On the Pursuit of a Cure: The Experience of a Young Leader at The University of Southern California

By Emily Zhang, Freshman Majoring in Health and Humanity

***Abhishek Verma**, a freshman at the University of Southern California, enjoys research not only because of the skills that he acquires through the experience, but also because of the pride he feels in being able to help in the continued advancement of modern science.*

Ever since high school, Abhishek Verma knew he wanted to pursue a career in medicine. Born in India, Verma lived in a number of places before settling down in Palo Alto, California, where he has resided since 5th grade. Although he has always been slightly interested in science, it was not until his biology courses in high school that his interest in the subject truly began to blossom. Armed with new knowledge and an eagerness to learn, Verma looked for opportunities to implement his interest in science. This opportunity came through a research partnership between his high school and Stanford University. "The partnership allowed me to conduct research for Stanford University," recalls Verma, "It turned out to be really fun, and I also got to meet a lot of cool people. I guess you could say it was my starting point."

Verma continued his research through the Young Scholars Program at UC Davis, a six week residential program during the summer for qualified high school students. During his six weeks stay at UC Davis, Verma researched the effect of fertilizer use and agricultural methods in Belize, South America, on malaria. "Just like the first time I did research, the research I did at Davis was extremely enjoyable. From those experiences, I knew I wanted to continue researching." With a clear mindset, Verma entered the University of Southern California in August 2010 as one of the few students accepted into the highly selective USC Baccalaureate/MD Program. Through persistence and a little bit of luck, he was able to land a position as a research assistant for Dr. Chien-Ping Ko, a Professor of Biological Sciences at

USC. Ambitious and talented, Abhishek Verma represents one of the few freshman students at USC who has already secured a research position.

Currently, Dr. Chien-Ping Ko's research focuses on finding the mechanism and causes of Spinal Muscular Atrophy (SMA). Every year, hundreds of infants are diagnosed with SMA, an inherited neuromuscular disease characterized by the loss of voluntary muscle function resulting from the degeneration of nerve cells in muscles of affected infants. SMA is the number one inherited cause of death in children under the age of two, and of those diagnosed before the age of two, fifty percent will die before their second birthday. One in every 40 people are carriers of the gene responsible for SMA, and one in every 6000 babies are born with the inherited disease. SMA usually has three different levels of severity, ranging from severe to mild. Children born with mild SMA can live up to adulthood, although they might experience trouble walking, bending, or getting up by themselves. Children born with intermediate SMA are usually able to sit with assistance but cannot stand, while children born with severe SMA cannot sit or stand, and may even have trouble lifting their heads or accomplishing the normal motor skills expected early on in infancy. To date, there is no known cure for SMA. Individuals with the disease are born with normal motor neurons in muscle tissue (Figure 1). However, for reasons that are still unknown, these motor neurons begin to degenerate, leading to undeveloped receptors of the neurons. Normally, the receptors of neurons allow for signal transmission between neurons and the environment. Muscle tissues contract or relax when motor neurons embedded in the tissues receive signals to do so. However, in the case of SMA, undeveloped receptors do not receive signals from the environment or other motor neurons, leading to loss of muscle control. Consequently, individuals diagnosed with SMA will eventually experience breathing and feeding difficulties, as well as progressive weakness due to a lack of muscle control.

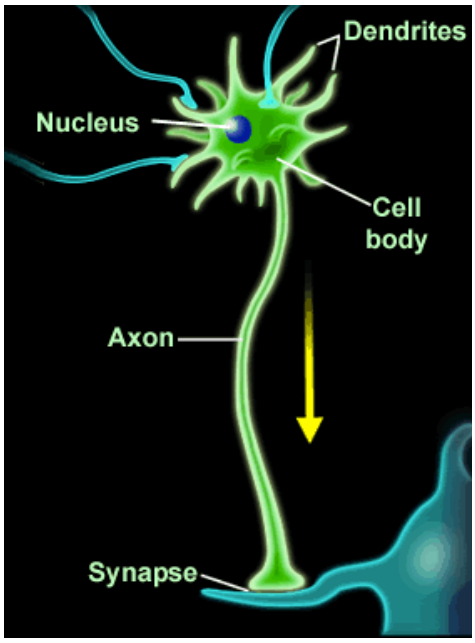

Figure 1 *The above diagram depicts a neuron, with each part labeled. Normal neurons receive signals at the dendrites. The electrical signal is then transferred down the axon towards the synapse, the space between the neuron transmitting the signal, and the neuron or target cell receiving the signal*

