECHNOLOGY

END SEMESTER EXAMINATIONS, NOV/DEC 2016

SUBJECT: GENOMICS & PROTEOMICS BIO 431

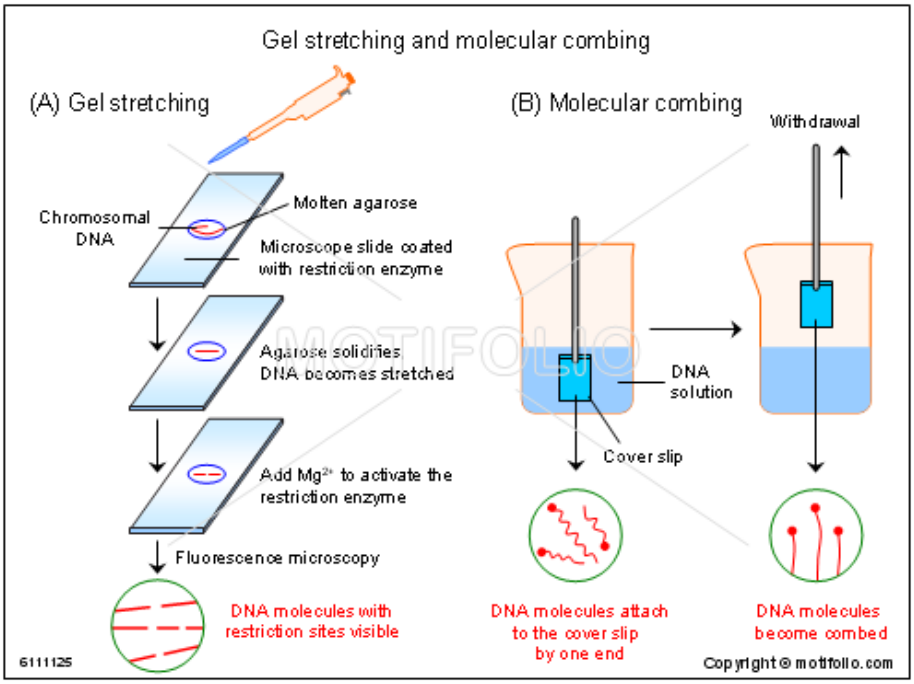
REVISED CREDIT SYSTEM
(06/12/2016)

Time: 3 Hours MAX. MARKS: 50

Instructions to Candidates:

- Answer **ANY FIVE FULL** questions.
- Missing data may be suitable assumed.

1A.	The rise of genomics and proteomics has generated knowledge which is revolutionizing the field of medical diagnostics and could yield a powerful arsenal of therapies that offer the promise of cures instead of just amelioration of symptoms. What analytical challenges proteomics approach possess compared to genomics?	**4**
1B.	An infamous sequencing company claimed that their read length is 100 Kbp in sequencing endangered species 'Tasmanian Tiger' using their contemporary NGS sequencing method. However scientific community raised doubts against this claim. Why scientific community questions this results?	**3**
1C.	Most of the well-characterized prokaryotic genomes consist of double-stranded DNA organized as a single circular chromosome 0.6-10 Mb in length and one or more circular plasmid species of 2 kb-1.7 Mb. The past few years, however, have revealed some major variations in genome organization. In addition, a recent accumulation of data has shown that the location and orientation of the genes and repeated sequences (including prophages and transposons) on and among these elements is not always random. Why it is essential to understand or study prokaryotic features and genome organization. What kind of clues we shall obtain from this?	**3**
2A.	Harvard researchers interested in sequencing extinct Eurasian wild aurochs, *Bos primigenius* used a technique called "Emulsion PCR" to create "hairy beads". This technique revolutionized the speed at which DNA sequencing was done. How	**5**

	emulsion PCR is different from standard PCR.	
2B.	How does this technique enable the Next-Generation sequencing technology used to sequence *Bos primigenius* DNA?	**3**
2C.	What interaction occurs between oligo's and bead substrate for creating hairy beads?	**2**
3A.	Genome editing, or genome editing with engineered nucleases (GEEN) is a type of genetic engineering in which DNA is inserted, deleted or replaced in the genome of a living organism using engineered nucleases, or "molecular scissors." Discuss the different genome editing tools.	**4**
3B.	Molecular combing is a technique in which single DNA molecules are bound by one or both ends to a surface and stretched in a uniform and parallel manner by a receding meniscus. This technique is gentle on the molecules, rapid, and easy to perform. Reliable, quantitative information for genome-wide studies can be obtained without the need for other techniques and a large number of accurate measurements can be made in a single experiment. How can we leverage this method to obtain sequence of methylated cytosine patterns in DNA? Explain it.  	**4**

