

Question 1 Translation

Here is a sequence (180 bases) from the baker yeast (*Saccharomyces cerevisiae*) genome. This sequence is located directly downstream from a promoter: it is a transcribed sequence. The sequence of the coding strand is given below: the mRNA produced by transcription has the same sequence, except for T replaced with U. There is no intron in this sequence.

1060

```
5' c a c c c g a a a c   g a c g t c g t a a   g t c c c g t t c a   g g g c c t t c g t   t e t c a c g g t a   t a a c c a t g a t
   c g g t g a c c g a   a c t t t c a g c t   g t c t c a t g g t   g c c a a c g t c a   a g c g a a g c g g   g g g t t g t t g t
   t g c g a g a a t g   t a t c c t t a g c   a t g a t g c t g g   c g g c g c c a c g   c a a a t t t t c t   g a g t g t a t t t   3'
```

1.1 What is the size in amino acids of the protein encoded by this sequence ?

_____ 27 amino acids _____

1.2 What is the size of the protein when the only mutation introduced into the sequence is at position 20 : A is replaced by T (20 a → t)

_____ 15 amino acids _____

1.3 What is the size of the protein when the only mutation introduced into the sequence is at position 56 : A is replaced by T (56 a → t)

_____ 17 amino acids _____

1.4 What is the size of the protein when the only mutation introduced into the sequence is at position 64 : T is replaced by A (64 t → a)

_____ 27 amino acids _____

1.5 What is the size of the protein when the only mutation introduced into the sequence is at position 82 : T is replaced by A (82 t → a)

_____ 8 amino acids _____

1.6 What is the size of the protein when the only mutation introduced into the sequence is at position 166 : a T is added during DNA replication by stuttering

caaattttct → caaat~~t~~ttct

_____ 27 amino acids _____

1.7

You want to PCR amplify the sequence given above. To do that you have to order 20 nucleotides long primers. Indicate the sequences of the primers you order to do the PCR :

Sequence of primer 1 : 5' _____ c a c c c g a a a c g a c g t c g t a a _____ 3'

Sequence of primer 2 : 5' _____ a a a t a c a c t c a g a a a a t t t g _____ 3'

Question 2

During translation, the energy cost of one round of elongation is

- A. 1 ATP
- B. 1 GTP
- C. 3 ATP
- D. 1 ATP + 2 GTP
- E. 2 ATP + 2 GTP

To attach an amino acid to the corresponding tRNA : the equivalent of 2 ATP are used.
 $\text{ATP} \rightarrow \text{AMP} + \text{Pi} + \text{Pi}$ represents the same energy cost as $2 \text{ATP} \rightarrow 2 \text{ADP} + 2 \text{Pi}$

During elongation each elongation factor (eEF-1 and eEF-2) hydrolyses one $\text{GTP} \rightarrow \text{GDP}$

Question 3

Amino acids are attached to

- a. the 3' end of tRNA
- b. the 5' end of tRNA

Question 4

What amino acid should be attached to a tRNA with the anticodon sequence 5'-UGA-3' ?

Base pairing must be anti-parallel :

on tRNA Anticodon 3' - A G U - 5'

on mRNA Codon 5' - U C A - 3' → Serine

		Second mRNA base					
		U	C	A	G		
First mRNA base (5' end of codon)	U	UUU } Phe UUC } UUA } Leu UUG }	UCU } UCC } Ser UCA } UCG }	UAU } Tyr UAC } UAA Stop UAG Stop	UGU } Cys UGC } UGA Stop UGG Trp	U C A G	Third mRNA base (3' end of codon)
	C	CUU } CUC } Leu CUA } CUG }	CCU } CCC } Pro CCA } CCG }	CAU } His CAC } CAA } Gln CAG }	CGU } CGC } Arg CGA } CGG }	U C A G	
	A	AUU } Ile AUC } AUA } AUG Met or start	ACU } ACC } Thr ACA } ACG }	AAU } Asn AAC } AAA } Lys AAG }	AGU } Ser AGC } AGA } Arg AGG }	U C A G	
	G	GUU } Val GUC } GUA } GUG }	GCU } GCC } Ala GCA } GCG }	GAU } Asp GAC } GAA } Glu GAG }	GGU } GGC } Gly GGA } GGG }	U C A G	

Question 5

When one of the enzymes linking amino acids to tRNA makes a mistake (e.g. an Alanine is linked to a tRNA with a 5' CCC 3' anti-codon assigned to Glycine), what is the consequence?

- a. the tRNA will not participate to translation because it will be unable to bind eEF-1.
- b. the tRNA will not participate to translation because it will be unable to bind eEF-2.
- c. the tRNA will not participate to translation because it will be rejected by the ribosome.
- d. the tRNA will participate to translation and an Alanine will be incorporated instead of a Glycine in one protein.

The 20 enzymes loading amino acids onto tRNA carry the entire responsibility for the accuracy of their job. There is no quality control downstream from these enzymes. The ribosome checks only the codon / anti-codon pairing, nothing else.