BME 1532-CELL BIOLOGY

Gene Expression

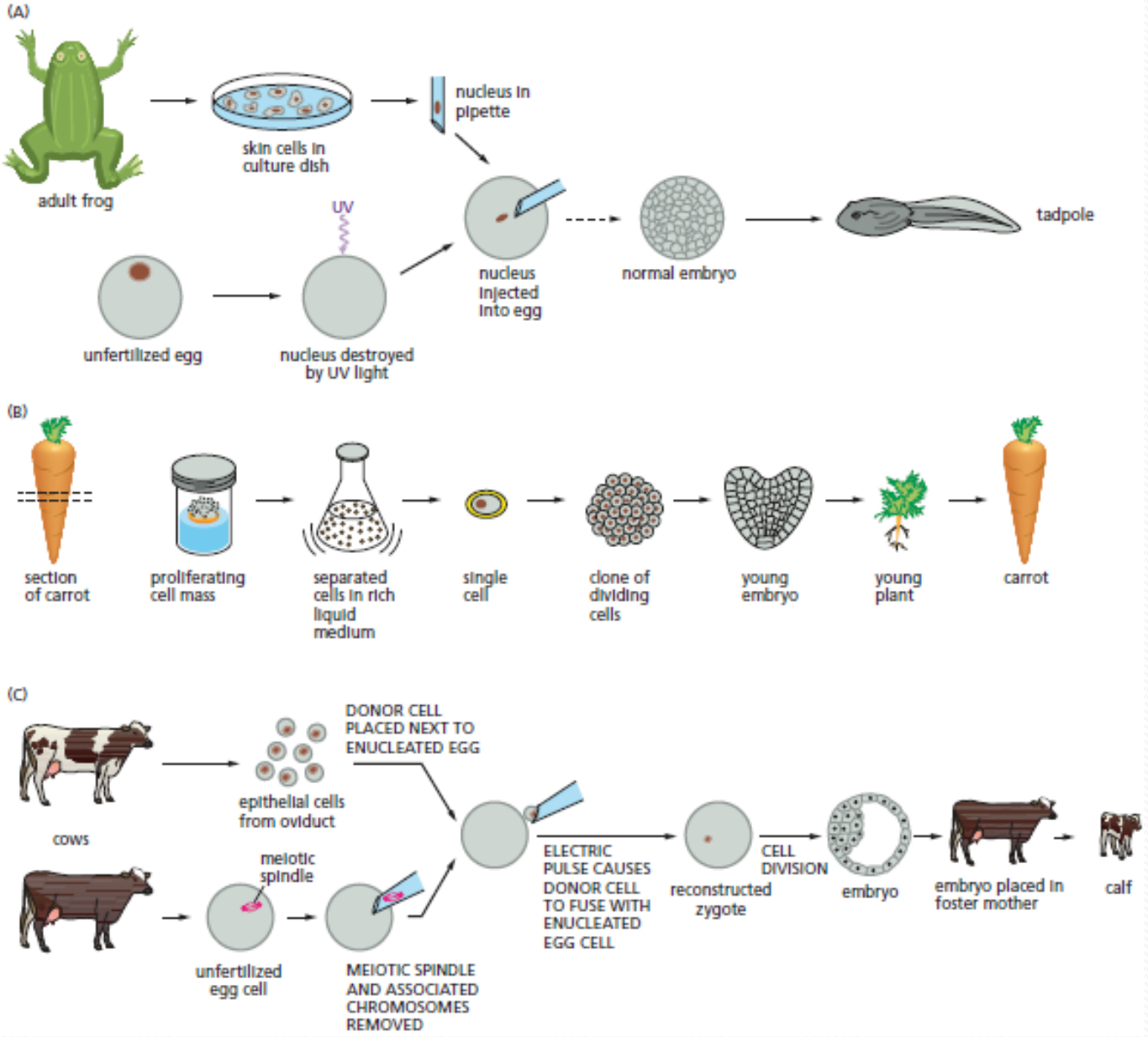
by Assist. Prof. Görke Gürel Peközer

Yıldız Technical University
Biomedical Engineering Department
Spring 2020

Last week on BME 1532

- Central Dogma
- Transcription
  - Promoters, Terminators,
    - Prokaryotic Transcription
      - Sigma factor
    - Eukaryotic Transcription
      - Regulatory DNA sequences, general transcription factors, RNA polymerase types
- mRNA proceesing
  - Capping
  - Polyadenylation
  - Splicing
    - Alternative Splicing
- Translation
  - Genetic code, codon, reading frame
  - tRNAs
  - Ribosomes

- Over the course of embryonic development, a fertilized egg cell gives rise to many cell types that differ dramatically in both structure and function.
- This occurs through cell differentiation which is achieved by changes in ***gene expression***.
- The evidence that cells have the ability to change which genes they express without altering the nucleotide sequence of their DNA comes from experiments in which the genome from a differentiated cell is made to direct the development of a complete organism.
- If the chromosomes of the differentiated cell were altered irreversibly during development, they would not be able to accomplish this.
- DNA in specialized cell types of multicellular organisms still contains the entire set of instructions needed to form a whole organism. The various cell types of an organism therefore differ not because they contain different genes, but because they express them differently.


(A)
adult frog
skin cells in culture dish
nucleus in pipette
UV
unfertilized egg
nucleus destroyed by UV light
nucleus injected into egg
normal embryo
tadpole
(B)
section of carrot
proliferating cell mass
separated cells in rich liquid medium
single cell
clone of dividing cells
young embryo
young plant
carrot
(C)
cows
epithelial cells from oviduct
DONOR CELL PLACED NEXT TO ENUCLEATED EGG
meiotic spindle
unfertilized egg cell
MEIOTIC SPINDLE AND ASSOCIATED CHROMOSOMES REMOVED
ELECTRIC PULSE CAUSES DONOR CELL TO FUSE WITH ENUCLEATED EGG CELL
reconstructed zygote
CELL DIVISION
embryo
embryo placed in foster mother
calf

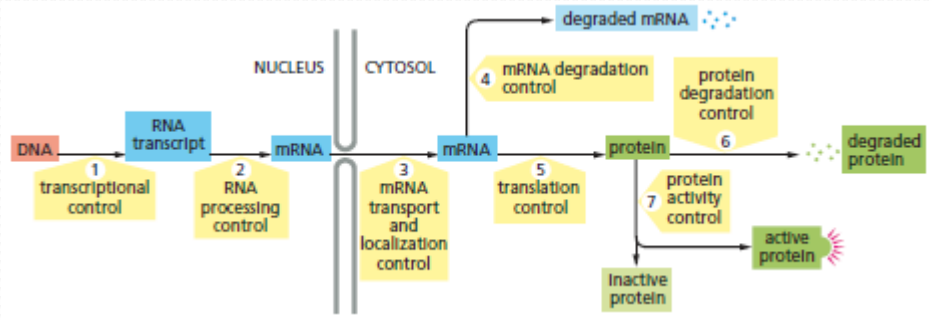
- Nearly all the cells of a multicellular organism contain the same genome.
- However, the differences between an information-processing nerve cell and a blood-filtering kidney cell, for example, are so extreme that it is difficult to imagine that the two cells contain the same DNA.
- In mammals, hundreds of different cell types carry out a range of specialized functions that depend upon genes that are switched on in that cell type but not in most others: for example, the β cells of the pancreas make the protein hormone insulin, while the α cells of the pancreas make the hormone glucagon; the B lymphocytes of the immune system make antibodies, while developing red blood cells make the oxygen-transport protein hemoglobin.
- The differences between a neuron, a white blood cell, a pancreatic β cell, and a red blood cell depend upon the precise control of gene expression.
- A typical differentiated cell expresses only about half the genes in its total repertoire.

