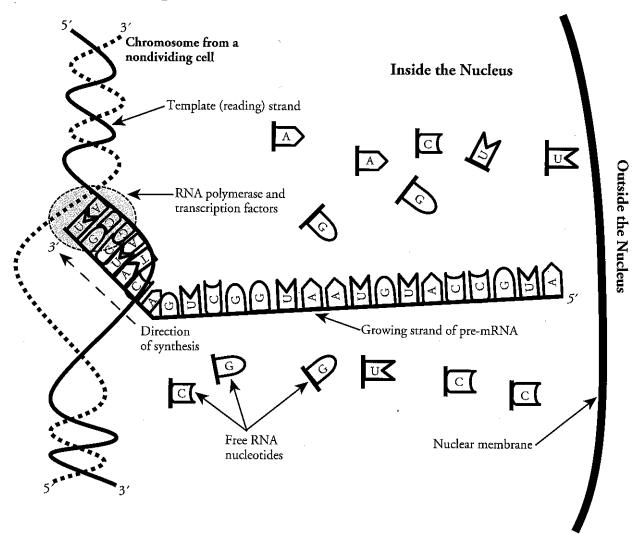
Gene Expression—Transcription

How is mRNA synthesized and what message does it carry?

Why?

DNA is often referred to as a genetic blueprint. In the same way that blueprints contain the instructions for construction of a building, the DNA found inside the nuclei of cells contains the instructions for assembling a living organism. The DNA blueprint carries its instructions in the form of genes. In most cases the genes direct the production of a polypeptide, from which other more complex proteins, such as enzymes or hormones, may be constructed. These polypeptides and other molecules run the organism's metabolism and, in multicellular organisms, dictate what each cell's job is. So, what is the language of these instructions and how are they read and decoded by the cellular organelles? This activity will focus on the decoding of genes in eukaryotes.

Model 1 – Transcription



- 1. Consider the eukaryotic cell in Model 1.
 - a. Where in the cell is the DNA found?

In the nucleus.

b. Where in the cell does transcription take place?

In the nucleus.

- 2. Refer to Model 1.
 - a. What polymer is synthesized during transcription?

Pre-mRNA.

b. What monomers are used to construct this polymer and where are they found? Free RNA nucleotides found within the nucleus (or nucleoplasm).

3. According to Model 1, what enzyme is required for transcription? (*Hint:* Think about how enzymes are named. What ending is used for enzyme names?)

RNA polymerase.

- 4. Refer to Model 1.
 - a. What is the base-pair rule for a DNA strand matching an RNA strand?

C to G, T to A and A (DNA) to U (RNA).

b. Compare this base-pair rule with that of two DNA strands.

DNA does not contain uracil, so T matches to A and A matches to T in DNA. However, in a DNA/RNA match, T bonds with A and A bonds with U.

5. Which strand of the DNA contains the "blueprint" for the pre-mRNA?

The template strand.

- 6. Consider Model 1.
 - a. In which direction is the DNA molecule read?

3' to 5'.

b. The DNA strand and pre-mRNA strand are anti-parallel. With this in mind label the 3' and 5' ends of the pre-mRNA strand in Model 1.

See Model 1.

c. In which direction is the pre-mRNA molecule constructed?

5' to 3'.

7 D C ...

7. Before printing presses were available, books had to be transcribed in order to share the information in them. Consider the definition of transcription and explain why the process in Model 1 is described using that word.

To transcribe is to make an exact copy. Transcription is making a copy of the information in DNA as RNA.

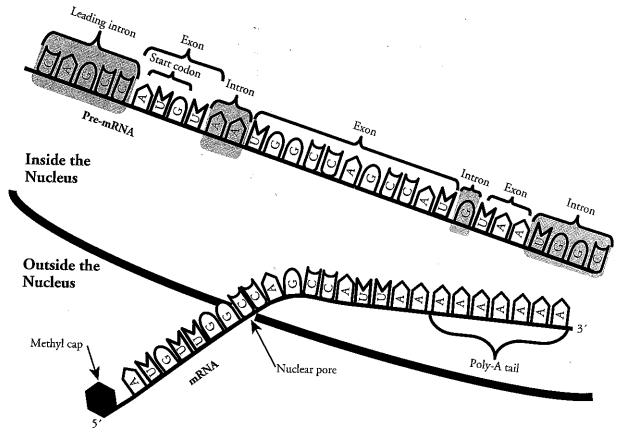
Read This!

