Transcription - Summary

starting input: DNA template (3' -> 5' strand)

initiation:

* recognition of the promoter region on the non-template DNA strand
* RNA polymerase binding to the promoter region

Elongation:

* separation of the double DNA strand
* RNA polymerase moves along DNA template strand 3' -> 5' direction
* binding of complementary nucleoside triphosphate to DNA template strand
* linking / condensation of nucleoside into RNA with the release of phosphate groups

Termination:

* recognition of terminator region by RNA polymerase
* RNA polymerase and mRNA detach from DNA

End / Output : mRNA

Translation - Summary

Starting input:

* binding of small subunit of ribosome to mRNA
* recognition of binding site on mRNA

Initiation:

* start coclon in mRNA, AUG
* binding of tRNA with amino acid methionine on mRNA
* addition of large subunit of the ribosome s.t. the tRNA is located on binding site

Elongation:

* tRNA located at A site and contains next amino acid
* tRNA located at P site contains polypeptide
* movement of polypeptide chain to the amino acid at A-site
* formation of a peptide bond and tRNA at p=side loses its polypeptide
* movement of large ribosome subunit s.t. the tRNA with no amino acid now occupies E-site
* tRNA with polypeptide occupies p-site

Termination:

* presence of one 3 stop codon (UAA, UAG or UGA) on the mRNA
* binding of release factor at the A-site
* release of the polypeptide release of tRNA, opening of ribosome in 2 subunits

DNA -transcription-> RNA -translation-> Protein

transcription:

* RNA synthesized using DNA template strand
* different types sythesized
* promoter
* terminator

Prokaryotes vs Eukaryotes

Prokaryotes:

* no nuclear membrane
* no physical separation between chromosomes and ribosomes
* often mRNA will bind to a ribosome and begin translation immediately
* only 1 type of RNA polymerase

Eukaryotes

* possess a nuclear membrane
* physical separation between chromosomes and ribosomes
* mRNA is subject to modifications before it exits to the cytoplasm through nuclear pores
* existance of pre mRNA
* contains coding information (exon)
* and non coding information (intron)
* pre mRNA: 5' ex | in | ex | in | ex 3'
* mRNA 5' (ex | ex | ex ) 3' (tail to 3' provides protection against degration)
* addition of capt o 5' acts as recognition for binding ribosome

Translation

* base sequence of mRNA converting ito amino acid sequence
* occurs in ribosomes located in cytoplasm

3 stages

1) initiation

2) elongation

3) termination

3.5 viruses

* small particles that cannot replicate by themselves
* consist of either DNA or RNA which is wrapped in a protein coat (capsid)
* sometimes has a membrane envelope
* lacks enzymes and ribosomes
* cannot self-replicate
* multiplies by invading a host cell
* ie: special case: bacteriophages (Bacteria virus)

3.6 Plasmids

* present in many prokaryotic cells (eg. bacteria)
* stand alone circular DNA molecule, distinct from the chromosome and can self-replicate
* contains a small number of genes that are not essential for the primary function of the cell, but beneficial for bacterial living in stressful environment (can counter antibiotic resistance)
* can manipulate plasmid to expand and amplify genes to produce target proteins

3.7 Restriction enzymes

* occurs naturally in bacteria
* provide protection against the action of foreign DNA (eg. bacteriophages)
* act by recognizing and cutting specific DNA sequences (typically between 4 and 8 nucleotides)
* over 3000 identified and characterized; over 600 commercially available

DNA manipulation

* objective/ purpose is to cut and/or paste DNA fragments from different sources
* DNA cutting -> involving restriction enzymes
* DNA pasting -> involves DNA ligase enzyme

Application

combine plasmid (circular DNA) with a piece of DNA fragment

restriction enzyme

plasmid with restriction enzyme fragment