

Name: _____

The Genetic Code

2nd base in codon

		U	C	A	G	
1 st base in codon	U	UUU phe	UCU ser	UAU tyr	UGU cys	3 rd base in codon U C A G
		UUC phe	UCC ser	UAC tyr	UGC cys	
		UUA leu	UCA ser	UAA STOP	UGA STOP	
		UUG leu	UCG ser	UAG STOP	UGG trp	
	C	CUU leu	CCU pro	CAU his	CGU arg	U C A G
		CUC leu	CCC pro	CAC his	CGC arg	
		CUA leu	CCA pro	CAA gln	CGA arg	
		CUG leu	CCG pro	CAG gln	CGG arg	
	A	AUU ile	ACU thr	AAU asn	AGU ser	U C A G
		AUC ile	ACC thr	AAC asn	AGC ser	
		AUA ile	ACA thr	AAA lys	AGA arg	
		AUG met	ACG thr	AAG lys	AGG arg	
	G	GUU val	GCU ala	GAU asp	GGU gly	U C A G
		GUC val	GCC ala	GAC asp	GGC gly	
		GUA val	GCA ala	GAA glu	GGA gly	
		GUG val	GCG ala	GAG glu	GGG gly	

Name: _____

7.012 Exam Two -- 2006

Exam starts at 10:05 am and ends at 10:55 am.

There are 9 pages including this cover page & the genetic code.

Please write your name on each page.

Only writing on the **FRONT** of every page will be graded.
(You may use the backs, but only as scratch paper.)

Question 1 **24 pts**_____

Question 2 **11 pts**_____

Question 3 **24 pts**_____

Question 4 **25 pts**_____

Question 5 **16 pts**_____

TOTAL **out of 100**_____

Name: _____

1. (24 pts) You are working with a piece of DNA of the sequence:

5' -TATTGAGCTCCCGGAT-3'

3' -ATAACTCGAGGGCCTA-5'

(a, 5 pts) You cut the above piece of DNA with a restriction enzyme that recognizes the sequence 5'-GAGCTC-3' and cuts on the 3' side of the A within this sequence. Draw all products that result from this digestion. Make sure to draw the nucleotide sequence of both strands, and label all 5' and 3' ends.

(b, 5 pts) Now you ligate the DNA you produced in part **(a)** to the sequence below, which you have also cut with the same restriction enzyme. Draw the shortest DNA product that could form from ligating a piece of DNA from part **(a)** to a piece of DNA from part **(b)**. Make sure to draw the nucleotide sequence of both strands, and label all 5' and 3' ends.

5' -TAGAGCTCCGCAATG-3'

3' -ATCTCGAGGCGTTAC-5'

Name: _____

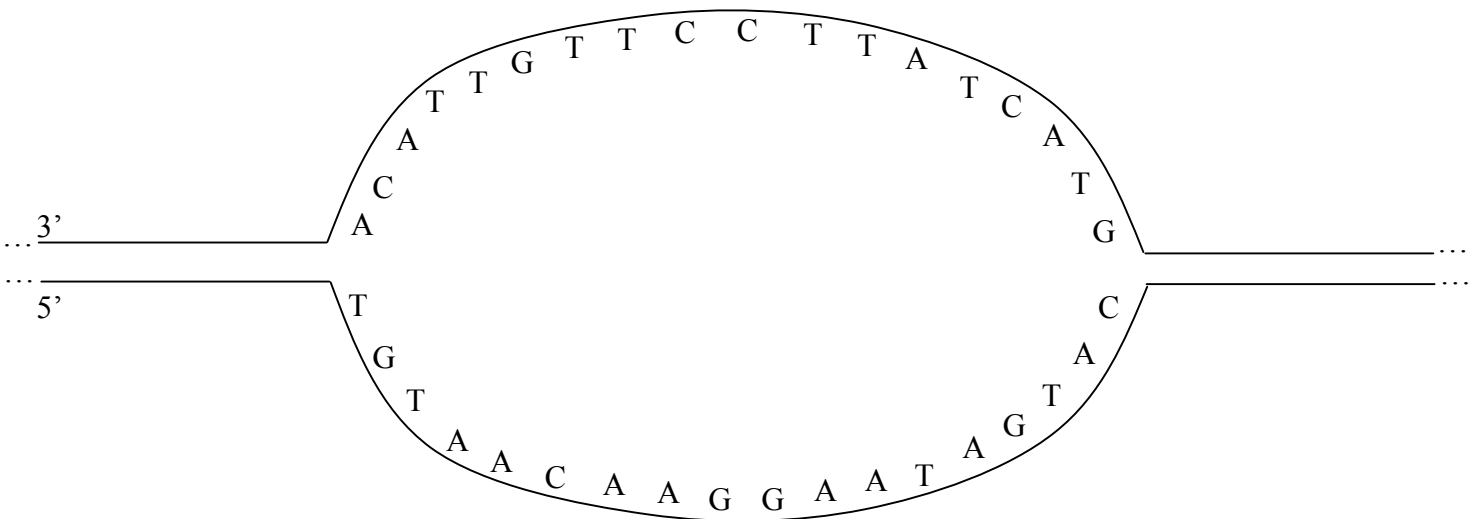
(c, 6 pts) Restriction enzymes have DNA binding domains. Name three other different types of proteins (that are not restriction enzymes) that have DNA binding domains.

Which reaction is catalyzed by each of the enzymes listed below? Answer by stating which specific type of bond is affected (e.g. "covalent" is not specific enough), and whether each enzyme catalyzes the formation or the breaking of that type of bond.