- It is the expression of a different collection of genes in each cell type that causes the large variations seen in the size, shape, behavior, and function of differentiated cells.
- The extent of the differences in gene expression between different cell types can be also investigated by comparing the protein composition of cells in liver, heart, brain, and so on.
- These investigations reveal that many proteins are common to all the cells of a multicellular organism. These ***housekeeping proteins*** expressed from ***housekeeping genes*** include, for example, the structural proteins of chromosomes, RNA polymerases, DNA repair enzymes, ribosomal proteins, enzymes involved in glycolysis and other basic metabolic processes, and many of the proteins that form the cytoskeleton.
- In addition, each different cell type also produces specialized proteins that are responsible for the cell's distinctive properties.

- Besides, requirements for some gene products change over time.
- For example, the need for enzymes in certain metabolic pathways may increase or decrease as food sources change or are depleted.
- Or the specialized cells in a multicellular organism can alter their patterns of gene expression in response to extracellular cues.
- For example, if a liver cell is exposed to the steroid hormone cortisol, the production of several proteins is dramatically increased. When the hormone is no longer present, the production of these proteins returns to its resting level.
- Other cell types respond to cortisol differently, while some other cell types do not respond to cortisol at all.
- The fact that different cell types often respond in different ways to the same extracellular signal contributes to the specialization that gives each cell type its distinctive character.

Control of Gene Expression

- The control of gene expression is achieved at different levels since there are many steps in the pathway leading from DNA to protein, and all of them can in principle be regulated.
- Thus a cell can control the proteins it contains by;
  1. controlling when and how often a given gene is transcribed,
  2. controlling how an RNA transcript is spliced or otherwise processed,
  3. selecting which mRNAs are exported from the nucleus to the cytosol,
  4. regulating how quickly certain mRNA molecules are degraded,
  5. selecting which mRNAs are translated into protein by ribosomes, or
  6. regulating how rapidly specific proteins are destroyed after they have been made;
  7. regulating the activity of individual proteins



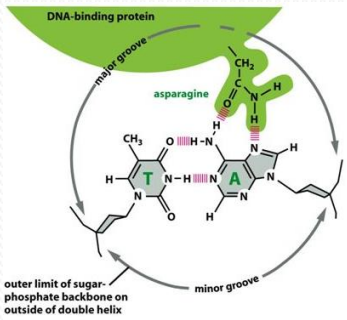
Transcriptional Control of Gene Expression

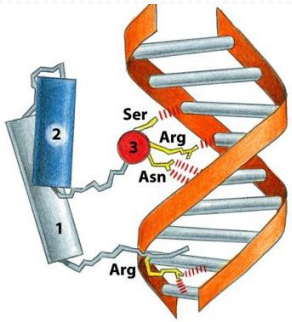
- For most genes, the control of transcription is paramount.
- This makes sense because only transcriptional control can ensure that no unnecessary intermediates are synthesized.
- Until 50 years ago, the idea that genes could be switched on and off was revolutionary.
- This concept was a major advance, and it came originally from studies of how *E. coli* bacteria adapt to changes in the composition of their growth medium.
- Many of the same principles apply to eukaryotic cells. However, of gene regulation in higher organisms is enormously complex.

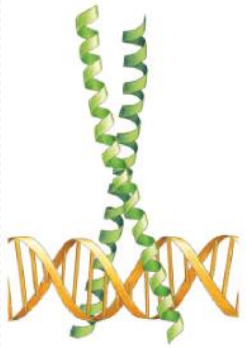
Regulatory DNA sequences

- Control of transcription is usually exerted at the step at which the process is initiated.
- We saw that the promoter region of a gene binds the enzyme *RNA polymerase* and correctly orients the enzyme to begin its task of making an RNA copy of the gene.
- The promoters of both bacterial and eukaryotic genes include a *transcription initiation site*, where RNA synthesis begins, plus a sequence of approximately 50 nucleotide pairs that extends upstream from the initiation site.
- This upstream region contains sites that are required for the RNA polymerase to recognize the *promoter*, although they do not bind to RNA polymerase directly.
- Instead, these sequences contain recognition sites for proteins that associate with the active polymerase—sigma factor in bacteria or the general transcription factors in eukaryotes.
- In addition to the promoter, nearly all genes, whether bacterial or eukaryotic, have ***regulatory DNA sequences*** that are used to switch the gene on or off.

- Regulatory DNA sequences must be recognized by proteins called transcription regulators to exert their effects.
- It is the binding of a transcription regulator to a regulatory DNA sequence that acts as the switch to control transcription.
- Regulatory proteins that recognize a specific nucleotide sequence because the surface of the protein fits tightly against the surface features of the DNA double helix in that region.
- Because these surface features will vary depending on the nucleotide sequence, different DNA-binding proteins will recognize different nucleotide sequences.
- In most cases, the protein inserts into the major groove of the DNA helix and makes a series of intimate molecular contacts with the nucleotide pairs within the groove.