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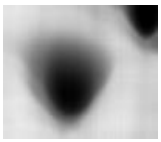
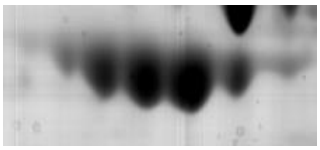
3C.	Nearly all proteins are made using only 20 standard amino acid building blocks. Two unusual genetically-encoded amino acids are selenocysteine and pyrrolysine. Pyrrolysine was discovered in 2002 at the active site of methyl-transferase enzyme from a methane-producing archeon, *Methanosarcina barkeri*. In what way, these two amino acids are different from hydroxylysine, methyllysine, and hypusine,	**2**
4A.	A long-held goal in nanopore sequencing has been to use a voltage-biased nanoscale pore in a membrane to measure the passage of a linear, single-stranded (ss) DNA or RNA molecule through that pore. With the development of enzyme-based methods that ratchet polynucleotides through the nanopore, nucleobase-by-nucleobase, measurements of changes in the current through the pore can now be decoded into a DNA sequence using an algorithm. Graphene has replaced membrane proteins. In what way, graphene can be considered as an alternative for membrane proteins. Give reasons.	**4**
4B.	The developers of Ion Torrent semiconductor sequencing have marketed it as a rapid, compact and economical sequencer that can be utilized in a large number of laboratories as a bench top machine with a read length of 400 bp. Is it advisable to use ion torrent to sequence human genome?	**3**
4C.	In a murder investigation, forensic scientists have collected the mitochondrial and nuclear DNA of the accused. Among the two DNA (mitochondrial and nuclear), which one you will be suggesting to determine the gender of the accused.	**3**
5A.	Identification of components of protein complexes by mass spectrometry and SDS-PAGE have become an important and powerful approach to understand cell biology. In both these techniques the accelerating force arise from an electric field acting on a charged molecule. The retarding force, which is the crucial parameter in selecting either mass spectrometry or SDS-PAGE. From researcher's perspective, detail the retarding force in these techniques.	**3**
5B.	Cerebrospinal fluid (CSF) is a clear, colorless body fluid found in the brain and spine. It is produced in the choroid plexuses of the ventricles of the brain. It acts as a cushion or buffer for the brain's cortex, providing basic mechanical and immunological protection to the brain inside the skull. CSF contains approximately 0.3% plasma proteins, or approximately 15 to 40 mg/dL, depending	**3**

	on sampling site. There are quantitative differences in the distributions of a number of proteins in the CSF. In general, globular proteins and albumin are in lower concentration in ventricular CSF compared to lumbar or cisternal fluid. The *IgG index* of cerebrospinal fluid is a measure of the immunoglobulin G content, and is elevated in multiple sclerosis. A B.Tech student is interested in separating CSF proteins. What technique would you suggest him/her for successful and cost effective protein separation? Rationalize your answer.	
5C.	Explain the working principle of Mass spectrometry and different separation techniques that can be combined with it.	**4**
6A.	Enolase, also known as phosphopyruvate hydratase, is a metalloenzyme responsible for the catalysis of the conversion of 2-phosphoglycerate (2-PG) to phosphoenolpyruvate (PEP), the ninth and penultimate step of glycolysis. The protein is represented by one spot when IEF was performed on pH 3-10 non-linear IPG strips (A), and by six spots when IEF was performed on pH 4-7 strips (B). Rationalize the difference between the images and reason behind it. (A)  (B) 	**3**
6B.	Explain and draw the schematic representation of two different work-flows involved in protein separation.	**4**
6C.	A protease (also called a peptidase or proteinase) is an enzyme that performs proteolysis, that is, begins protein catabolism by hydrolysis of the peptide bonds that link amino acids together in a polypeptide chain. A wide range of proteases have been utilized for protein digestion and separation techniques. Praveen has added proteases to his sample (comprised of Insulin and serpin family proteins), could not succeed. What can be the possible reason for malfunctioning of proteases?	**3**