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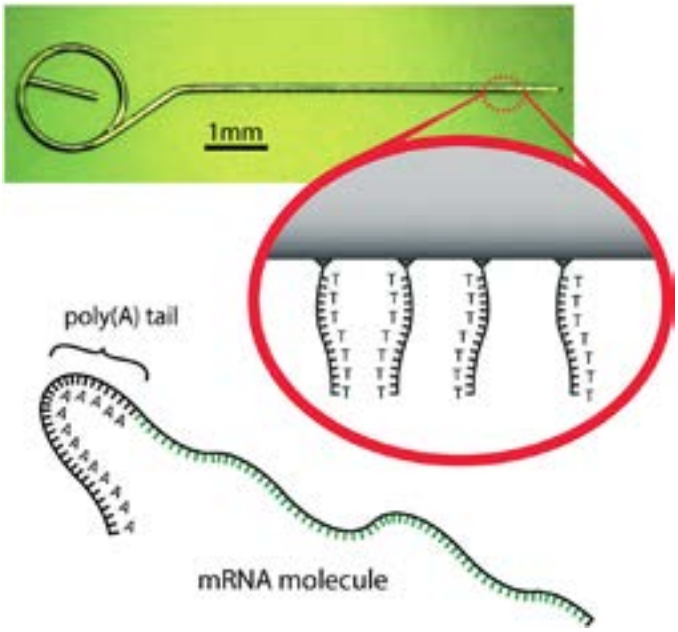

Diagram of a functionalized needle and a mRNA molecule

mRNA Extraction Using Functionalized Needles

by Thomas Holland, Biomedical Engineering Junior

DNA sequencing and testing has a special interest in the scientific world. Through DNA testing we can create specialized medical treatments, understand mutation, track genetics, target specific viruses and tumors, and access a host of other possibilities. One of the key aspects of any DNA testing is the process of PCR or polymerase chain reaction, which is the replication and amplification of a specific segment of DNA. While PCR is a simple process, scientists must go through the lengthy process of extracting tissues and then isolating the target DNA. However, through the use of specialized needles, this process can be shortened to a simple pinprick.

PCR begins with a mixture of buffers, proteins, enzymes and primers. The buffer protects the cells from damage; the enzymes and proteins facilitate the replication, and the primers enable the replication of a specific segment of the target DNA. A small amount of DNA is added to the mixture, and the mixture goes through repeated cycles of heating and cooling. This cycle causes the proteins and enzymes to activate and replicate the DNA. This process is capable of turning a single strand of DNA into millions of strands. Now that the segment of DNA has been amplified, any number of tests and experiments can be performed.

The only question now is where does the DNA come from. Since all cells contain the same DNA, a sample can be taken from any part of the specimen. There are multiple techniques for extracting and purifying the DNA, but they all involve the same basic aspects. First, the cytoplasm of the cells is broken down using chemicals and enzymes. Then, the proteins and RNA are similarly broken down. Proteins that bind to the DNA are then added, these proteins typically enlarge the DNA in some way. Next, the sample is put into a centrifuge. This centrifuge separates the bonded DNA which then can be extracted from the main mixture. This process takes time and precision to ensure accuracy.

To aid in DNA extraction, Louisiana Tech faculty Dr. Gergana Nestorova and Dr. Niel Crews created a method of extracting mRNA using functionalized needles. To facilitate the extraction of mRNA, the needles have immobilized oligos attached to them. Oligos are artificially created strands of nucleotides. These strands have the same form and substance of DNA, although, they do not contain any genetic information. These oligos are made specifically to be complimentary to the tails of mRNA. This complimentary relationship allows the mRNA to bind to the oligos. Once the needles have been functionalized, they can now be inserted into sample tissues directly into the target cell. Only the mRNA will bind to the oligos. Despite other particles clinging to the needles, the irrelevant portions can simply wash off.

Now that the mRNA has been extracted, it must go through a process called reverse transcription or RT. RT converts mRNA back into DNA using enzymes and proteins. RT is the same method that the body uses to convert DNA into mRNA but in reverse. This process takes a matter of minutes. After reverse transcription, PCR can be accomplished like normal and the DNA tested.

This method of mRNA extraction is quicker than traditional methods. It is also cost efficient because the only equipment required is the needles and a few other reagents. The major downside to this process is the difficulty of handling the needles, which is currently being addressed. Once a more convenient handling method is created, these needles will increase the ease and availability of DNA testing. This technique even has great potential for applications on the international space station.

Sources
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