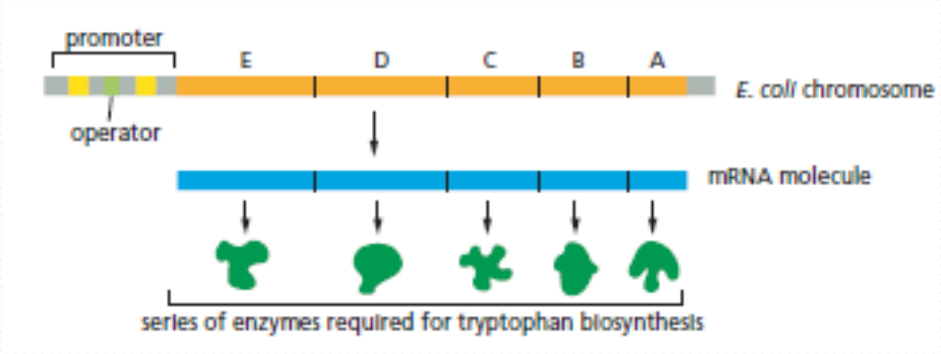


Prokaryotic Gene Regulation

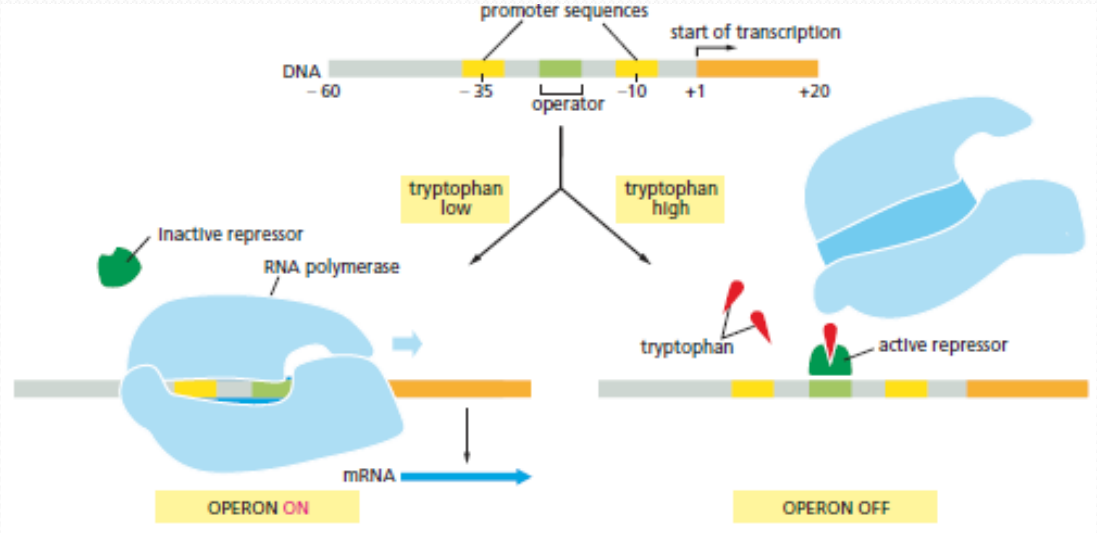
- The simplest and best understood examples of gene regulation occur in bacteria.
- The genome of the bacterium *E. coli* consists of a single circular DNA molecule of about 4.6×10^6 nucleotide pairs. This DNA encodes approximately 4300 proteins, although only a fraction of these are made at any one time. Bacteria regulate the expression of many of their genes according to the food sources that are available in the environment.
- For example, in *E. coli*, 5 genes code for enzymes that manufacture the amino acid tryptophan.
- These genes are arranged in a cluster on the chromosome and are transcribed from a single promoter as one long mRNA molecule; such coordinately transcribed clusters are called ***operons***.
- Although operons are common in bacteria, they are rare in eukaryotes, where genes are transcribed and regulated individually.

Transcriptional Repressors: Trp Operon

- When tryptophan concentrations are low, the operon is transcribed; the resulting mRNA is translated to produce a full set of biosynthetic enzymes, which work in tandem to synthesize tryptophan.
- When tryptophan is abundant, the amino acid is imported into the cell and shuts down production of the enzymes, which are no longer needed.
- Within the operon's promoter is a short DNA sequence, called the ***operator***, that is recognized by a transcription regulator.
- When this regulator binds to the *operator*, it blocks access of RNA polymerase to the promoter, preventing transcription of the operon and production of the tryptophan-producing enzymes.
- The transcription regulator is known as the *tryptophan repressor*, and it is controlled in an ingenious way: the repressor can bind to DNA only if it has also bound several molecules of tryptophan.



- The binding of tryptophan causes a subtle change in its three-dimensional structure so that the protein can bind to the operator sequence.
- When the concentration of free tryptophan in the bacterium drops, the repressor no longer binds to DNA, and the tryptophan operon is transcribed.
- The repressor is thus a simple device that switches production of a set of biosynthetic enzymes off according to the availability of the end product of the pathway that the enzymes catalyze.

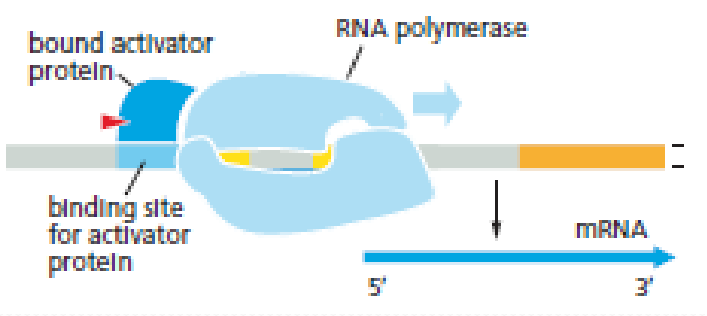


Transcriptional Activators

- The tryptophan repressor, as its name suggests, is a transcriptional repressor protein: in its active form, it switches genes off, or *represses* them.
- Some bacterial transcription regulators do the opposite: they switch genes on, or *activate* them.
- These ***transcriptional activator proteins*** work on promoters that—in contrast to the promoter for the tryptophan operon—are only not able to bind and position RNA polymerase on their own.
- These poorly functioning promoters can be made fully functional by activator proteins that bind nearby and contact the RNA polymerase to help it initiate transcription.

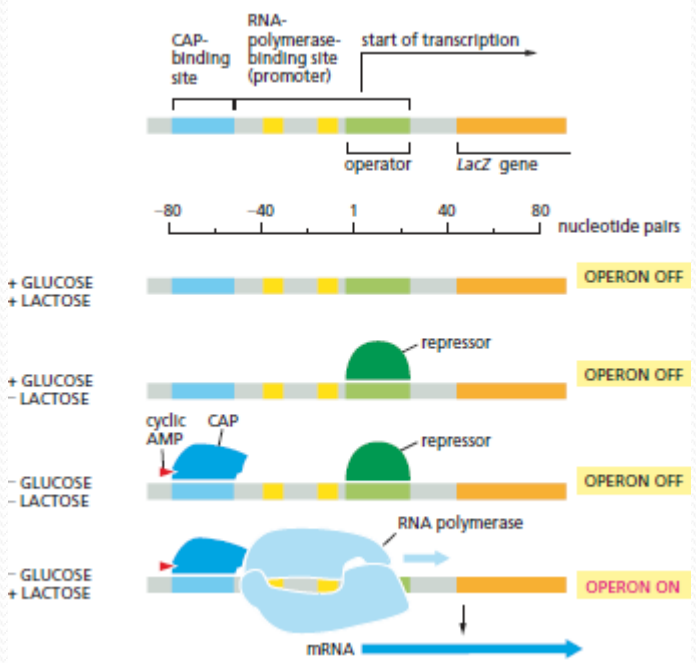
Lac Operon

- Like the tryptophan repressor, activator proteins often have to interact with a second molecule to be able to bind DNA.
- For example, the bacterial activator protein *CAP* has to bind cyclic AMP (cAMP) before it can bind to DNA. Genes activated by CAP are switched on in response to an increase in intracellular cAMP concentration, which rises when glucose, the bacterium's preferred carbon source, is no longer available; as a result, CAP drives the production of enzymes that allow the bacterium to digest other sugars.
- In many instances, the activity of a single promoter is controlled by two different transcription regulators.
- The *Lac operon* in *E. coli*, for example, is controlled by both the *Lac repressor* and the CAP activator.



