DNA Replication and Protein Synthesis

**Introduction:**

The genetic material of nearly every organism is Deoxyribonucleic acid, or DNA. Cells have their own DNA, but that doesn’t help when the cell is trying to create a new cell and has no genetic material to pass on to the daughter cell. To compensate for this issue, DNA replicates to create new strands of DNA identical to the original strand in order to pass the genetic material onto any daughter cells it might make. However genetic material isn’t enough to perform all the functions in the cell. DNA acts as a template for the creation of proteins which actually do the work in the cell. The process of reading DNA and eventually creating proteins is actually relatively similar to how DNA replication occurs.

**Starting DNA Replication:**

DNA has a structure called a double helix. This means that DNA is actually made of two strands of nucleic acids connected to each other by hydrogen bonds. The resulting shape is one where both connected strands twist to form a helix. This shape can pose some problems for DNA replication, however, because all the information that is needed to create a new strand of DNA is on the inside of the double helix and very hard to get to. In order to get to the nitrogenous bases at the core of DNA, the double helix has to unwind and an enzyme called helicase comes in to break the hydrogen bonds between the base pairs of the two strands. This disconnects the two strands of the double helix from each other and exposes the nitrogenous bases for easier access later in replication. The separating also untwists the two strands from their double helix shape, but this too runs into an issue. Unwinding one end of something that is twisted without moving the other end too makes one end get much more twisted than it was previously to compensate for the other end getting untwisted, and this is exactly what happens in DNA. As you unwind one end of DNA, the other end gets more twisted and can get coiled around itself and make it very hard for helicase to sever the hydrogen bonds in those sections. This is called supercoiling and can make it impossible for DNA replication to occur. There is a remedy for this problem, however, in the form of an enzyme called topoisomerase, or gyrase. This enzyme attaches to where the supercoiling is occurring and will sever the phosphate backbone of the DNA to allow that section of the DNA to rotate, almost like a top, and relieve any supercoiling that was occurring. So the helicase can continue to unwind and unzip the double helix of the DNA, but separating the two strands of DNA actually makes the strands more fragile than they were together, and more likely to break down, so to help the single stranded DNA remain intact until replication can be completed, a single-stranded binding protein will come in and bind to the DNA to keep it together and protected from nucleases that might have degraded them otherwise. This isn’t all the single-stranded binding proteins, or SSB proteins, do however. They also work to remove secondary structures on the single stranded DNA, which makes enzymes function more effectively on the DNA than it would have with the secondary structures still attached. This function might even make DNA replication up to one hundred times faster than it before. With all of these enzymes and proteins working together, the DNA double helix is unwound, unzipped, protected and primed for the rest of DNA replication.

**DNA Replication:**

After helicase and all the other enzymes and proteins move down the DNA, the remaining two strands of single stranded DNA and the gap between them is called the replication fork. It looks a bit like a ‘Y’ and is where the rest of replication will take place. Once the two strands are separated and ready for replication, DNA polymerase will come in and start to lay down new nitrogenous bases that would pair with the existing nitrogenous bases. DNA polymerase, however, only adds bases in the 5’ to 3’ direction, and one of the two strands is going in the wrong direction for this to happen easily. The two strands of DNA run anti-parallel, which means that while one strand will run 5’ to 3’, the other will run 3’ to 5’. This second strand is called the lagging strand in DNA replication, because it goes in the wrong direction of replication and bases have to be added discontinuously, unlike the other strand, which is added to continuously. However, before replication can run into this problem, DNA polymerase has to have a strand of nucleotides to add to in the first place. DNA polymerase cannot just start laying down the base pairs of the strand of DNA, there has to be a something to add to, which is where primers come in. A primer is a short strand of eight to ten nucleotides and is laid down at the very beginning for DNA polymerase to recognize and start adding bases to. These primers are usually provided by the aptly named enzyme primase. Now that a primer has been laid down and DNA polymerase has recognized it, it will start to add bases onto the strand of DNA towards the 3’ end of the strand. The 5’ to 3’ leading strand has an easy time of this, but the 3’ to 5’ lagging strand will be replicated in pieces called Okazaki fragments. These fragments will later be connected, but first let’s get a deeper understanding into the nitrogenous bases and how they are added to the strand of DNA.

