

Name \_\_\_\_\_

Your ID Number 3-159

**BIOCHEMISTRY 353, SPRING 2002 FIRST HOUR EXAM, Feb. 8, 2002**

Before you start, PRINT your name and record your ID number. Be sure to print your name at the top of each page. Notes of any kind are NOT permitted.

Confine your answers to the space provided. Only answers in the space provided will be graded. If you cross out an entire answer, you may write your new answer in the same amount of space on the back of the page. Be sure to clearly state in a box on the front of the page that the answer is on the back of the page, or your answer will not be seen by the grader.

**THIS EXAM IS 8 PAGES LONG. BEFORE STARTING CHECK THAT YOU HAVE ALL OF THE PAGES**

**DO NOT TURN THIS PAGE OVER UNTIL INSTRUCTED TO DO SO BY A PROCTOR**

Part I \_\_\_\_\_/30

Part II \_\_\_\_\_/20

Part III \_\_\_\_\_/15

Part IV \_\_\_\_\_/16

Part V \_\_\_\_\_/19

**TOTAL \_\_\_\_\_/100**

**Part I. TRUE-FALSE (3 points each)** Circle T for statements which are True and F for statements which are False. (**GRADED RIGHT MINUS WRONG**; This means that if you got 8 right and 1 wrong and left 1 blank you would get  $8-1 = 7 \times 3$  or 21 points.)

- T F 1. In the processing of mRNA precursors, the smallest intron is almost always removed first and the largest intron is almost always removed last.
- T F 2. In analyzing the base composition of most double-stranded DNAs,  $T+C=A+G$ ,  $C=G$  and  $T=A$ .
- T F 3. If you synthesize the 5'-capped form of an mRNA that is normally capped, divide the mRNA into equal portions, chemically decap half of it and inject the capped mRNA into one set of living eukaryotic cells and the decapped mRNA into a second similar set of cells, one day after injection the level of the decapped mRNA will probably be lower than the level of the capped mRNA.
- T F 4. A mutation that inactivated the 3'→5' exonuclease activity of *E. coli* DNA polymerase III would probably have no effect on the rate of mutations in newly replicated *E. coli* DNA.
- T F 5. Since there are 61 trinucleotide codons that specify amino acids, and 20 amino acids, three codons specify each amino acid.
- T F 6. A graduate student isolated a DNA polymerase from an organism living in an Antarctic glacier. This enzyme is very stable at low temperatures and is very unstable at high temperatures (assume this is true). When this enzyme was used as the DNA polymerase in a standard PCR reaction, the target DNA hardly increases in amount.
- T F 7. A key step in demonstrating that DNA is the hereditary material was the demonstration that heating a pathogenic *Pneumococcus* bacterium had no effect on the bacterium's ability to infect and kill mice.
- T F 8. EDTA chelates magnesium ions (assume this is true). If high levels of EDTA are added to a solution containing an unspliced type II self-splicing intron, the rate of self-splicing will be reduced.
- T F 9. If you very rapidly label the newly synthesized DNA in an *E. coli* containing only 10% of the normal level of *E. coli* DNA ligase, the fragments of newly synthesized DNA (hint: Okazaki fragments) isolated from the bacteria containing the reduced DNA ligase activity will be shorter than those in the bacteria containing normal DNA ligase levels.
- T F 10. The most common intranucleotide linkage in nucleic acids is a 2'-3' phosphodiester bond.

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**Part II. Multiple Choice Questions (20 points 4 points each) Circle the letters corresponding to ALL of the correct answers for each question.** (If 4 answers are correct and you circle one, that is not the correct answer). **Grading these questions: If you circle all of the correct answers you get all 4 points. If you make one error you lose 2 points (half credit). If you make two errors in a question you lose all 4 points (no credit).** (For example: If a question has three correct answers and you circle two of the three correct answers and no incorrect answers, you get two points [half credit]. If the question has three correct answers, and you circle two correct and one incorrect answer, you have made two errors, and get no points.)

1. At the same time that *E. coli* are infected with bacteriophage T2, the radioisotopes  $^{32}\text{P}$  and  $^{35}\text{S}$  are added. Which of the following events then occur.

- A. A new set of radioactively labeled proteins appears.
- B. The radioactive isotopes are not incorporated into RNA or protein
- C. A new class of labeled RNA is synthesized
- D. Newly synthesized radioactively labeled RNA becomes associated with ribosomes.
- E. Newly synthesized RNA becomes associated only with newly synthesized ribosomes.

2. Chemically synthesized oligonucleotides are commonly used:

- A. As primers in PCR reactions.
- B. As probes for Western blots (protein blots).
- C. As primers for sequencing DNA.
- D. To construct linkers containing restriction sites.
- E. To introduce mutations into cloned DNA molecules.

3. Which of these DNA sequences, if they were contained within a much longer double stranded DNA molecule, would be likely to be the recognition sequence of a restriction enzyme?

