所別:<u>生命科學系碩士班 分子與細胞生物組(一般生)</u> 科目:<u>分子生物學 共 5 頁 第 / 頁</u> \*請在試卷答案卷 (Pt) 內作答

Part I. Multiple Choice (單選題; 2.5 points each, total 75 points) Choose the one alternative that best answers the question.

1. A DNA fragment whose sequence is 5'CTAGAGTCTGGCTACA3' 3'GATCTCAGACCGATGT5'

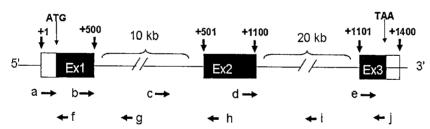
Its mRNA sequence from transcription is 5'CUAGAGUCUGGCUACA 3'. Which of the following is true?

- (a) The DNA sequence is mostly likely an intron sequence.
- (b) The DNA sequence is mostly likely an enhancer sequence.
- (c) The upper DNA strand is the coding strand.
- (d) The lower DNA strand is the sense strand.
- (e) Both strands can be the coding strand.
- (f) The upper DNA strand is the template strand.
- 2. Which of the following is incorrect?
- (a) Genomic libraries can be constructed using phages or bacterial plasmids.
- (b) Full sets of proteins encoded by genomes are studied in the field of proteomics.
- (c) Most of the genomes that have been sequenced to date are eukaryotes.
- (d) Current estimates are that there are 25,000-30,000 genes in the human genome.
- (e) Much of the DNA between genes consists of repetitive DNA.
- 3. A homeotic gene
- (a) turns on the housekeeping genes in all eukaryotic cells.
- (b) serves as a master control gene important in controlling the developmental fate of groups of cells.
- (c) produces a product that controls the transcription of other genes.
- (d) functions as a repressor for gene expression.
- (e) All of the choices are correct.
- 4. The lac operon
- (a) encodes enzymes that permit a cell to metabolize the milk sugar lactose.
- (b) is turned off as long as the repressor binds to the promoter.
- (c) is also controlled by the tryptophan operon.
- (d) uses activators to turns the operon on by binding to DNA.
- (e) is used to control both glucose and lactose concentrations in E. coli.
- 5. A student just isolated and purified mRNA from mouse liver tissues. After 10 times dilution, the absorbance measured at 260 nm is 0.25. What is the concentration of this mRNA sample before dilution?
- (a)  $2.5 \mu g/ml$  (b)  $25 \mu g/ml$  (c)  $75 \mu g/ml$  (d)  $100 \mu g/ml$  (e)  $125 \mu g/ml$
- 6. Which of the following statements about DNA replication is incorrect?
- (a) DNA replication in *E. coli* is semi-discontinuous.
- (b) Both leading and lagging strands are replicated in the 5' → 3' direction.
- (c) The lagging strand is replicated discontinuously as 1-2 kb Okazaki fragments.
- (d) Synthesis of Okazaki fragments in E. coli is initiated with DNA primers of 10-12 bases long.
- (e) Most eukaryotic and bacterial DNAs replicate bidirectionally.
- 7. Helicase is an enzyme that
- (a) degrades the RNA strand of an RNA-DNA hybrid.
- (b) unwinds a nucleic acid double helix.
- (c) degrades the DNA strand of an RNA-DNA hybrid.
- (d) introduces transient single-stranded breaks into substrate DNAs.
- (e) joins two double-stranded DNAs end to end.
- 8. Neither of your parents has cystic fibrosis, but your cousin, with whom you share a set of grandparents, does. How can you determine if you are a carrier for the disease?



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- (a) plasmid analysis (b) restriction fragment analysis (c) karyotyping (d) gene cloning (e) The only way to find out if you are a carrier is to reproduce and see if any of your children exhibit cystic fibrosis.
- 9. Which of the following statements about the genetic code is incorrect?
- (a) Among the total of 64 codons, 61 code for 20 different amino acids, showing a high degeneracy of the code.
- (b) The code is nonoverlapping: that is, each base is part of only one codon.
- (c) The degeneracy of the code can be accommodated by the so called 'wobble position'.
- (d) Each aminoacyl-tRNA can recognize and pair with only one codon.
- (e) none of the above
- 10. Which of the following is not true about cDNA library and cDNA cloning
- (a) A cDNA library is a set of clones representing as many as possible of the mRNAs in a given cell type.
- (b) cDNA fragments cloned into the M13 phage vectors can be recovered in single-stranded form.
- (c) Cosmids behave both as plasmids and as phages, thus they have been popular as cDNA cloning vectors.
- (d) Particular clones in a cDNA library can be detected by colony hybridization with antibodies if an expression vector is used.
- (e) Particular clones in a cDNA library can be detected by colony hybridization with radioactive DNA probes.
- 11. The genomic organization of a gene with 3 exons is shown below. The exons are shown as 3 boxes and their boundaries are denoted according to their location in mRNA. The size of the introns as well as location of start and stop codons are also shown on the top. If one wants to detect its expression level with quantitative real time RT-PCR, then which set of the primers is best suited for this purpose?
- (a). (a, g) (b). (c.i) (c). (b, i) (d). (c, h) (e). (d, j) (f). (a, j)