Lac Operon

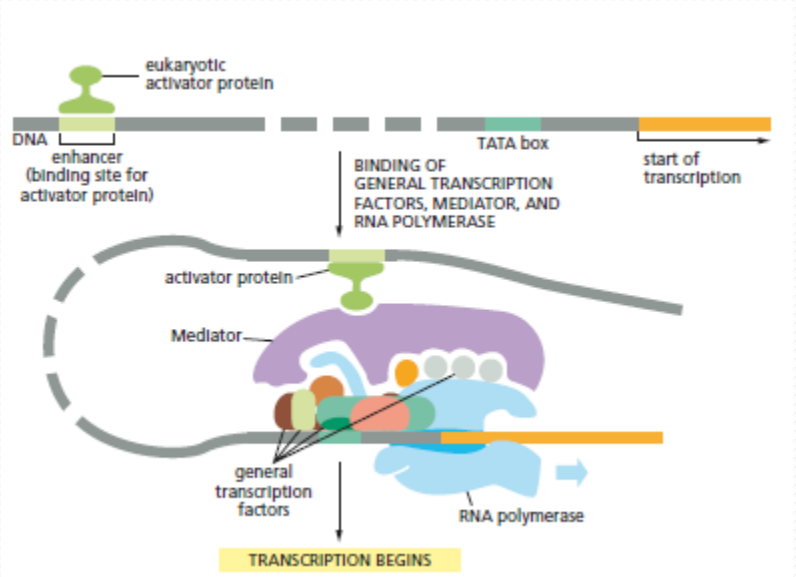
- The *Lac* operon encodes proteins required to import and digest the disaccharide lactose.
- In the absence of glucose, the bacterium makes cAMP, which activates CAP to switch on genes that allow the cell to utilize alternative sources of carbon—including lactose.
- It would be wasteful, however, for CAP to induce expression of the *Lac* operon if lactose itself were not present. Thus the Lac repressor shuts off the operon in the absence of lactose.
- This arrangement enables the control region of the *Lac* operon to integrate two different signals, so that the operon is highly expressed only when two conditions are met: glucose must be absent and lactose must be present.
- When lactose is present AND glucose is absent, the cell executes the appropriate program—in this case, transcription of the genes that permit the uptake and utilization of lactose.



Eukaryotic Gene Regulation

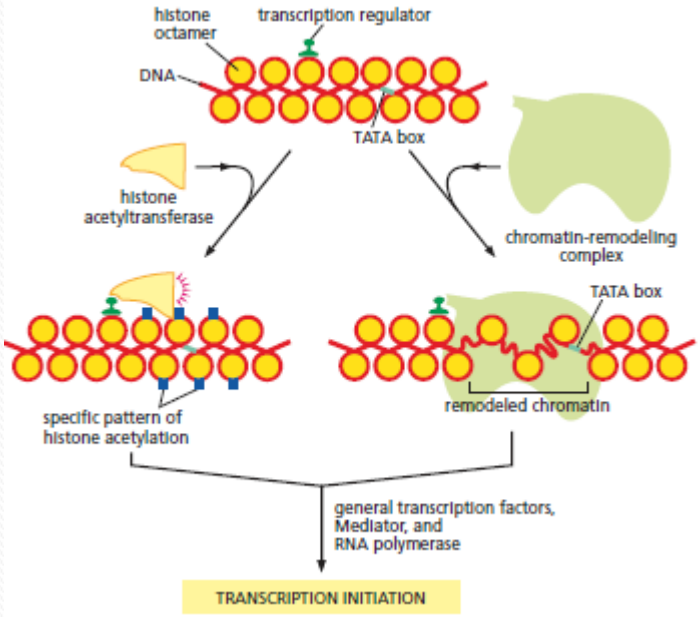
- Eukaryotes, too, use transcription regulators—both activators and repressors—to regulate the expression of their genes. The DNA sites to which eukaryotic gene activators bind are termed ***enhancers***, because their presence dramatically enhances the rate of transcription.
- Activator proteins could enhance transcription even when they are bound thousands of nucleotide pairs away from a gene's promoter.
- They also work when bound either upstream or downstream from the gene.

- The DNA between the enhancer and the promoter loops out to allow eukaryotic activator proteins to influence directly events that take place at the promoter. The DNA thus acts as a tether, allowing a protein that is bound to an enhancer—even one that is thousands of nucleotide pairs away—to interact with the proteins in the vicinity of the promoter—including RNA polymerase and the general transcription factors.
- Often, additional proteins serve to link the distantly bound transcription regulators to these proteins at the promoter; the most important of these regulators is a large complex of proteins known as ***Mediator***.
- One of the ways in which activator proteins function is by aiding the assembly of the general transcription factors and RNA polymerase to form a large ***transcription complex*** at the promoter.
- Eukaryotic repressor proteins do the opposite: they decrease transcription by preventing the assembly of the same protein complex.

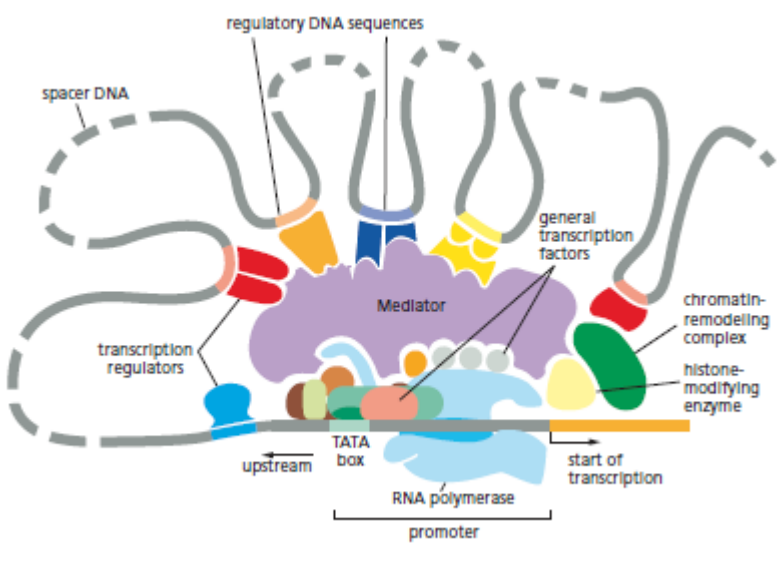


- In addition to promoting—or repressing—the assembly of a transcription initiation complex directly, eukaryotic transcription regulators have an additional mechanism of action: they attract proteins that modify chromatin structure and thereby affect the accessibility of the promoter to the general transcription factors and RNA polymerase.
- Eukaryotic DNA is packed into nucleosomes, which, in turn, are folded into higher-order structures. So transcription regulators, general transcription factors, and RNA polymerase should somehow gain access to such DNA for transcription to occur.
- Nucleosomes can inhibit the initiation of transcription if they are positioned over a promoter, because they physically block the assembly of the general transcription factors or RNA polymerase on the promoter.
- Such chromatin packaging may have evolved in part to prevent leaky gene expression by blocking the initiation of transcription in the absence of the proper activator proteins.
- In eukaryotic cells, activator and repressor proteins exploit chromatin structure to help turn genes on and off.

- As we discussed, chromatin structure can be altered by *chromatin-remodeling complexes* and by enzymes that covalently modify the histone proteins that form the core of the nucleosome.
- Many gene activators take advantage of these mechanisms by recruiting such chromatin-modifying proteins to promoters. For example, the recruitment of *histone acetyltransferases* promotes the attachment of acetyl groups to selected lysines in the tail of histone proteins.
- This modification alters chromatin structure, allowing greater accessibility to the underlying DNA; moreover, the acetyl groups themselves attract proteins that promote transcription, including some of the general transcription factors.
- Likewise, gene repressor proteins can modify chromatin in ways that reduce the efficiency of transcription initiation. For example, many repressors attract *histone deacetylases*—enzymes that remove the acetyl groups from histone tails, thereby reversing the positive effects that acetylation has on transcription initiation.



