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8. Human growth hormone (~22 KD) was one of the first recombinant therapeutic proteins produced in *E. coli* to be approved for human use in the world. However, its relatively short half-life in plasma, make the therapy of growth hormone inconvenient and expensive. How would you modify growth hormone to make it longer-acting? (10 points)

9. Fossil fuels, such as oil and gas are diminishing. Brazil produces large amount of ethanol from the fermentation of sucrose derived from sugarcane, and the United State produces ethanol from corn starch. Following table list the compositions of some lignocellulosic materials, please discuss: the basic principle and the advantages/disadvantages using the alternative fuel source from lignocelluloses. (10 points)

Table: Typical compositions of some lignocellulosic materials.

Raw material	Amount (%) of:		
	Lignin	Cellulose	Hemicellulose
Pine wood	27.8	44.0	26.0
Birch wood	19.5	40.0	39.0
Sugarcane bagasse	18.9	33.4	30.0
Rice straw	12.5	32.1	24.0
Cotton	12.5	32.1	24.0

10. The most effective and most often used microbial insecticide are the toxins synthesized by *Bacillus thurigiensis*. Following table list some properties of toxin from various strains of *B. thuringiensis*. Please answer: a. What is the mechanism of toxin to kill insect? b.What strategies could be applied to prevent the occurrence of the resistant insects? (10 points)

Table: Some properties of the insecticidal toxins from various strains of *B. thuringiensis*.

B. thuringiensis strain or subspecies	Protoxin size (kDa)	Target insects	Serotype
berliner	130–140	Lepidoptera	1
kurstaki KTO, HD-1	130–140	Lepidoptera	3
entomocidus 6.01	130–140	Lepidoptera	6
aizawai 7.29	130–140	Lepidoptera	7
aizawai IC 1	135	Lepidoptera, Diptera	7
kurstaki HD-1	71	Lepidoptera, Diptera	3
tenebrionis (san diego)	66–73	Coleoptera	8
morrisoni PG14	125–145	Diptera	8
israelensis	68	Diptera	14

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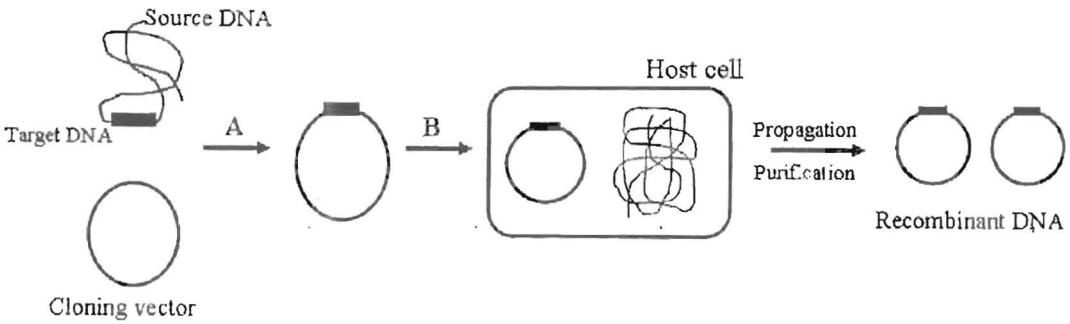
1. Following the diagram is the general procedures to manipulate the recombinant DNA in *E. coli*. Please answer the questions: (12 points)



a. What enzymes are required in **A** stage? What are their function and respective features? (4 points)

b. What did you need to do in the **B** stage? Please briefly describe the methods you used and their principles! (4 points)

c. What are the features to constitute a useful cloning vector for recombinant DNA technology? Please explain briefly (4 points)

2. What is the principle of error-prone PCR used in protein engineering? What are the possible factors to affect its performance? (8 points)

3. Please describe the process to develop an edible vaccine against cholera, caused by bacterium *Vibrio cholerae*. What are the advantages of edible vaccine as compared to traditional vaccine? (10 points)

4. Please briefly describe the procedures to create the transgenic plants overexpressing the human antibody such as IgG! (10 points)

5. What is nuclear cloning? Please given an example and explain how it works! (10 points)

6. What is DNA fingerprinting? How is it used to characterize traces of DNA in forensic samples? (10 points)

7. Please briefly explain how the HAT selection for hybridomas works! (10 points)

(背面仍有題目,請繼續作答)