- 12. Following question 11, if this gene is expressed in *E. coli* without any post-translational modification, the most possible molecular weight of its protein is
- (a). 43 KD (b). 80 KD (c). 200 KD (d). 20 KD (e). 100 daltons (f). 60 daltons
- 13. Following question 11, if one uses primers a and h to amplify this gene, what will be the size of the amplicons amplified from genomic DNA and mRNA, respectively?

(c). (550 bp, 1.5 kb)

(d). (10.5 kb, 0 bp)

- (a). (10.5 kb, 550 bp) (b). (550 bp, 10.5 kb)
- (e). (31.5 kb, 550 bp) (f). (0, 0)
- 14. The sequence of a gene's mRNA can be summarized as below:

5'-uaguauaauaucggccaggAUGccg......cggguauucucauuuUAAggcagguucuua-3'
The start and stop codons are shown in **bold capital**. If you want to amplify the coding region of this gene with RT-PCR, which of the following primer sets will work for you?

- (a) 5'-cggccaggATGccg-3'/5'-taagaacctgccTTAaaatgagaa -3'
- (b) 5'-gccggtcCTAcggc -3'/5'-aaaATTccgtccaagaat -3'
- (c) 5'-eggeeaggATGccg-3'/5'-aaaATTccgtccaagaat -3'
- (d) 5'-cggccaggATGccg-3'/5'-ttctcatttTAAggcaggttctta-3'
- (e) 5'-gccggtcCTAcggc -3'/5'-taagaacctgccTTAaaatgagaa -3'
- (f) 5'-aaaATTccgtccaagaat -3'/5'-ttctcatttTAAggcaggttctta-3'



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- 15. The elements of a gene with 2 exons are listed as below:
  - (1). Exon 1; (2). promoter; (3). Exon 2; (4) intron; (5) terminator.

What is their most possible order in this gene when you are reading from downstream to upstream?

- (a). 5-3-4-1-2 (b). 1-3-2-4-5
- (c). 5-4-3-2-1
- (d). 2-1-4-3-5
- (e). 4-2-3-1-5

(f). 3-1-2-4-5

16. A student likes to make a fusion protein of GFP and p53 so he can trace the subcellular localization of p53 after the cells have been assaulted with UV radiation. He found that there is no compatible restriction site between his insert and GFP-containing vectors, so he has to make this construct by blunt end ligation. The insert was released from the vector with EcoRI digestion and its 5'end sequence is 5'-AATTCCGATGGAGGAC. The translational initiation codon is shown in bold. Which of the restriction sites in the GFP vector is suitable for making this fusion protein?

		MCS				
GFP TAC AAG	Xho I aga tct cga gct	caa gct t		<u>:(।</u> jca gtc ccg (	BamHI ggg atc c	
	Bg/ II	Hind III	EcoR I	Smal		

▼/▲: 5' and 3' cutting position

- (a). Xho I
- (b). BamH I
- (c). Hind III
- (d). Sma I
- (e). EcoR I

(f). Pst I

- 17. A contig of genomic DNA represents
- (a) A region of overlapping genes
- (b) A region of overlapping mRNA
- (c) A region of overlapping DNA fragments covering contiguous region of genome
- (d) A region of overlapping DNA fragments covering various regions of genome
- (e) The assembly of genes from expression sequence tags
- (f) The assembly of genes from non-expressed sequence tags
- 18. How is the 3' end of a mammalian gene mRNA generated?
- (a) Created by intrinsic terminator that causes termination of transcription
- (b) Created by Rho-dependent terminator that causes termination of transcription
- (c) Poly (A) added to the 3' end by Poly (A) polymerase
- (d) The 3' end of mRNA is where transcription stopped.
- (e) The 3' end of mRNA is created by cleavage at the termination site.
- (f) Poly (A) is added to the 3' end where termination happened.
- 19. The catalytic center active site of a spliceosome is formed by which snRNAs?
- (a) U1/U2 (b) U1/U3
- (c) U2/U3/U4
- (d) U3/U4
- (e) U6/U2 (f) U6/U4
- 20. One of the general transcription factors in the RNA polymerase II machinery contains ATPase, kinase, and helicase activity and also play important in elongation. This general factor is
- (a). TFIIA (b). TFIIB
- (c). TFIIH
- (d). TFIIJ

- 21. About TATA box and TBP, which of the following descriptions is false?
- (a). TATA box is found in all Pol II target gene promoters.
- (b). TBP binds to the minor groove of DNA
- (c). TBP positions RNA polymerase I to the correct initiation sites
- (d). TBP positions RNA polymerase II to the correct initiation sites
- (e). TBP positions RNA polymerase III to the correct initiation sites
- (f). TFIIDs containing different TAFs (TBP associated factors) could recognize different promoters.