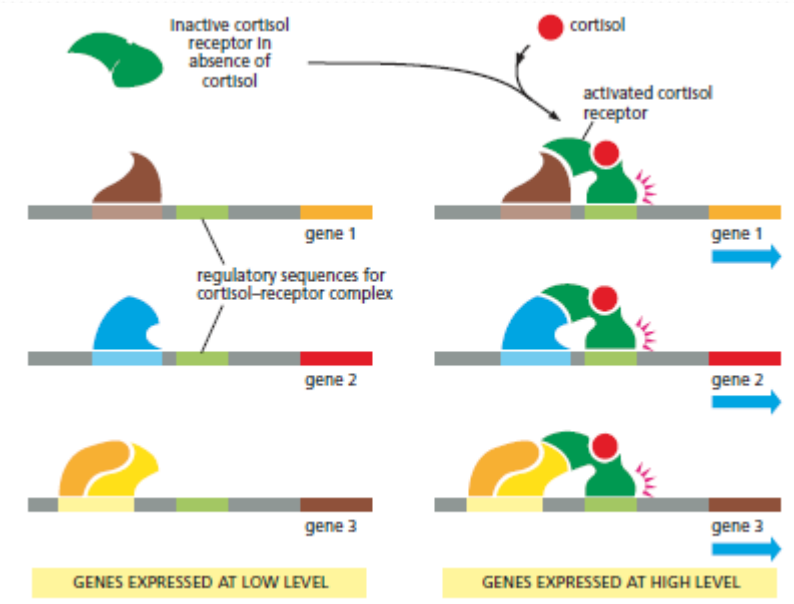
- In contrast, the simplest changes in gene expression in both eukaryotes and bacteria are often only transient.
- Because eukaryotic transcription regulators can control transcription initiation when bound to DNA many base pairs away from the promoter, the nucleotide sequences that control the expression of a gene can be spread over long stretches of DNA.
- In animals and plants, it is not unusual to find the regulatory DNA sequences of a gene dotted over tens of thousands of nucleotide pairs, although much of the intervening DNA serves as *"spacer"* sequence and is not directly recognized by the transcription regulators.
- Most eukaryotic transcription regulators work as part of a "committee" of regulatory proteins, all of which are necessary to express the gene in the right place, in the right cell type, in response to the right conditions, at the right time, and in the required amount.
- The term ***combinatorial control*** refers to the way that groups of transcription regulators work together to determine the expression of a single gene.



- In eukaryotes, the regulatory inputs have been amplified, and a typical gene is controlled by dozens of transcription regulators.
- These help assemble chromatin-remodeling complexes, histone-modifying enzymes, RNA polymerase, and general transcription factors via the multiprotein Mediator complex.
- In addition to being able to switch individual genes on and off, all cells—whether prokaryote or eukaryote—need to coordinate the expression of different genes.
- When an eukaryotic cell receives a signal to divide, for example, a number of unexpressed genes are turned on together to set in motion the events that lead eventually to cell division.
- As discussed earlier, one way in which bacteria coordinate the expression of a set of genes is by having them clustered together in an operon under the control of a single promoter.
- Such clustering is not seen in eukaryotic cells, where each gene is transcribed and regulated individually.

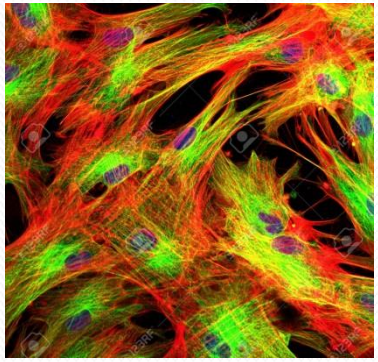
- Even though control of gene expression is combinatorial, the effect of a single transcription regulator can still be decisive in switching any particular gene on or off, simply by completing the combination needed to activate or repress that gene.
- The same protein can complete the combination for several different genes. As long as different genes contain regulatory DNA sequences that are recognized by the same transcription regulator, they can be switched on or off together, as a coordinated unit.
- An example of such coordinated regulation in humans is seen with the *cortisol receptor protein*.

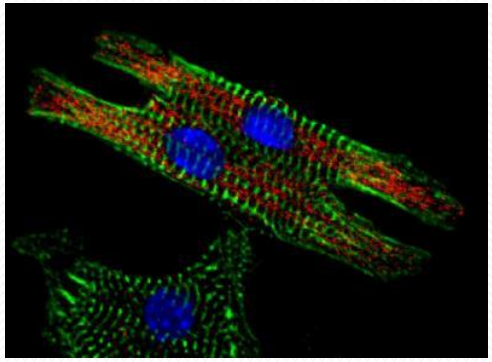
- In order to bind to regulatory sites in DNA, transcription regulator *cortisol receptor protein* must first form a complex with a molecule of cortisol.
- In response to cortisol, liver cells increase the expression of many genes, one of which encodes the enzyme tyrosine aminotransferase.
- All these genes are regulated by the binding of the cortisol–receptor complex to a regulatory sequence in the DNA of each gene.
- When the cortisol concentration decreases again, the expression of all of these genes drops to its normal level.
- In this way, **a single transcription regulator can coordinate the expression of many different genes.**



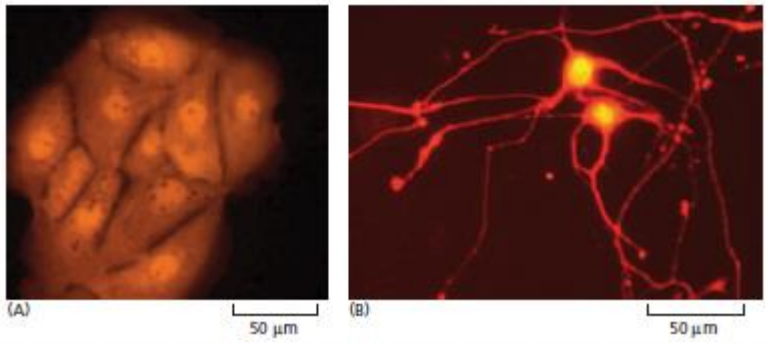
- The ability to switch many different genes on or off using a limited number of transcription regulators is not only useful in the day-to-day regulation of cell function.
- It is also one of the means by which eukaryotic cells diversify into particular types of cells during embryonic development.
- A striking example is the development of muscle cells. A mammalian skeletal muscle cell is distinguished from other cells by the production of a large number of characteristic proteins, such as the muscle-specific forms of actin and myosin that make up the contractile apparatus as well as the receptor proteins and ion channel proteins in the plasma membrane that make the muscle cell sensitive to nerve stimulation.
- The genes encoding these muscle-specific proteins are all switched on coordinately as the muscle cell differentiates.
- Studies of developing muscle cells in culture have identified a small number of key transcription regulators, expressed only in potential muscle cells, that coordinate muscle-specific gene expression and are thus crucial for muscle-cell differentiation.
- This set of regulators activates the transcription of the genes that code for muscle-specific proteins by binding to specific DNA sequences present in their regulatory regions.