 DNA has four different nitrogenous bases: adenine, thymine, guanine, and cytosine. The single strand of DNA can have these four bases in any order possible. Nitrogenous bases come in two types, purines and pyrimidines. Cytosine and thymine are both pyrimidines, while adenine and guanine are purines. One way to easily remember this is that cytosine, thymine and pyrimidine all have the letter ‘y’ in the word, while adenine, guanine and purine do not. Each base has a pair that it will connect to on the other strand of DNA that it will form hydrogen bonds with to keep the two strands of DNA together in the double helix shape. Adenine will always pair with thymine, and guanine will always pair with cytosine. The pairs will also form different numbers of hydrogen bonds between them. Adenine and thymine will create two hydrogen bonds to hold them together, while cytosine and guanine will form three. The way that DNA polymerase knows what base to add to the single strand of DNA to create a complementary strand is that it reads the base pair on the strand of DNA and will add the base that is its pair. If the base on the single strand of DNA is a cytosine, it will add a guanine to the growing strand, and if it’s an adenine, it will add a thymine and so on. This process will continue for both the leading and the lagging strand, but the lagging strand will require a new primer every time it starts a new fragment. After DNA polymerase adds the bases to the single strands of DNA, another form of polymerase will come through and scan the bases that the first DNA polymerase added to the strand of DNA. This polymerase is proofreading the bases that have been laid down to make sure that there haven’t been any mistakes in the added bases. A mistake in the code of the DNA could potentially be disastrous for the organism, so any mistakes have to be caught and corrected early. This second type of DNA polymerase will make sure that no wrong bases have been added or not added at all, and if any of these problems arise, it will correct the issue by either taking out the wrong base and adding the right one, or just adding the correct base in the first place. After this DNA polymerase has finished proofreading the DNA strands to make sure that everything is correct, DNA replication can finally come to an end.

**The End of DNA Replication:**

While it might seem like DNA replication is finished after proofreading, it is not. The lagging strand is still in fragments, and that cannot be the case for a complete strand of DNA. In addition, the primers from the very beginning of DNA replication have been replaced with matching nitrogenous bases by the second, proofreading DNA polymerase. These short sections of bases are also not connected to the rest of the strand of DNA, so not only do the Okazaki fragments of the lagging strand need to be connected, any primers for both strands also have to be connected to the rest of the DNA. The solution to both of these problems is the same, the enzyme DNA ligase. DNA ligase will connect the phosphate backbone back together at any of the breaks, connecting the fragments and primers to the together with other bases. All of these processed happen at generally the same time, making DNA replication very efficient, and by the time helicase makes it to the other end of the strand of DNA, you are left with two identical double helixes of DNA, each with one strand of DNA from the original double helix and one new strand of DNA. This is because DNA replication is semiconservative, meaning any new double helix of DNA is actually half the old strand and half new.

**The Start of Protein Synthesis:**

Protein synthesis is actually relatively close in some of the processes to DNA replication. The product is different and the names are different, but having a good grasp on DNA replication will certainly help understanding protein synthesis. DNA can’t do much by itself, it is more of a master code to make other things that can help the cell function. One of the things that DNA is a template for is the synthesis of proteins. Like in DNA replication, the DNA double helix has to be unwound and unzipped for protein synthesis to start. The biggest difference between the two processes is that when helicase separates the two strands of DNA from each other, it is not forever, and the two strands to eventually reconnect. This is due to the fact that DNA isn’t used up in this process, it is simply used as a guide. Another big difference is that DNA polymerase isn’t used, it uses instead RNA polymerase, because the product of this process will be RNA, not DNA. The first step is the eventual creation of RNA that will act as a messenger, because protein synthesis occurs in the ribosomes, but DNA doesn’t leave the nucleus and therefore cannot go to the ribosomes. So, when the two strands of the double helix are separated, RNA polymerase will add the matching bases to one of the strands of DNA. RNA polymerase doesn’t add bases in quite the same way that DNA polymerase adds bases, however. RNA polymerase will still add cytosine to any guanine, guanine to any cytosine, and adenine to any thymine, but it will not add thymine to adenine. This is due to the fact that RNA polymerase doesn’t have thymine at all, it instead uses uracil, and will add that to any adenine on the DNA strand. Uracil is a pyrimidine just like adenine and thymine, but it is uses exclusively in RNA. Once all the bases have been matched and added by the RNA polymerase, the new bases will not connect to the strand of DNA, and will instead leave the replication fork. These RNA bases are not yet ready to leave the nucleus, however, and are known as pre-messenger RNA. Pre-messenger RNA is a very long sequence of nitrogenous bases and includes introns. Introns are long noncoding parts of genetic material, and it was copied from the DNA into the pre-messenger RNA. For the strand to eventually become messenger RNA, or mRNA, these introns will have to be removed because they don’t serve any purpose to the mRNA. The process of removing the introns is called RNA splicing. RNA splicing removes all the introns and connects the exons, or the coding regions of the genetic material, together again to form a shorter, more compact strand of RNA, which is the messenger RNA that it was meant to be. mRNA’s purpose is to take the important part of the template that DNA provides out of the nucleus, where protein synthesis cannot occur, to the ribosomes where it can occur. Once the mRNA gets to the ribosomes in the cytoplasm, its job is finished and now it will act as a template for the rest of protein synthesis.

