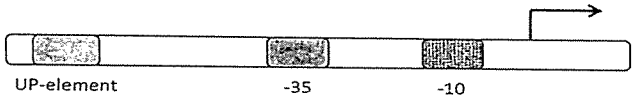
※ 注意：請於試卷內之「非選擇題作答區」依序作答，並應註明作答之大題及小題題號。

1. Describe the activities and functions of DNA polymerases in *Escherichia coli* and Human cells involved in DNA replication. (8 分)

2. The Nobel Prize in Chemistry 2015 awarded to Tomas Lindahl (base excision repair), Paul Modrich (mismatch repair) and Aziz Sancar (nucleotide excision repair) for their contributions to explain how the cell repairs its DNA and safeguards the genetic information. Please describe the DNA repair pathways and biological significance each Nobel laureates studied. (9 分)

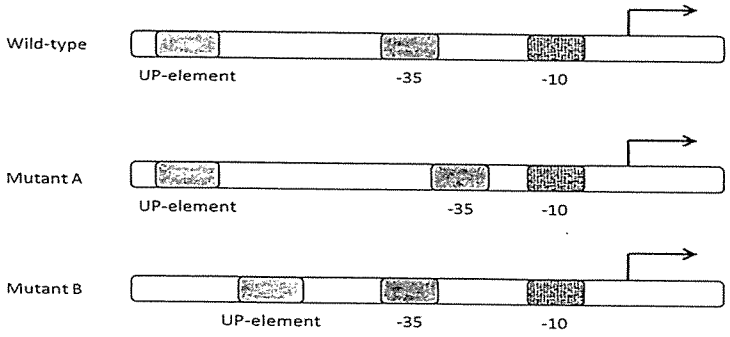
3. Double strand breaks (DSBs) in DNA if not repaired the consequence to the cell is disastrous. Please describe two mechanisms used to repair DSBs in cells.(8 分)

4. Below is a bacteria promoter with -10, -35 and UP elements.

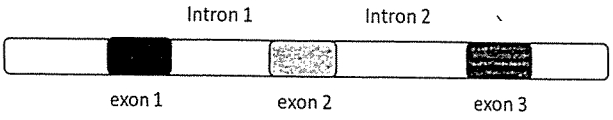



(1) Please describe what will be the best assay to determine the interactions between the RNA polymerase and the different elements on the promoter.(5 分)

(2) David wants to determine whether the distance between -10/-35 and -35/UP will affect the transcription efficiency of the promoter. He will construct two mutant promoters (Mutant A and Mutant B) and put a reporter gene downstream of the promoter. What will be the expected results? (10 分)




5. Below is a gene encoding three exons.




(1) Please describe the sequence features which are required in splicing. (5 分)

(2) If this gene undergoes alternative splicing, what will be generated? (5 分)

見背面

6. A gene called *dbt1* has just been found to account for the familial diabetes for two families. Researchers found that, in several patients from family A, both mRNA and protein of *dbt1* are barely detectible. On the other hand, the *dbt1* mRNA isolated from patients from family B is significantly longer than normal group, resulting in a much longer DBT1 protein.

(1) (6 分) Suppose both families are affected by single nucleotide change at different regions of the DBT1 gene, please propose the molecular mechanisms that cause the diabetes in the two families

(2) (4 分) Do you think the mutations in family A and family B would be genetic dominant or recessive? Explain why.

(3) (6 分) Continued from above, the researchers found that the mutation of DBT1 gene in family B is also present in family C, which does not develop diabetes. Interestingly, the length of *dbt1* mRNA and protein are normal in family C. The researchers found that members of family C bare an additional mutation in *dbt2*, which encodes an RNA binding protein. The sizes and levels of *dbt2* mRNA and protein isolated from family C are comparable to control group. Please propose the molecular mechanisms that accounts for prevention of diabetes in family C.

(4) (9 分) Continued from above, suppose you have obtained both normal and mutated versions of *dbt1* and *dbt2* genes from the three families, please design experiments that would test your proposed models.