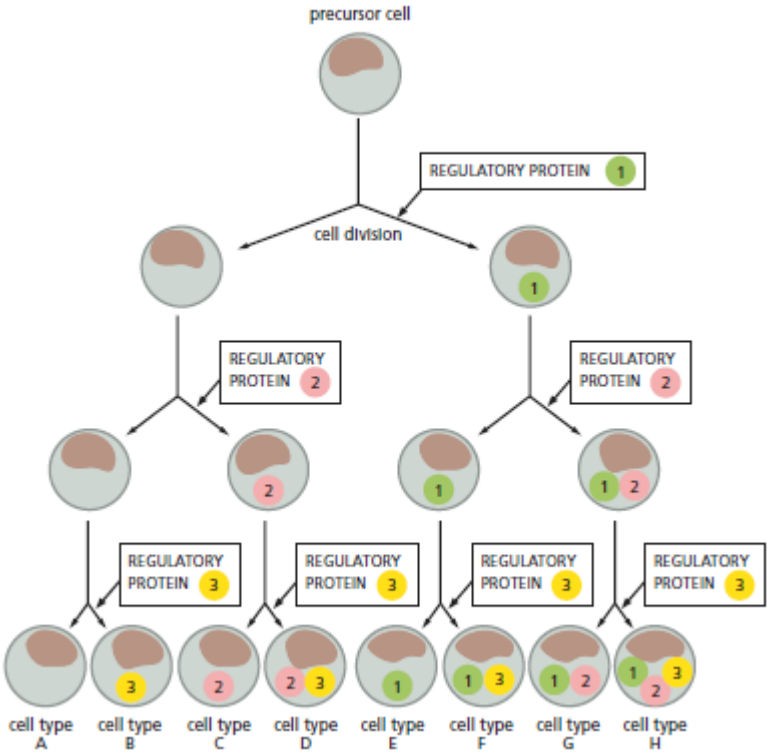
- Some transcription regulators can even convert one specialized cell type to another. For example, when the gene encoding the transcription regulator MyoD is artificially introduced into fibroblasts cultured from skin connective tissue, the fibroblasts form musclelike cells.
- It appears that the fibroblasts, which are derived from the same broad class of embryonic cells as muscle cells, have already accumulated many of the other necessary transcription regulators required for the combinatorial control of the muscle-specific genes, and that addition of MyoD completes the unique combination required to direct the cells to become muscle.



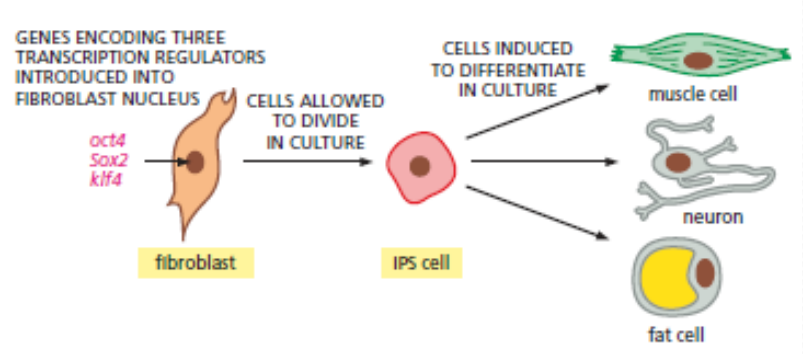


- This type of reprogramming can produce even more dramatic effects.
- For example, a set of nerve-specific transcription regulators, when artificially expressed in cultured liver cells, can convert them into functional neurons.
- Such dramatic results suggest that it may someday be possible to produce in the laboratory any cell type for which the correct combination of transcription regulators can be identified.





- We have seen that, in some cases, one type of differentiated cell can be experimentally converted into another type by the artificial expression of specific transcription regulators.
- Even more surprising, transcription regulators can reprogramme various differentiated cells into pluripotent stem cells that are capable of giving rise to all the specialized cell types in the body, much like the embryonic stem (ES) cells.
- Using a defined set of transcription regulators, cultured mouse fibroblasts have been reprogrammed to become induced pluripotent stem (iPS) cells—cells that look and behave like the pluripotent ES cells that are derived from embryos.
- The approach was quickly adapted to produce iPS cells from a variety of specialized cell types, including cells taken from humans. Such human iPS cells can then be directed to generate a population of differentiated cells for use in the study or treatment of disease.



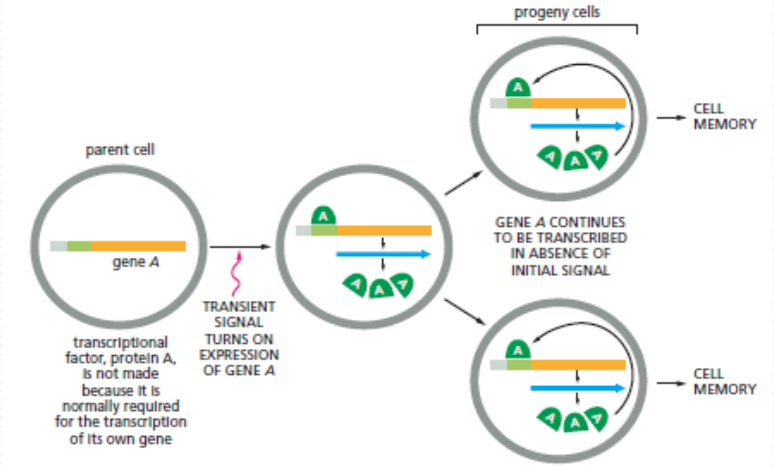
- We have seen that a small number of transcription regulators can control the expression of whole sets of genes and can even convert one cell type into another.
- But an even more stunning example of the power of transcriptional control comes from studies of eye development in *Drosophila*.
- In this case, a single "master" transcription regulator called Ey could be used to trigger the formation of not just a single cell type but a whole organ.
- In the laboratory, the *Ey* gene can be artificially expressed in fruit fly embryos in cells that would normally give rise to a leg. When these modified embryos develop into adult flies, some have an eye in the middle of a leg.

- Ey functions like any other transcription regulator, controlling the expression of multiple genes by binding to DNA sequences in their regulatory regions.
- Some of the genes controlled by Ey encode additional transcription regulators that, in turn, control the expression of other genes.
- In this way, the action of a single transcription regulator can produce a cascade of regulators that, working in combination, lead to the formation of an organized group of many different types of cells.

Cell memory

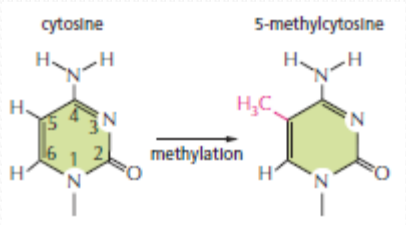
- Once a cell in a multicellular organism becomes committed to differentiate into a specific cell type, the choice of fate is generally maintained through subsequent cell divisions.
- Some highly specialized cells, including skeletal muscle cells and neurons, never divide again once they have differentiated—that is, they are *terminally differentiated*.
- But many other differentiated cells—such as fibroblasts, smooth muscle cells, and liver cells—will divide many times in the life of an individual.
- When they do, these specialized cell types give rise only to cells like themselves: smooth muscle cells do not give rise to liver cells, nor liver cells to fibroblasts.
- For a proliferating cell to maintain its identity—a property called ***cell memory***—the patterns of gene expression responsible for that identity must be remembered and passed on to its daughter cells through all subsequent cell divisions.
- This means that the changes in gene expression, which are often triggered by a transient signal, must be remembered by the cell. This phenomenon of *cell memory* is a prerequisite for the creation of organized tissues and for the maintenance of stably differentiated cell types.