(d, 4 pts) the *T. aquaticus* "Taq" polymerase used in PCR

(e, 4 pts) a restriction enzyme

2. (11 pts) Below is shown a picture of a replication bubble.



(a, 3 pts) The primer 5'-CUU-3' is being used to replicate this piece of DNA. Would that primer anneal to the upper strand of DNA in the picture or the lower strand?

Name: _____

(b, 5 pts) Write what the product of DNA replication would be if the first five nucleotides had been added onto this primer by DNA polymerase. Label the 5' and 3' ends. Make sure to include the primer.

(c, 3 pts) Would that strand that you have drawn in part **(b)** be a leading strand or a lagging strand?

3. (24 pts) You are studying a very short protein-encoding gene whose sequence is shown below. The region of sequence shown is from the transcriptional start site to the transcriptional stop site. The gene's one small intron is shown for you in bold.

5' -CTACGTACTAGCTATTCC**GATCTATA**CTCGATCTAGTCGCATTCCGATTTCGATCGTAC-3'
3' -GATGCATGATCGATAAGG**CTAGATAT**GAGCTAGATCAGCGTAAGGCTAAGCTAGCATG-5'

(a, 4 pts) Which strand is used as a template in transcription, the upper strand or the lower strand?

(b, 4 pts) How many nucleotides long would the final processed mRNA made from this gene be (not including the 5' cap and the 3' polyA tail)?

(c, 4 pts) How many amino acids long would the protein product of this gene be?

(d, 4 pts) What is the sequence of the anticodon of the tRNA that would bind to the third codon of this mRNA as it was being translated? Make sure to label the 5' and 3' ends of the anticodon sequence.

Name: _____

(e, 4 pts) What is the amino acid that would be on the C-terminus end of the protein produced from this gene?

(f, 4 pts) Say that the C:G basepair that is underlined in the sequence above were changed to an A:T basepair to make a mutant form of this gene. Which kind of mutation would this be? Your choices are: a frameshift mutation, a nonsense mutation, a silent mutation, or a missense mutation.

4. (25 pts) You are interested in how a specific protein-encoding mouse gene named Hrt1 is regulated. Full activity of the Hrt1 gene product is present in heart cells, but no activity of this gene product is present in liver cells. You hypothesize that the Hrt1 gene product is regulated in one of the following ways (which are listed in no particular order):

- 1) whether the mRNA is translated or not
- 2) whether the protein product is stable or immediately completely degraded
- 3) whether the gene is transcribed or not
- 4) whether the protein product is phosphorylated or not
- 5) whether the pre-mRNA is spliced to include all 5 exons or only 4 of those exons

You prepare four samples from cells: all of the mature mRNAs in heart cells, all of the mature mRNAs in liver cells, all of the proteins in heart cells, all of the proteins in liver cells. You load each sample into a separate lane in a gel, and separate the components by size via gel electrophoresis. You then detect presence/absence of any band corresponding to Hrt1 RNA or protein.

(a, 5 pts) Towards which pole of the gel will the Hrt1 RNA run? Your choices are: positive, negative, neither, both, or inconclusive. Explain your answer.

Name: _____

(b, 5 pts) Towards which pole of the gel will the Hrt1 protein run? Your choices are: positive, negative, neither, both, or inconclusive. Explain your answer.

You perform the type of experiment described above, and detect any Hrt1 mRNA that is present in heart cells or in liver cells. At which level (of the five levels listed above) do you think the Hrt1 gene product is regulated? List all possible levels (i.e. #1 through #5) given that result.

(c, 5 pts)

RNA from heart cells:	a band is present
RNA from liver cells:	a band with the same intensity is present that ran the same distance in the gel

Name: _____

Now say that, INSTEAD, you perform the type of experiment described above, but detect any Hrt1 **protein** that is present in heart cells or in liver cells. At which level (of the five levels listed above) do you think the Hrt1 gene product is regulated? For each potential result, list **all** possible levels (i.e. #1 through #5) given that one result.

(d, 5 pts) Protein from heart cells: a band is present
Protein from liver cells: a band with the same intensity is present that ran a different distance in the gel

(e, 5 pts) Protein from heart cells: a band is present
Protein from liver cells: no band is present

5. (16 pts) You are studying a **mouse** gene named MtoA. You know that the protein product of this gene localizes to mitochondria in mouse cells. Your final goal is that you want to know whether mouse MtoA protein is capable of localizing to mitochondria if produced in **yeast** cells. You decide to make a plasmid that contains a gene that is a fusion of the mouse MtoA gene to the **jellyfish** GFP gene. You make the plasmid correctly in a test tube, transform it into ***E. coli***, select for ampicillin, and allow the *E. coli* to grow up into colonies so as to make many copies of the plasmid. You then extract the plasmid from *E. coli*, and then transform it into yeast. You allow the transformed yeast to grow up into colonies, and you then look at these yeast containing the plasmid under a fluorescence microscope, and see green fluorescence in the mitochondria.

(a, 4 pts) Answer YES or NO for each of the following organisms. Must your plasmid contain an origin of replication that works...

...in mice?

...in yeast?

...in jellyfish?

...in *E. coli*?

Name: _____

(b, 3 pts) Must your plasmid contain a promoter that works in mice? If yes, state which specific gene on the plasmid will be controlled by that promoter.

(c, 3 pts) Must your plasmid contain a promoter that works in yeast? If yes, state which specific gene on the plasmid will be controlled by that promoter.

(d, 3 pts) Must your plasmid contain a promoter that works in jellyfish? If yes, state which specific gene on the plasmid will be controlled by that promoter.

(e, 3 pts) Must your plasmid contain a promoter that works in *E. coli*? If yes, state which specific gene on the plasmid will be controlled by that promoter.