- |    |                              |                              |                              |                          |                          |
|----|------------------------------|------------------------------|------------------------------|--------------------------|--------------------------|
| A. | 5'-TAGCTA-3'<br>3'-ATCGAT-5' | B.                           | 5'-GACCAG-3'<br>3'-CTGGCC-5' | C.                       | 5'-AGTC-3'<br>3'-TCAG-5' |
|    | D.                           | 5'-GAATTC-3'<br>3'-CTTAAG-5' | E.                           | 5'-ACGT-3'<br>3'-TGCA-5' |                          |

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4. Assume that you have cloned the chromosomal copy of a gene that is present in a single copy in the genome of a turtle. The cloned gene contains one EcoRI site near its center. If you label the entire length of the cloned gene by nick translation, denature the labeled DNA to make it single stranded and probe a Southern blot of turtle DNA digested to completion with EcoRI, how many bands are you most likely to see on the autoradiogram? You should assume that the technical aspects of the experiment were carried out correctly. (Since there is only one correct answer, there is no partial credit for this question.)
- A. 0
  - B. 1
  - C. 2
  - D. 3
  - E. 4
5. In the standard and most common structure of DNA.
- A. The phosphate backbone of the two strands is linked in the center of the helix by bridging magnesium ions.
  - B. The DNA is in 2 helical chains coiled around a common axis running in opposite directions.
  - C. The nucleotides are on the inside of the helix and the sugar phosphate backbone is on the outside.
  - D. The planes of the nucleotide bases are perpendicular to the axis of the helix.
  - E. The helix rotates one complete turn approximately every 10-10.4 nucleotides.

**Part III. (15 points, 5 points each)** Based on your knowledge of splicing pathways, predict whether an RNA transcript made from each of the following hybrid genes will be spliced if incubated under conditions appropriate for splicing of a natural RNA precursor. If you think it will be spliced, briefly describe the mechanism. If you think it will not be spliced, briefly explain the reason for your conclusion. **Simple drawings should be included for full credit.**

1. The first exon and 5' half of the first intron of a gene encoding chicken ovomucoid joined by recombinant DNA techniques to the 3'-half of the second intron and 3'-exon of the mouse  $\beta$ -globin gene.



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3. In converting an mRNA into a double stranded by PCR, the mRNA is first copied into single stranded DNA using the enzyme \_\_\_\_\_.
4. These two nucleotides \_\_\_\_\_ forma a base pair that normally contains two hydrogen bonds.
5. The enzyme \_\_\_\_\_ requires a primer to copy DNA.
6. In dideoxy DNA sequencing which carbon on the sugar contains a hydrogen (H) instead of its normal OH group. \_\_\_\_\_
7. DNA sequenes are normally written 5' → 3' on the \_\_\_\_\_ strand.
8. The enzyme \_\_\_\_\_ is widely used in recombinant DNA research to covalently join blunt ended double stranded DNA fragments in the presence of ATP.

**V. Short Answer (19 points, 3 points each unless otherwise noted)** provide a brief biochemical explanation for each of the following observations or statements. Your answer should reflect the contents of this course. Only answers in the space provided will be graded, so think before you write. If you cross out an entire answer make a box indicating clearly that the answer is on the back, and use an equivalent amount of space on the back of the page. You do not have to fill in the entire space. If you know the answer, one or two short sentences or a simple diagram may be sufficient. If you keep writing, and write something that is incorrect, some credit will be deducted.

1. Bacteriophage T2 was used in the experiments performed by Hershey and Chase to help determine the nature of the genetic material. Bacteriophage M13 infects *E. coli* using a strategy that is different strategy than the one employed by T2. The M13 protein coat is removed in the inner membrane of the bacterial cell, where it is sequestered and subsequently used to help envelop the progeny phage DNA. Why would M13 have been much less well suited than T2 was for the experiments carried out by Hershey and Chase?

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2. (9 points, 3 points each) A solution containing active DNA polymerase I and the  $Mg^{+2}$  salts of dATP, dGTP, dCTP and dTTP is present. The molecules listed below are added to aliquots of this solution. State whether or not you would expect observe DNA synthesis and provide a brief (1-2 sentence) explanation of your reasoning. (Assume that incubation temperature and buffers were appropriate for testing for DNA synthesis.)
- A. A double-stranded closed circular negatively supercoiled DNA 3,000 nucleotides in length.
- B. A linear single-stranded DNA 3,000 nucleotides in length base paired near the middle of the DNA with a 50 nucleotide linear single-stranded RNA containing at its ends a 5'-hydroxyl group and a 3'-phosphate
- C. A single-stranded DNA 1,000 nucleotides in length that contains 5'-phosphate and 3'-hydroxyl groups at its ends that contains a sequence of 30 nucleotides that can perfectly base pair with a sequence near the center of the DNA.

