In vivo volume analysis of biomaterials using photoacoustic imaging (PAI)

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WeARE Research Area

Dr. Brey has given me the opportunity to work in Photoacoustic imaging project. I worked under the mentorship of Dr. Shrestha. The project was to identify and monitor biomaterials in vivo using photoacoustic imaging. I helped her with image analysis of a hydrogel in vivo and look at how the gel degrades in the mice by observing the photoacoustic images. She allowed me to shadow her on how she was able to obtain the images and taught me how to use ImageJ to do the analysis.

Motivation or Background

The research project is based on using photoacoustic (PA) imaging to monitor biomaterial following implantation over the course of 8 weeks in vivo. PA is a bybrid imaging technique that uses both optical and acoustics to generate an image. Since observing biomaterials in the body can become difficult and many other imaging techniques could harm the patient, PAI is being used as a tool to investigate/evaluate biomaterials in vivo. was involved in image analysis of photoacoustic images to measure volume of implanted biomaterials. This will be achieve using Fiji (image analysis software) to quantify vascular and biomaterial parameters. I will also contribute to the preparation and staining of histological slides of harvested tissues for analysis. This project will provide insight into the use of PA as a noninvasive tool to assess biomaterials following implantation. I will be exposed to concepts of biomaterials, photoacoustic imaging, image analysis and histological staining. The motivation to do this is to get a hands-on experience and an expectation on how a future job will be like. This also allows me to decide what concentration I want to do in the biomedical engineering field

Objectives

1. To observe and monitor the changes in the hydrogel over the period of 8 weeks. Evaluate change in volume of hydrogel by obtaining volumetric data of hydrogel at each week. The volume is obtained by adding all the areas of each sliced image and multiplying the sum they the slice size.
2. Compare the normalized volumetric data obtained from ImageJ software with the data generated from the ViewMSOT software. The volume of hydrogel at each week was normalized to week1 volume.

Methodology

The methodology of the research is quite simple, but very time consuming. When obtaining the images, there is a peak the signal intensity is maximum. Considering this slice as the middle cross-section, 4.5 mm on each side of the hydrogel was included for volume measurement since the actual diameter was ~9mm. An important part before image analysis occurs is setting a set scale so the area is accurate. Once getting that range and setting the scale, the image analysis occurs, by finding the area of each slice. After finding all the area of all the slices the sum will be obtained and multiplied by 0.33 mm due to the size of each size to find the volume of the gel. The volumes are then compared to other groups, samples, and type of gel being observed. Then the normalization will take place, and the mean of all the samples normalization will take place so the standard deviation can be found to observe the margin of error in each observation of images.

