no start codon in transcription

Questions Transcription

November 2021

1) What would be the most likely effect of a mutation at the following locations in an *E. coli* gene?

A→T
A→G

a) -10 . . . doesn't bond
b) -20 → located between the consensus sequences of the E. coli promoter. A mutation here may not have any effect.
c) -35 affect the binding of sigma factor to the promoter. Transcription would probably be reduced or inhibited.
d) start site of transcription little effect on transcription. The position of start site relative to the promoter is more important than the sequence of the start site

see photo. 01.12.21 A-tr.just

2) The following sequence of nucleotides is found in a single-stranded DNA template. Assume the RNA polymerase proceeds along this template from the left to the right. → move from left to right.
a) Which end of the DNA template is the 5′ end? read from 3′→5′, synthesizes RNA strand in the 5′ to 3′ direction
b) Give the sequence of the RNA copied from this template and label its 5′ end 3′ ends.

→

3′ GACAATGTCCATGCCA 5′

5′ CUGUUACAGGUACGGU 3′

3) The following DNA nucleotides are found near the end of a bacterial transcription unit. Find the terminator in this sequence.

3′ --AGCATACAGCAGACCGATCTTGGTCTGAAAAAAAGCATACA--5′

stem loop stem

a) Mark the point at which transcription will terminate.
b) Is this terminator rho independent or rho dependent? hairpin. no. rho-independent
c) Draw a diagram of the RNA that will be transcribed from this DNA, including its nucleotide sequence and any secondary structures that form.

see photo. 5′ AGCAUACAG GAGAGCC GAUCUU GGUCUG

4) What protein associated with a transcription factor is common to all eukaryotic promoters? What is its function in transcription?

ad. TBP (TATA Box binding protein) in TFIID. It binds the TATA Box and positions the polymerase over the transcriptional start site.

TBP. it binds to the TATA Box promoter

what does it do.

nucleic acids generation

5) Compare and contrast transcription and replication. How are these processes similar and how are they different?

DNA→RNA | DNA→DNA

Cell division ↔ gene ~~expression~~ expression

one strand | both strands

any time | specific time

T occurs only once

6) The gene for muscular dystrophy is dystrophin. With 2'400'000 nucleotides it is the longest human gene known. Assuming a rate of 40 bases per second, how long does it take the polymerase to transcribe this gene? 1000 min

The full length protein contains 3685 amino acids. If you construct a cDNA clone with 200 bp for leader and trailer sequences, and you transfered it into cultured cells, how long would it take to be transcribed?

(3685×3+200)/40/60 = 4.69 min

15h.

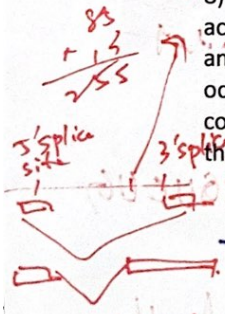
7) The doublestranded circular DNA molecule that forms the genome of the SV40 tumor virus can be denatured into singlestranded DNA molecules. Because the base composition of the two strands differs, the strands can be separated on the basis of their density into two strands designated W(atson) and C(rick). When each of the purified preparations of the single strands was mixed with mRNA from cells infected with the virus, hybrids were formed between the RNA and DNA. Closer analysis of these hybridizations showed that RNAs that hybridized with the W preparation were different from RNAs that hybridized with the C preparation. What does this tell you about the transcription templates for the different classes of RNAs?

see. photo.

simple | large T-ag protein

sequence similar to 3' splice site

8) In studying normal and mutant forms of a particular human enzyme, a geneticist came across a particularly interesting mutant form of the enzyme. The normal enzyme is 227 amino acids long, but the mutant form was 312 amino acids long. The extra 85 amino acids occurred as a block in the middle of the normal sequence. The inserted amino acids do not correspond in any way to the normal protein sequence. What are possible explanations for this phenomenon? How would you distinguish among them?



The 85 amino acids could have come from an unspliced intron due to mutation in a splice site sequence. Northern blot of RNA of mutant is 255 nts longer than the RNA of normal cell. Or from a mutation caused by insertion of DNA sequence. An intron sequence would be present in the genomic DNA of the wildtype, but an inserted sequence would not.