In eukaryotes the enzyme RNA polymerase joins with several transcription factor proteins at the promoter, which is a special sequence of base pairs on the DNA template strand that signals the beginning of a gene. The transcription factor proteins, along with the RNA polymerase, is called the transcription initiation complex. This moves along the DNA template strand at about 40 base pairs per second producing pre-mRNA. When the RNA polymerase reaches the terminator sequence of base pairs on the DNA template strand, it completes the production of pre-mRNA and releases it into the nucleoplasm.

- 8. What parts make up the transcription initiation complex? RNA polymerase and transcription factors.
- 9. Where on the DNA strand does the transcription initiation complex form? *It forms on the promoter.*
- 10. Nearly all cells in an organism contain identical DNA, and each DNA strand may contain hundreds or thousands of individual genes. Is it likely that a cell would transcribe all the genes within its nucleus simultaneously? Justify your answer using complete sentences.
 - No, cells only transcribe genes as they are needed to make specific proteins.
- 11. Considering the many types of cells in a multicellular organism, and their different functions, is it likely that all cells transcribe all their genes at some point in their lifetime? Justify your answer using complete sentences.
 - No, certain genes will be switched on in specific cells and switched off in others depending on the location and function of the cell.



Model 2 - mRNA Processing



- 12. Compare the pre-mRNA to the mRNA leaving the nucleus in Model 1.
 - a. What has been removed from the pre-mRNA to make it into mRNA? *Introns.*
 - b. What has been added to the mRNA that was not present in the pre-mRNA, and where on the mRNA strand are the additional items located?
 - A methyl cap has been added to the 5' end of the mRNA and a poly-A tail has been added to the 3' end of the mRNA.
- 13. Identify the structure through which the mRNA leaves the nucleus. *The nuclear pore.*
- 14. The nucleotides on the mRNA will be "read" in the next step to producing a polypeptide. What sequence of bases indicates the starting point for the polypeptide "blueprint"? *AUG*.
- 15. The "m" in mRNA is short for "messenger." Why is this molecule called messenger RNA?

 The mRNA molecule carries the code for a polypeptide from the nucleus to the cytoplasm (or ribosome).

 Therefore, it is like a messenger.

Read This!

Introns are sections of pre-mRNA that are noncoding. That is, they don't provide useful information for the production of the polypeptide being synthesized. There is evidence that suggests these introns allow certain sections of DNA to code for different polypeptides when different sections are removed. The removal of specific sections is triggered by a signal response in the cell. The portions of the pre-mRNA that remain are called exons. The methyl cap (sometimes called the GTP cap or 5' cap) helps the mRNA molecule move through the nuclear pore and attach to a ribosome, its final destination. mRNA is a short-lived molecule. Once in the cytoplasm the mRNA will be subject to exonucleases that immediately start removing individual nucleotides from the 3' end of a nucleic acid. The individual mRNA nucleotides will then be free to be used again during the process of transcription.

- 16. The human genome contains about 25,000 genes and yet produces about 100,000 different polypeptides. Propose an explanation of how this is possible.
 - A single sequence of pre-mRNA can produce different mRNA sequences depending on what introns are removed.
- 17. Using the information in the *Read This!* box, develop a hypothesis to explain the advantage of the poly-A tail added to the 3' end of the mRNA.
 - It prevents the information-carrying part of the mRNA from being destroyed by the exonucleases before a polypeptide can be formed.



- 18. Different mRNA molecules can have poly-A tails of different lengths. Considering the purpose of adding the poly-A tail (from the previous question), why are some tails longer than others? Justify your answer using complete sentences.
 - An mRNA with a short tail will have a shorter lifespan. Proteins that are needed over long periods of time might come from long-tail mRNAs whereas proteins that are only needed briefly may come from short-tailed mRNAs.
 - 19. Summarize the steps of transcription.
 - Pre-mRNA is made from the template strand of DNA starting at the promoter by the transcription initation complex (RNA polymerase and transcription factors). When the RNA polymerase reaches the terminator, the pre-mRNA is released. Introns are removed and the methyl cap and poly-A tail are added to make mRNA.

Extension Questions

- 20. What type of biological molecule is an exonuclease?
 - Enzyme (protein).
- 21. Free nucleotides must be available in a cell's nucleus to produce mRNA strands. Where do these free nucleotides come from?
 - The free nucleotides come from the cytoplasm where older mRNA has been hydrolyzed by exonucleases.
- 22. Even though bacterial cells do not contain a nucleus, transcription occurs in a similar way to eukaryotic cells. How might biologists use transcription mechanisms to support the theory of evolution?
 - Scientists could use the fact that all species use the same nucleotides in their DNA and mRNA. All species use a promoter region to initiate transcription. All species use a similar RNA polymerase enzyme. Slight variations in the mechanisms could help connect species in an evolutionary phylogenic tree or cladogram.