科目:生物化學(C)

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※ 注意:全部題目均請作答於試卷內之「非選擇題作答區」,請標明題號依序作答。

- 1. 胺基酸(amino acids)根據其極性(polarity)可分成三類,試問:
 - (1) 屬於高親水性(highly hydrophilic)的胺基酸有哪幾個?
 - (2) 屬於高疏水性(highly hydrophobic)的胺基酸有哪幾個?
 - (3) 屬於中極性(intermediate polarity)的胺基酸有哪幾個? (共 20 分)
- 2. 除了加熱之外,還有哪些方法會使蛋白變性(denaturation)?(5分)
- 二、以下題目請標明題號,詳細作答。
- 1. 選擇題 (複選, 每題2分)
 - (1) Which of the following statements about DNA double helix is **NOT** correct?
 - A. The two strands of a DNA double helix are antiparallel.
 - B. With B form DNA, each complete turn of the helix contains approximately 10 bp.
 - C. The bases of opposite strands are paired to each other by covalent bonds.
 - D. When DNA is denatured by heating, its absorbance in at 260 nm decreases.
 - E. The pairing of a purine (A or G) with a pyrimidine (T or C) within the helix is important for the integrity of the helix.
 - F. Most DNA-binding proteins bind DNA through the major groove.
 - G. At melting temperature (Tm), more than half of the DNA is denatured.
 - (2) Which of the following statements about RNA transcription is NOT correct?
 - A. Like DNA replication, transcription occurs in three phases, namely initiation, elongation, and termination.
 - B. A short RNA oligomer is required for initiation of RNA transcription.
 - C. The RNA is synthesized from the 5' to 3' direction.
 - D. The DNA strand directing mRNA synthesis is the coding strand.
 - E. Transcription usually begins at specific DNA sequences downstream of promoters.
 - F. The promoters of prokaryotic genes are highly conserved in the -10 and -35 sequences.
 - G. The RNA polymerase of eukaryotes is unable to recognize promoter sequences on its own.

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(3) Which of the following statements about the lac operon of *E. coli* is **NOT** correct?

- A. The lac operon of *E. coli* includes three genes which encode β-galactosidase, permease, and transacylase, respectively.
- B. The lac operon contains three promoters and an operator.
- C. When the bacterium is grown in a medium that contains only glucose, expression of the lac operon is completely repressed.
- D. In the absence of lactose, the lac I repressor binds to the operator and thereby blocks transcription of the lac operon.
- E. In the absence of glucose, the catabolite activator protein (CAP) binds to the operon and enhances transcription of the lac operon.
- F. Expression of the lac operon is induced in the presence of IPTG (isopropyl β -D-1-thiogalactopyranoside).
- G. When the lac operon is induced, β-galactosidase, permease, and transacylase are expressed as a polyprotein.
- (4) Which of the following statements about DNA replication and DNA polymerases of *E. coli* is **NOT** correct?
 - A. The process of DNA replication is semi-conservative.
 - B. The leading strand is synthesized continuously, while the lagging strand is replicated in a discontinuous manner.
 - C. In contrast to the leading strand, the lagging strand is synthesized in the 3' to 5' direction.
 - D. The initiation of DNA replication occurs at random sites around the genome.
 - E. DNA polymerase I is the main enzyme for the replication of DNA in *E. coli*.
 - F. DNA polymerase II is involved in DNA repair.
 - G. DNA polymerase III is an enzyme of multiple subunits.
- (5) Which of the following statements about protein translation is NOT correct?
 - A. The nucleotide sequence of the mRNA is read as a series of triplets.
 - B. Shine-Dalgarno sequence is complementary to the 3'-end of the 23S ribosomal RNA of *E. coli*.
 - C. Translation begins at an initiation codon, AUG, which is found at the 5'-end of an open reading frame (ORF).
 - D. In eukaryotes, ribosome finds the ORF in a way different from that of *E. coli*.
 - E. ORF ends with one of three different termination codons, UUA, UGA, or UAG.
 - F. The phenomenon that some amino acids are encoded by only one

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codon is described as "degeneracy".

- G. A frame-shift mutation may be caused by the insertion of a single base in the ORF or the 5'-untranslated region (UTR).
- (6) Which of the following statements about RNA processing in eukaryotes is NOT correct?
 - A. Once synthesized, the 5'-end of mRNA is capped by the addition of a 7-methyl guanosine residue via a 5'-3' condensation reaction.
 - B. The function of the cap is to protect the mRNA and also to help protein translation.
 - C. Introns are removed by RNA splicing.
 - D. RNA ligase is required for the joining of two exons.
 - E. All mRNAs contain at the 3'-end poly(A) tails, which are synthesized by poly(A) polymerase.
 - F. Poly(A) confers stability to the transcript, and is also required for translation to occur.
 - G. More than one form of mature mRNA may be generated from a primary transcript.

2. 問答題:

- (1) In regard to transcription by RNA polymerase II in eukaryotes, please describe in detail the process for initiation (5 分), and the characteristics of class II promoters (3 分).
- (2) How does "gene silencing" occur? (5 分)

= (30%)

- 1. 何謂 Photorespiration?C4 植物如何减低 Photorespiration。(10%)
- 2. 如何建立一生化代謝反應之途徑?(5%)若A → B 為其中之一反應, 且其需要一 enzyme 來催化其反應,設計一方法來純化此酵素(5%)? 酵素找到後設計一方法來找出其基因(5%)?基因找到後,您的下一步 實驗是什麼(5%)? (共20%)

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四、生化基本分析技術(共20分)

- 1. Electrophoretic techniques are common and useful for separation and purification of biological macromolecules, such as proteins and nucleic acids. What are the principles of electrophoresis? (2 pts) And what are the practical use (experimental component(s)) for protein and nucleic acid electrophoresis, respectively, in general? (2 pts).
- 2. Chromatographic techniques are applied for analysis and purification of biological molecules. Please briefly describe the principle of the following chromatographic techniques used in biological molecular analyses. (6 pts)
 - a. Thin-layer chromatography
 - b. Ion-exchange chromatography
 - c. Affinity chromatography
- 3. Viroids, exclusively bearing a circled, single-strand, non-coding RNA, are emerging pathogens to enable infecting a variety of plants. To study viroid pathogenesis in the corresponding host plants, several RNA-protein interactions are focusing, in particular, the identification of viroid RNA-binding proteins (from host/experimental plants). Please design a sequence/flow chart of the following experiments to explore the possibility of *Citrus Exocortis viroid* (CEVd; one species of viroids enables infecting both of citron and tomato) interaction with host DNA-dependent RNA polymerase II both in vivo and in vitro. (10 pts)

Note: The materials start from the CEVd infectious cDNA construct, 3-week old susceptible tomato seedlings, anti-Pol II (DNA-dependent RNA polymerase II) antibodies, recombinant Pol II protein, and all reagents for the following experiments.

- a. In vitro transcription of RNA (with remove DNA template)
- b. Mobility shift analysis
- c. Sucrose gradient analysis
- d. Mechanical inoculation of CEVd
- e. UV cross-linking
- f. Preparation of protein lysate from plant tissue(s)
- g. Mass spectrum analysis
- h. SDS-polyacrylamide gels electrophoresis
- i. Reverse transcriptase-polymerase chain reaction (RT-PCR).
- j. Specific (radio-)labelling of RNA at an internal site
- k. Immunoprecipitation