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- 22. Which of the following regions in the mammalian genome can have the highest density of CpG doublets in general?
- (a). Inactivated transposon relics.
- (b). Active transposons
- (c). Regulatory regions of house-keeping genes
- (d). Regulatory regions of tissue-specific genes.
- (e). Insulators
- (f). Euchromatin.
- 23. About RNA polymerase, which of the following statements is true?
- (a). There is only one type of sigma factor in prokaryotes
- (b). Prokaryotic RNA polymerase can bind promoter specifically without sigma factor
- (c). Sigma factor is required for prokaryotic RNA polymerase binding to general DNA sequence
- (d). Prokaryotic RNA polymerase holoenzyme can not recognize promoter by itself
- (e). Eukaryotic RNA polymerase can not recognize promoter by itself
- (f.) All eukaryotic genes are transcribed by RNA polymerases II.
- 24. Which of the following combination of cis-elements best represents minimal Pol II target promoters? (upstream~downstream, and N represent number of bases in between)
- (a). Initiator-N<sub>20</sub>-TATA box-N<sub>24</sub>-DPE
- (b). Initiator-N20-TATA box
- (c). TATA box-N<sub>20</sub>-Initiator-N<sub>24</sub>-TATA box-DPE
- (d). TATA-N<sub>20</sub>-Initiator-N<sub>24</sub>-TATA-DPE
- (e). TATA-N<sub>20</sub>-Initiator-N<sub>24</sub>--DPE
- (f). TATA box-N<sub>20</sub>-Initiator
- 25. The function of insulators on gene expression is?
- (a). anti-initiation (b). anti-termination (c). block the effect of adjacent enhancer (d). enhance the spreading of heterochromatin (e). alternative initiator. (f). anti-attenuation
- 26. Which of the following factors mediates the repression effect of glucose on *lac* operon?.
- (a). Calcium ion (b). cAMP (c). cGMP (d). PI(3.4)P<sub>2</sub> (e). PI(3.5)P<sub>2</sub> (f).
- 27. One has used *pfu* DNA polymerase to amplify the CDS of a gene from product of reverse transcription. How can she clone this PCR product into a vector?
- (a). directly ligated into a blunted linear vector site.
- (b). modify the PCR product with klenow enzyme before ligated into a blunted linear vector site
- (c). modify the PCR product with ligase before ligated into a blunted linear vector site
- (d). directly ligated into a linear vector with 5'-T overhang
- (e). directly ligated into a linear vector with 3"-T overhang
- (f). modify the PCR product with polynucleotide kinase before ligated into a blunted linear vector site
- 28. Which of the following elements in a prokaryotic core promoter is the unwinding site of the promoter?
- (a). initiator (b). TATA box (c). -10 box (d). -35 box (e). Pribnow box (f). region between -1 and -35 boxes
- 29. The binding affinity of lamda phage CI repressor is strongest for?
- a).  $O_R 1$  (b).  $O_R 2$  (c).  $O_R 3$  (d).  $P_{RM}$  (e).  $P_{RE}$  (f).  $P_L$ .
- 30. Which of the following factors is required for CPSF to bind to the AAUAA site to mediate the cleavage of mRNA 3' end?
- (a). CPSF (b). CstF (c). PABP (d). poly (A) polymerase (e). CFI f. CFII



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#### Part II. Essay questions (total 25 points)

- 1. Please list the general transcription factors that contain TBP (TATA-binding protein)? (4 points)
- 2. Duchene Muscular Dystrophy (DMD) is a degenerative disorder of muscle, which affects 1 in 3500 of human male births and significantly less females. The genetic mutation (gene) causing this disorder was identified by traditional molecular biology methods, including genomic DNA hybridization/mapping, linkage analysis, and chromosome walking, without modern high-throughput screening technology. Can you recapitulate the way that this gene was discovered by using these approaches? (6 points)
- 3. What is the difference between reverse transcriptase PCR (RT-PCR) and standard PCR? Please outline their step-by-step methods. For what purpose would you use RT-PCR? (10 points)
- 4. Draw the cloverleaf tRNA structure and point out the important structural elements. (5 points)