In order to gain insight on the cause of SMA, Verma compares sample neurons from mice with normal genes to mice with the necessary mutated genes that leads to SMA. Both normal mice and SMA affected mice have green fluorescent protein (GFP) in all nerves, resulting in nerves that appear green under the microscope (Figure 2). Although the nerves are

visible under the microscope due to the presence of GFP, the receptors at the nerve synapses are not. Since the number of functional synapses in SMA affected individuals is of interest in Dr. Chien-Ping Ko's research, their visiblity under the microscope is important. Therefore, Verma utilizes an immunocytochemistry assay, a process that specifically adds fluorescent colors to the neurons of interest. For the purposes of this research, immunocytochemistry with ChAT-specific antibodies is used (Figure 3). This type of immunocytochemistry relies on the fact that acetylcholine (ACh) is the neurotransmitter that signals motor neurons to contract, and is therefore synthesized by all the motor neurons in the spinal cord and brain stem. ACh synthesis requires the presence of a specific enzyme known as choline acetyltransferase (ChAT), which is usually found at the axon terminal.

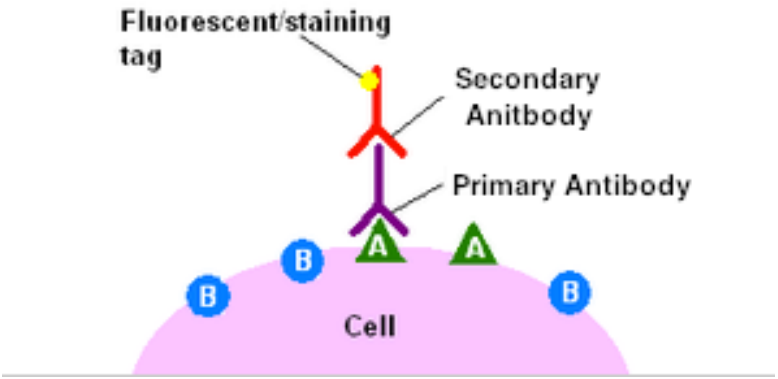

Figure 3 *Immunocytochemistry refers to the process of detecting antigens (usually proteins) in cells of a tissue section by using the principle of certain antibodies binding specifically to certain types of antigens in biological tissues. For the research discussed here, synaptophysin (primary antibody) binds to ChAT found in motor neurons (labeled A in the diagram). A secondary antibody with a fluorescent marker attached to it is then bound to the primary antibody*

Only cholinergic neurons, nerve cells that synthesize ACh in their terminals, contain ChAT, so this enzyme is a good marker for motor neurons responsible for muscle movement. In the immunocytochemistry process conducted by Verma, entire muscle cells are bathed in a solution containing an antibody called synaptophysin. By definition, an antibody is a specialized immune protein produced by the immune system in order to identify and attach to antigens, foreign substances that trigger an immune response. All complete proteins are antigenic, as are many bacterial and other polysaccharides. Therefore, since enzymes are proteins, synaptophysin antibody will bind to the enzyme ChAT, which acts as the antigen under such circumstances.