7. 申論題

(1) 你在攻讀研究所時，指導老師給你一個基因作為論文題目，並告訴你說這個基因可能與腫瘤細胞的轉移有關，請你提出一個研究計畫，如何利用各種分子生物學技術在活體外(in vitro)、活體內(in vivo)以及臨床醫學應用等不同的層次來證明"該基因確實會影響腫瘤細胞的轉移" (5 分)

(2) 新一代定序儀(NGS, Next-Generation Sequencer)儼然已為分子生物學基因體研究中最有力的工具之一，請論述你對新一代定序儀的了解(如:原理或發展)、應用範圍、與其他定序方法的優缺點比較 (4 分)

(3) 你的指導老師希望你探討細胞經由藥物處理後，某 A 基因的表現量改變，而下面是你利用即時定量聚合酶鏈鎖反應(Real-Time Quantitative PCR)技術得到的結果

| | 加藥前 | | 加藥後 | |
|---|---|---|---|---|
| | Internal control | Gene A | Internal control | Gene A |
| 第一重複 Ct 值 | 18.9 | 24.1 | 21.3 | 31.0 |
| 第二重複 Ct 值 | 19.0 | 24.6 | 25.6 | 33.8 |
| 第三重複 Ct 值 | 19.1 | 24.8 | 26.6 | 34.2 |

請問:

a. 即時定量聚合酶鏈鎖反應常應用的原理有哪幾種？(2 分)

b. 根據此結果，A 基因在加了藥物後表現量如何改變？(3 分)

c. 這組數據你覺得有沒有問題？為什麼？(3 分)

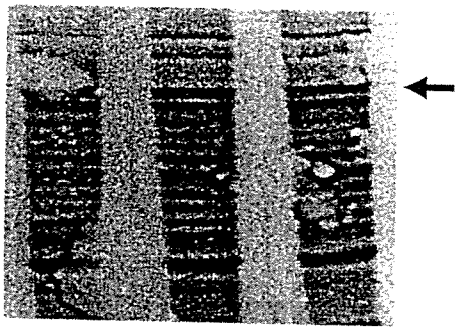
d. 除了利用即時定量聚合酶鏈鎖反應來分析 A 基因表現量改變外，還有什麼方法也可以分析 A 基因表現量的改變？(2 分)

接次頁

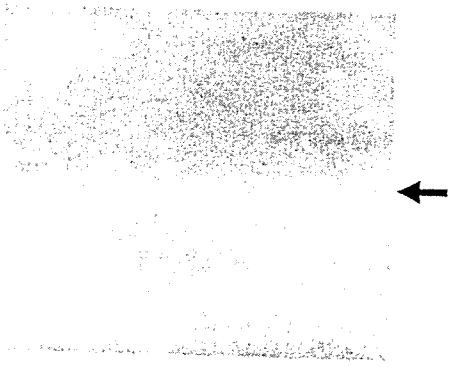
(4) 單一核苷酸多型性(SNP, Single Nucleotide Polymorphism)和基因突變如何區別？有哪些方法可以用來檢測單一核苷酸多型性及基因突變？(3 分)

(5) 西方點墨法(Western blot)是分子生物學上重要的技術，常用來分析蛋白質的表現，對於結果的判讀也是非常重要的一環，請你對下列 a. b. c. 三個西方點墨法的結果是否滿意？理由是？如不滿意，請問主要問題在哪邊？(箭號代表想要分析的蛋白質預期會出現訊號的質量大小處)(3 分)

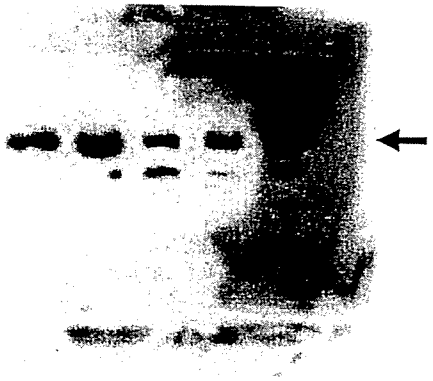
a.



b.



c.



試題隨卷繳回