

# UNIVERSITETET I OSLO

Det matematisk-naturvitenskapelige fakultet

Exam in: **MBV2020 Laboratory course in biochemistry and molecular biology**

Day of exam: **June 15, 2004**

Exam hours: **14:30-16:30 (2 hours)**

This examination paper consists of **3** pages.

Appendices: **None**

Permitted materials: **None**

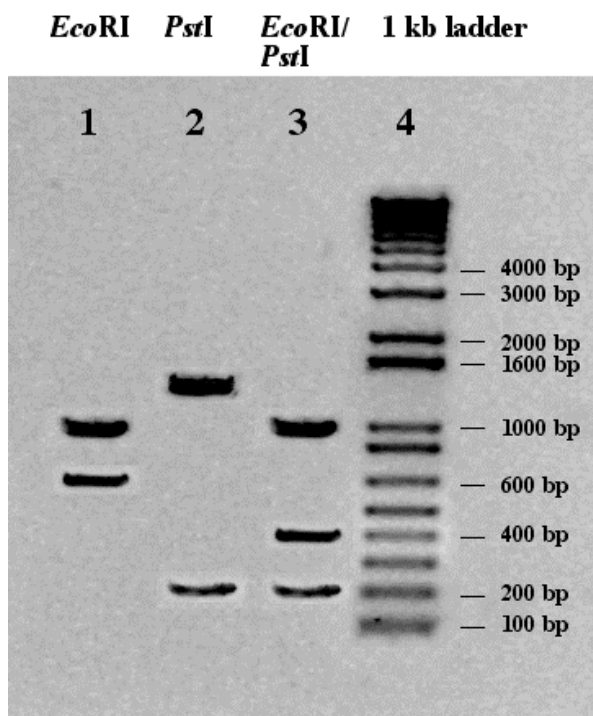
*Make sure that your copy of this examination paper is complete before answering.*

1. Explain very briefly (1-3 sentences) what function the following compounds had in the methods used in the MBV2020 course.

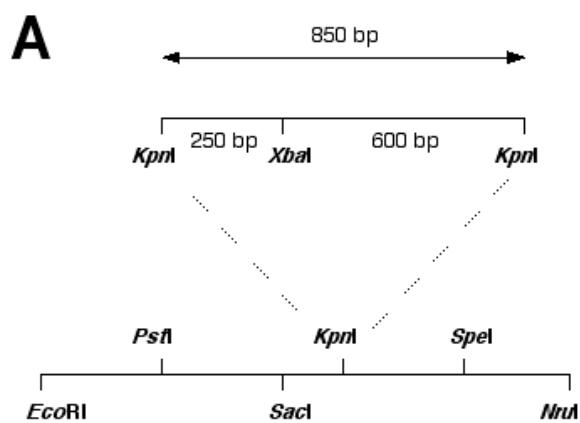
- a) Restriction enzymes
- b) Agarose
- c) Alcohol
- d) Gel loading buffer
- e) Ethidium bromide
- f) 1 kb ladder
- g) Ampicillin
- h) dNTPs (deoxynucleoside triphosphates)
- i) X-GAL
- j) IPTG

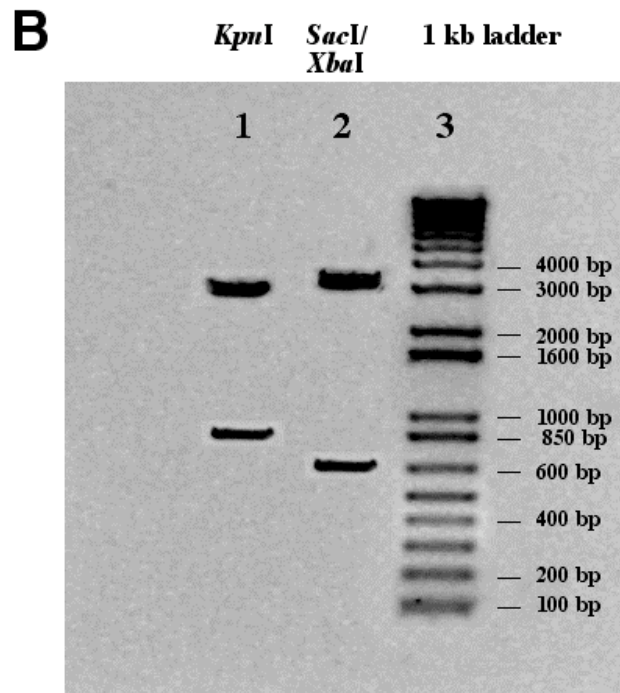
2. DNA digested with *EcoRI* and *PstI* was separated on an agarose gel as shown in the photo below. Please answer the following:

- a) What is the size of the original DNA molecule?
- b) Is the original DNA a linear or a circular molecule?
- c) Draw a restriction map of the DNA molecule indicating the approximate position of the restriction sites.



3. An 850 bp DNA fragment has been cloned into the *KpnI* site of a vector as shown in figure A below. To determine the orientation of the insert in the multiple cloning site, the final construct was digested with *SacI* and *XbaI* and separated on an agarose gel (figure B). Show in a drawing and explain in which orientation the insert is in the multiple cloning site.





4.

- Explain the principle of the polymerase chain reaction (PCR).
- A PCR was run with genomic DNA as template using two 20-mer primers and the following settings:

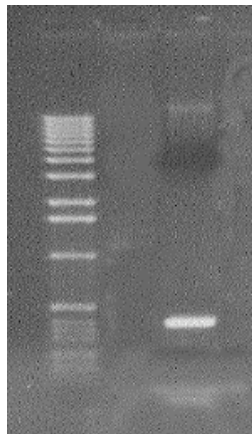
Denaturation: 1 min at 95°C

Annealing: 1 min at 60°C

Extension: 2 min at 72°C

30 cycles

The DNA fragment amplified under these conditions was visualized on an agarose gel (see photo below).



What could be expected to see on the gel when separately altering parameters as follows?

- a) The annealing temperature is lowered to 40°C (all other settings unchanged).
  - b) Primers are shortened to 10 nucleotides (all other parameters unchanged).
- Explain.