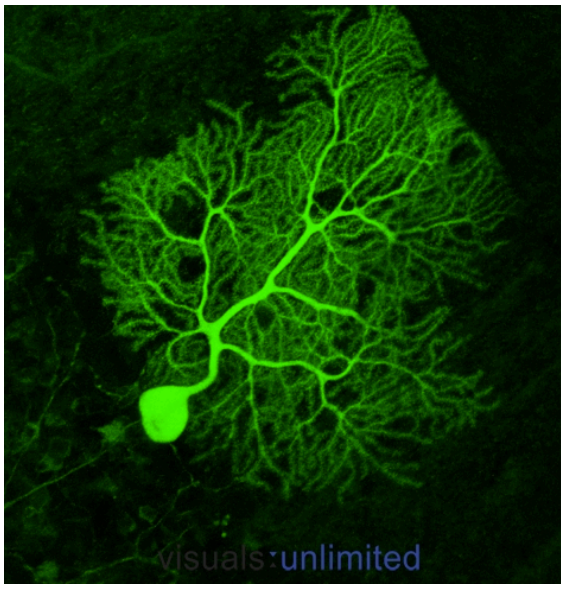

Figure 2 *The above is a picture of a neuron that contains GFP, resulting in the green color of the neuron when it is observed under the microscope. After immunocytochemistry for this research, there will also be red spots where ChAT is present*

This step in the immunocytochemistry assay marks the receptors of the motor neurons of interest. Next, a secondary antibody with a fluorescent marker is attached, making the receptors visible under the microscope. Through this process, receptors appear red under the microscope, while neurons appear

green due to GFP. These neuron samples are then observed and differences in normal and SMA affected neurons are noted.

Verma's results indicate that normal motor neurons have dendrites that reach all the way across the synaptic cleft, covering the entire receptor. However, motor neurons affected by SMA have dendrites that do not reach to the other end of the synaptic cleft. Oftentimes, only one or two dendrites of affected motor neurons actually reaches towards the receptor of a nearby neuron. If no dendrites are attached to the receptors of nearby neurons, then the neuron has undergone complete degeneration. A lack of dendrite attachment to receptors of nearby neurons means that the signals from the nervous system are not relayed from one neuron to the next, and the initial signal for muscle movement is not converted into a response, since the signal dies before it reaches the location where signals are converted into cellular responses. Therefore, the muscle does not move. Verma's results also indicate a lack of synapses in SMA affected neurons of muscle tissue in some parts of the body (Figure 4). This is supported by a lack of red fluorescent markers in SMA affected neurons observed under the microscope, which indicates the location of receptors in these neurons. These results suggest that for some muscles in the body of individuals affected by SMA, a lack of synapses might be the cause of reduced muscle control. Some time in the near future, Dr. Chien-Ping Ko's research may provide an answer as to what is causing the observed synapse loss. If the cause of synapse loss is identified, then a way to prevent synapse loss may be found. If a way to prevent synapse loss is found, then new medications can be created that can potentially treat SMA affected neurons in certain parts of the body. Therefore, the success of this research could have a huge impact in the medical science community. As Verma puts it, this research is an important step in the right direction. "It's for a great cause," says Verma, "and I'm glad that I can be a part of this amazing opportunity."

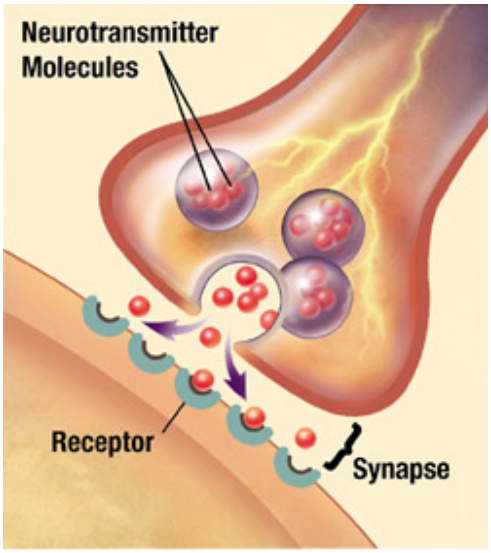

Figure 4 *The picture above depicts a neuron and its target cell zoomed in at the synapse. Due to the electrical signal relayed through the axon, the neuron releases neurotransmitters (in the case of motor neurons, the signal is for the movement of a muscle, and the neurotransmitters released would be ACh). Neurotransmitters then travel across the synapse and attach to specific receptors of the target cell. In SMA affected neurons in some muscles of the body, there is a lack of synapse*