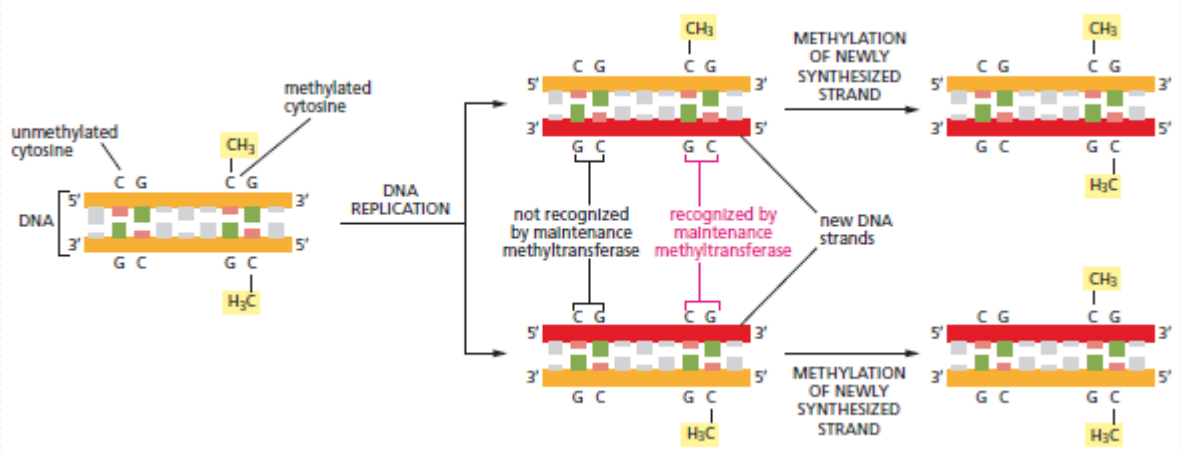
- Cells have several ways of ensuring that their daughters “remember” what kind of cells they are. One of the simplest and most important is through a ***positive feedback loop***, where a master transcription regulator activates transcription of its own gene, in addition to that of other cell-type–specific genes.
- Each time a cell divides the regulator is distributed to both daughter cells, where it continues to stimulate the positive feedback loop. The continued stimulation ensures that the regulator will continue to be produced in subsequent cell generations.
- The Ey protein discussed earlier functions in such a positive feedback loop. Positive feedback is crucial for establishing the “self-sustaining” circuits of gene expression that allow a cell to commit to a particular fate—and then to transmit that information to its progeny.



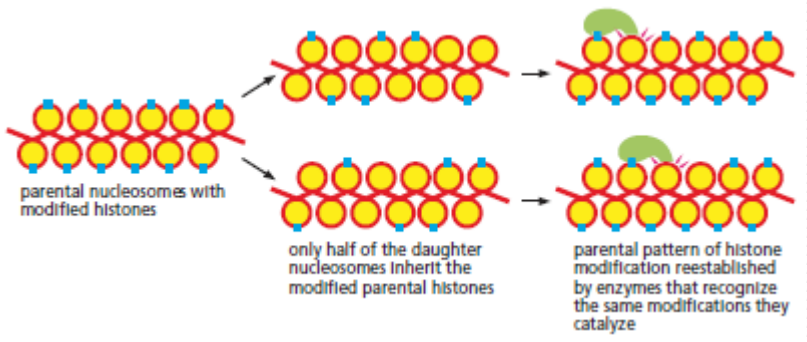
Epigenetics

- Epigenetic mechanism is another way of reinforcing cell identity.
- DNA methylation, one of the epigenetic mechanism, involves cytosine methylation that turns off genes by attracting proteins that bind to methylated cytosines and block gene transcription.
- DNA methylation patterns are passed on to progeny cells by the action of an enzyme that copies the methylation pattern on the parent DNA strand to the daughter DNA strand as it is synthesized.





- Another epigenetic mechanism for inheriting gene expression patterns involves the modification of histones.
- When a cell replicates its DNA, each daughter double helix receives half of its parent's histone proteins, which contain the covalent modifications of the parent chromosome.
- Enzymes responsible for these modifications may bind to the parental histones and confer the same modifications to the new histones nearby.
- This cycle of modification reestablishes the pattern of chromatin structure found in the parent chromosome.



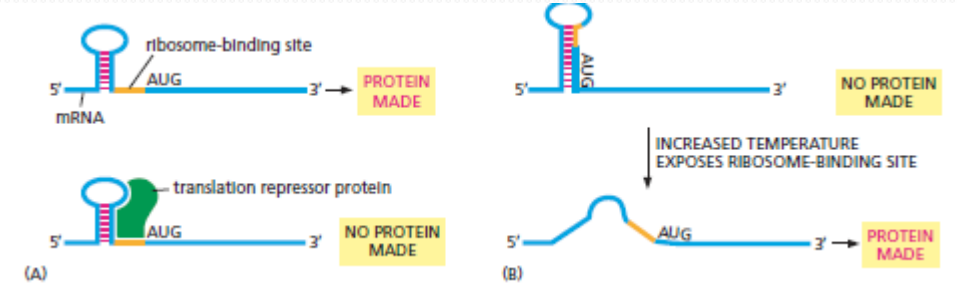
- Because all of these cell-memory mechanisms transmit patterns of gene expression from parent to daughter cell without altering the actual nucleotide sequence of the DNA, they are considered to be forms of epigenetic inheritance.
- Such epigenetic changes play an important part in controlling patterns of gene expression, allowing transient signals from the environment to be permanently recorded by our cells.

Post-Transcriptional Control of Gene Expression

- We have seen that transcription regulators control gene expression by promoting or hindering the transcription of specific genes. The vast majority of genes in all organisms are regulated in this way.
- But many additional points of control can come into play later in the pathway from DNA to protein, giving cells a further opportunity to regulate the amount or activity of the gene products that they make.
- These post-transcriptional controls, which operate after transcription has begun, play a crucial part in regulating the expression of almost all genes. Examples include:
  - Alternative RNA splicing allows different forms of a protein, encoded by the same gene, to be made in different tissues.
  - Post-translational modifications of a protein can regulate its concentration and activity.

- The more time an mRNA persists in the cell before it is degraded, the more protein it will produce.
- In bacteria, most mRNAs last only a few minutes before being destroyed. This instability allows a bacterium to adapt quickly to environmental changes. Eukaryotic mRNAs are generally more stable.
- Most eukaryotic mRNAs, however, have half-lives of less than 30 minutes, and the most short-lived are those that encode proteins whose concentrations need to change rapidly based on the cell's needs, such as transcription regulators.
- Whether bacterial or eukaryotic, an mRNA's lifetime is dictated by specific nucleotide sequences within the untranslated regions (UTR) that lie both upstream and downstream of the protein-coding sequence. These sequences often harbor binding sites for proteins that are involved in RNA degradation or blocking of translation.