**Translation:**

Translation is a bit more complicated than DNA replication or the synthesis or messenger RNA. When mRNA enters the ribosome, a small ribosomal subunit will attach to the ‘back’ of the mRNA, or the side of the mRNA where the bases are not sticking out. It will move along the strand until it reaches a very specific sequence of three nitrogenous bases, or a start codon. This codon is almost always the sequence AUG, and once the small ribosomal subunit finds it, a special kind of RNA called transfer RNA, or tRNA, which is connected to a specific amino acid, will attach to the codon. This prompts the large ribosomal subunit to bind to the other end of the mRNA, covering the tRNA that just attached to the mRNA as well. The large ribosomal is shaped almost like the letter ‘m’ except it has three gaps instead of two. These gaps have specific names and purposes. The large ribosomal subunit has gaps called, from right to left, the A site, the P site, and the E site. While the first tRNA is automatically placed in the P site, any new tRNAs will enter the A site first, an easy way to remember this is to consider the A site the ‘acceptor’ site. The E site is where the tRNA will be ejected, so you can think of it as the ‘exit’ site. The P site is a little more complicated. Each tRNA is connected to a very specific amino acids, which will eventually create the protein. The purpose of translation is the creation of a polypeptide chain from the template the mRNA provides, and a polypeptide chain is just a series of amino acids in a chain. You can think of the P site as a polypeptide holding site, because it is usually where the chain remains during translation. As a new tRNA recognizes the next codon in the mRNA and enters the large ribosomal subunit, the amino acid on the tRNA in the P site is shifted to the tRNA in the A site, making the amino acid originally on that tRNA the last and most recent addition to the polypeptide chain. Each new tRNA to enter the large ribosomal subunit will become the new holder of the polypeptide chain, and the other tRNA will lose all the amino acids that were attached to it. They all remain in a chain, however, and will just be shifted to the new tRNA. When this happens, the large ribosomal subunit shifts, and the tRNA in the A site enters the P site and the tRNA in the P site enters the E site and then leaves the subunit to be used again later. As this continues, the chain gets longer and longer and eventually it will reach the opposite of the start codon, the stop codon, which in RNA is UAG, UAA, or UGA. These codons don’t actually have an amino acid that they code for, so no tRNA will enter the A site and bind to the mRNA. Instead, a termination protein does. This termination protein won’t have any amino acid attached to it, and it won’t be able to take the chain from the tRNA in the P site. Instead, the polypeptide chain that is attached to the tRNA in the P site will detach from the tRNA. The tRNA will move into the E site and leave the large ribosomal subunit, and when the termination protein enters the P site, it will also cause both the large and the small ribosomal subunits to detach from the mRNA strand. The tRNA and both ribosomal subunits will be used again in later translations, because they have only been used, not used up. The polypeptide chain, now that it is detached from the tRNA, is will fold around itself to form a particular shape unique to the type of protein that it is. Every protein folds into a shape that actually aid in its eventual function. There are two different ways that the protein can fold after it is synthesized, called a secondary structure. It could fold into an alpha-helix, which is coiled like a spring and turns in a clockwise direction. It could also fold into a beta-pleated sheet, which is folded in an almost zig-zag pattern, with each fold lying next to each other in an anti-parallel fashion. These folds can happen along the polypeptide chain making up the protein, so it can have both types of folding at once. The final structure, or tertiary structure, of the protein is when all of the different alpha-helices and beta-sheets on the polypeptide chain fold in on each other to form a three-dimensional shape. These fully folded proteins can also interact and fold around other fully formed proteins to form a quaternary structure, but that’s usually for proteins that contain multiple polypeptide chains.

**Summary:**

DNA replication and protein synthesis are both complicated processes in the cell, but are essential to the functions that take place both inside and outside the cell. They both put a great deal of importance on the original strand of DNA, with replication using it to create new strands of DNA and protein synthesis using it as a template to eventually create proteins. This is understandable, because DNA is the genetic material of the cell and is used in a great many processes in the cell. To know what keeps cells running and what makes new cells, it is important to understand both the process of protein synthesis and the process of DNA replication.

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