Individuals at the National Partnership for Advanced Computational Infrastructure (NPACI) at the University of California at San Diego (UCSD) study shape analysis of microscopic data in neuroscience. The Cell Centered Database (CCDB) is a database developed by NPACI. It contains structural and protein distribution information from confocal, multiphoton, and electron microscopy, including correlated microscopy. The database query will be enhanced by shape analysis. Scientists are curious whether it is possible to classify dendrite shape automatically. At the Center for Imaging Science at Johns Hopkins University, we are currently developing methods to make this possible. The Large Deformation Metric Mapping (LDMM) software was developed for NPACI by Johns Hopkins University and has been of significant aid to the project. The LDMM software maps the differences in two given images by computing the metric distances between the two images. The images are compared, and various changes and differences are visualized. In the end we will develop a pipeline for the CCDB of all the information generated to achieve our goal.

My goal this summer was to conduct a thorough analysis of dendritic spines and eventually place them into a standard coordinate system. I first practiced on 2D images and later moved onto the analysis of 3D images. Dendritic spines are small protuberances that stem from the cell body, or soma, of certain neurons. They play a major role in receiving and processing excitatory synaptic inputs. The spines are not static structures; they extend and retract due to changes in activity and in response to environmental stimuli, often in a time frame of just a few minutes. The spines vary in size and morphology but usually are 1-4 µm in length and 0.1-1 µm in diameter. They usually end in a bulbous head that is attached to the dendrite by a narrow stalk or neck. Their distribution across a neuron is not homogeneous, so it is imperative to analyze numerous spines over a large area of the dendrite.

We analyzed the spines of a Purkinje neuron from a rat cerebellum. The image data was downloaded from the CCDB. The data set contained an image of the entire dendrite as well as 33 files of the spines in *.synu format. The spine parameters were measured using light microscopy and electron tomography. The dendrite had a volume dimension of 200.0 x 590.0 x 80.0 pixels, a resolution of 0.026 x 0.026 x 0.026 µm/pixel, and a magnification of 3000. The triangulated surfaces were to be visualized in a program called BrainWorks, which only reads files in the *.byu format. This required another student in the laboratory, Dominic, to create a program in Matlab to convert the *.synu files to the proper *.byu format. After this was done, we ran into some problems with the topology, such as holes or extra triangles on the surfaces, which were fixed by Dominic.

Once the surfaces were converted to the usable *.byu format and reconfigured so their topology was correct, I had to visualize each surface on the image to fit the
anatomy. Through this visualization, I also determined the neck, the narrow stalk that connects the spine to the dendritic shaft, and the head, the opposite, far end. I then placed 20 landmarks on each of the 33 surfaces. Certain areas that stood out were targeted, as if the eyes, nose, lips, and ears would be pinpointed on a face. I marked the neck, the head, certain points that either protruded in or out of the spine, certain points that were vertices of rather large triangles, and certain points that were vertices of numerous triangles. The landmarks were placed precisely at the same area on each surface, from neck to head, and in three levels from top to bottom which I labeled as "upper," "medial," and "lower."

Each of the 33 surfaces was then to be placed in a standard coordinate system measuring 64.0 X 64.0 X 64.0. This required that the centroid of each surface be at (32.0, 32.0, 32.0). With Dr. Tilak Ratnanather's help, I developed a Matlab program to change the coordinates of each surface so the centroids would all be at the required coordinates of (32.0, 32.0, 32.0). The coordinates were altered while the vertex number was maintained, which provided us with valuable reference points for the new surfaces. Each surface was still rotated the same and the topology stayed the same; it was strictly the coordinates that were altered. The landmark files were also converted to fit the new surface in the standard coordinate system, and this was completed by a third student, Russell.

Once the standard coordinate system was achieved our goal was to rotate the surfaces so that they all lined up precisely on the same axis. We wrote a program in Matlab using a rotation matrix to generate the new coordinates. For each surface, we used the coordinates of the neck and head from the landmark files to calculate the angle that the surface needed to be rotated in order to line up with the axis.

I learned a great deal this summer and got a lot of good experience out of the internship program. All of the small steps necessary to reach the end goal were extremely valuable. We dealt with a very specific subject, the dendritic spines of Purkinje neurons, as opposed to typical classroom learning where students barely brush upon general topics. Besides spending a good amount of time researching the anatomy of dendritic spines, I also improved my computer and programming skills. I was forced to develop ideas and methods myself rather than being told every step I needed to take, and this has been very beneficial to my theoretical skills. Getting the opportunity to work in a research setting has allowed me to get a sense of the type of work and environment I may end up in the future. I got the opportunity to meet several other individuals not just in the lab I worked in, but also within the entire Center for Imaging Science at Johns Hopkins University. It was very interesting to hear what everyone had to say about the field they were pursuing, what led them to it, their future goals, etc. In addition, I was also given some very valuable advice when considering my future plans.

I am extremely fortunate for this excellent summer internship opportunity. I know the skills and knowledge I acquired will aid me tremendously in the next few years, and many more after that.