- Some of these sequences control how often or how efficiently the mRNA will be translated into protein. These sequences control translation initiation.
- Bacterial mRNAs contain a short ribosome-binding sequence located a few nucleotide pairs upstream of the AUG codon where translation begins. This binding sequence forms base pairs with the RNA in the small ribosomal subunit, correctly positioning the initiating AUG codon within the ribosome.
- Because this interaction is needed for efficient translation initiation, it provides an ideal target for translational control. By blocking—or exposing—the ribosome-binding sequence, the bacterium can either inhibit—or promote—the translation of an mRNA.
- Eukaryotic mRNAs possess a 5′ cap that helps guide the ribosome to the first AUG, the codon where translation will start.
- Eukaryotic repressor proteins can inhibit translation initiation by binding to specific nucleotide sequences in the 5′ untranslated region of the mRNA, thereby preventing the ribosome from finding the first AUG—a mechanism similar to that in bacteria.
- When conditions change, the cell can inactivate the repressor to initiate translation of the mRNA.

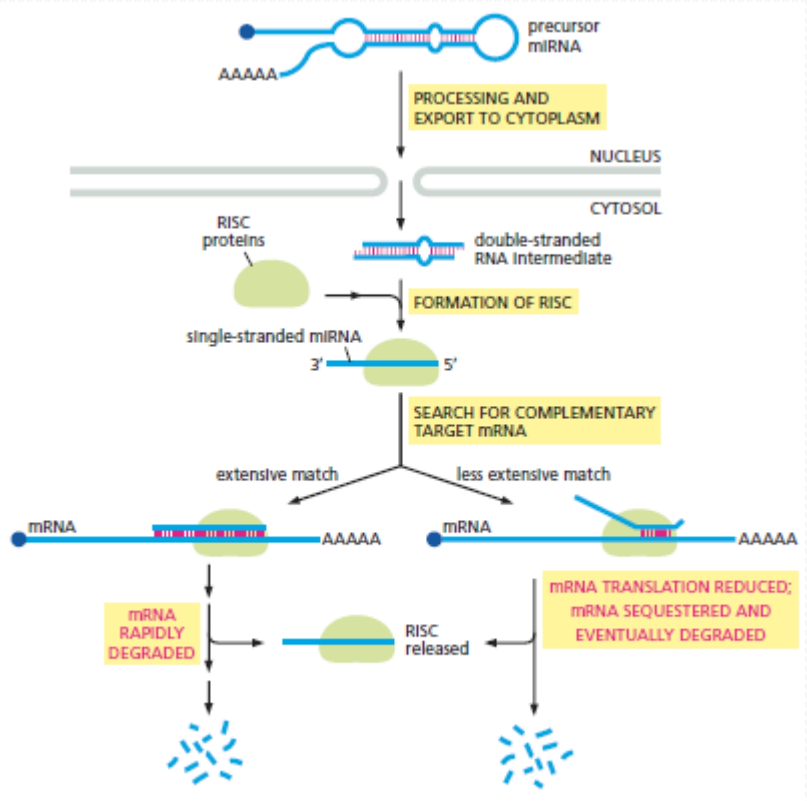


Regulatory RNAs

- In addition to the mRNAs, which code for proteins, *noncoding RNAs* have various functions.
- Some have key structural and catalytic roles, particularly in protein synthesis by ribosomes (tRNA and rRNA).
- Some others have important roles in regulating gene expression and are therefore referred to as ***regulatory RNAs***.
- There are at least three major types of regulatory RNAs:
  - *microRNAs*,
  - *small interfering RNAs*,
  - *long noncoding RNAs*.

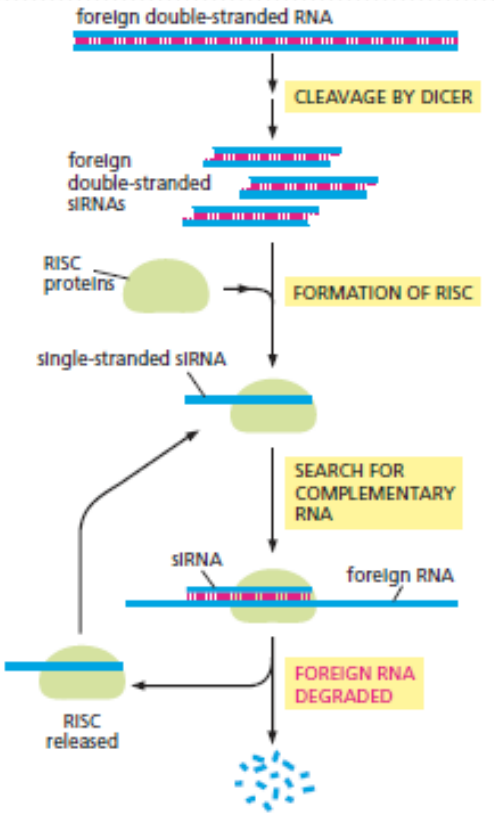
miRNA

- MicroRNAs, or miRNAs, are tiny RNA molecules that control gene expression by base-pairing with specific mRNAs and reducing both their stability and their translation into protein.
- Mature, functional miRNA molecule, which is only about 22 nucleotides in length.
- This small but mature miRNA is packaged with specialized proteins to form an *RNA-induced silencing complex* (*RISC*), which patrols the cytoplasm in search of mRNAs that are complementary to the bound miRNA molecule.
- Once a target mRNA forms base pairs with an miRNA, it is either destroyed immediately by a nuclease present within the RISC or its translation is blocked.



siRNA

- Some of the same components that process and package miRNAs also play another crucial part in the life of a cell: they serve as a powerful cell defense mechanism.
- In this case, the system is used to eliminate "foreign" RNA molecules—in particular, the double-stranded RNAs produced by many viruses and transposable genetic elements.
- siRNA resembles certain aspects of the adaptive immune responses of vertebrates; in both cases, an invading pathogen elicits the production of molecules—either siRNAs or antibodies— that are custom-made to inactivate the specific invader and thereby protect the host.



lncRNA

- Long noncoding RNAs are a class of RNA molecules that are more than 200 nucleotides in length.
- With few exceptions, their roles in the biology of the organism are not entirely clear.
- One of the best understood of the long noncoding RNAs is ***Xist***. This enormous RNA molecule, some 17,000 nucleotides long, is a key player in X inactivation—the process by which one of the two X chromosomes in the cells of female mammals is permanently silenced.
- Early in development, Xist is produced by only one of the X chromosomes in each female nucleus. The transcript then "sticks around," coating the chromosome and presumably attracting the enzymes and chromatin remodeling complexes that promote the formation of highly condensed heterochromatin.

- Other long noncoding RNAs may promote the silencing of specific genes in a similar manner.
- Some long noncoding RNAs arise from protein-coding regions of the genome, but are transcribed from the "wrong" DNA strand. Some of these *antisense* transcripts are known to bind to the mRNAs produced from that DNA segment, regulating their translation and stability.
- Regardless of how the various long noncoding RNAs operate—or what exactly they do—the discovery of this large class of RNAs reinforces the idea that a eukaryotic genome is densely packed with information that provides not only an inventory of the molecules and structures every cell must make, but a set of instructions for how and when to assemble these parts to guide the growth and development of a complete organism.