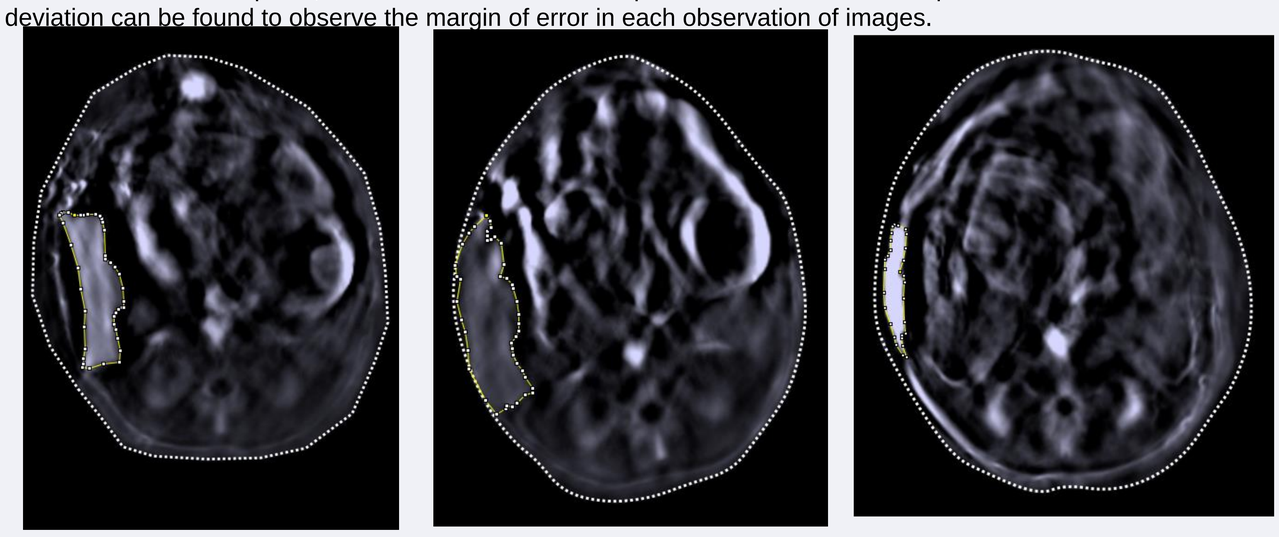
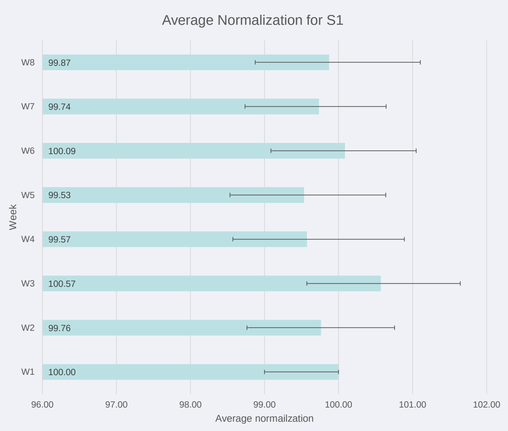
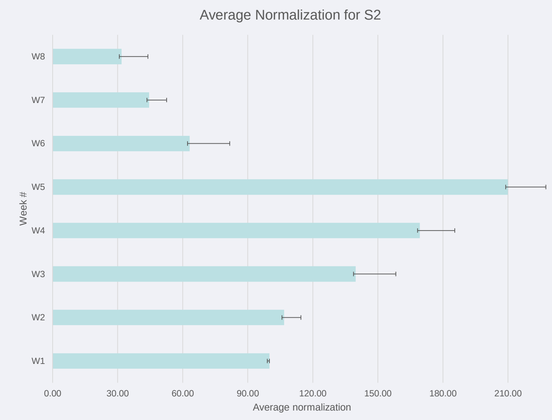


Fig. 1: Images of hydrogel in Week 1 (left), 4 (middle), and 8 (right) for both S2

Results



Graph 1a: Normalization average (mean) for S1 with error margin



Graph 1b: Normalization average (mean) for S2 with error margin. (note that the error margin is larger but seems small due to scale)

When doing image analysis, it is very important to get little to no background space in the object you are observing. Therefore, when getting the area for the hydrogel, selecting only the gel is key and avoid getting the arterial wall and background space. Since hydrogels expand before degrading G2M1S2 data will be very different. As seen in figure 2, G2M1S1 has a consistent volume, fluctuating ±2 mm^3 making the normalization to be very little. In G2M1S2, the gels expanding phase being in the third week and lasts till the fifth week. These trends have been common for all the samples. But S2 was much more difficult to be accurate due to the gel size increasing and hard to differentiate if a certain outline is part of the hydrogel or not, making S2 samples have a high margin error compared to S1 samples.

Skills and Experience

The experience in undergraduate research is highly recommended due to the amount of information and hands on experience obtained. The skills I have learned was how to use ImageJ to analyze images and to present data properly due to the weekly lab meetings, where they provide constructive feedback about experiments or the methods of data analysis. This is a good experience to see how research and labs function and the basic principles required for such a job in the future. .

What I Learned

I have learned many things within this research lab such as the use ImageJ to do image analysis, how to obtain the images by observing the protocol on PAI, and how histology (H&E staining) is done. I also learned how to observe PAI images and how the slicing of the images work. I've also learned more properties about hydrogels and the degradation of them before learning about them in my biomaterials class. This also showed me how broad the biomedical eningeering field is and how the labs allow you to see how a concentration would be before choosing that path.

Future Plans

To quantify the vascularization surrounding the hydrogel in vivo and examining the local tissue response using H&E staining (histology) and do analysis on the stained slides.

Acknowledgments

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References

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