

## Introduction

Bone is one of the most essential parts of all vertebrate animals, it holds them up, helps them move and protects other vital organs from damage and trauma. It is an area of research that is continuously advancing to improve the quality of life of people on Earth. Although bone has been studied extensively on the global level, more research has to be done on the local level, which can impact the properties of the entire bone. Advanced nano-CT scans will enable the future analysis of bone at the local level by looking at its microstructure and material properties. This study investigated how to implement image processing techniques to convert large compressed files to enhanced digital images and 3D models that can be visualized and operated on by FEA and other mechanical modeling techniques.

## Methods

### Image Acquisition and Processing

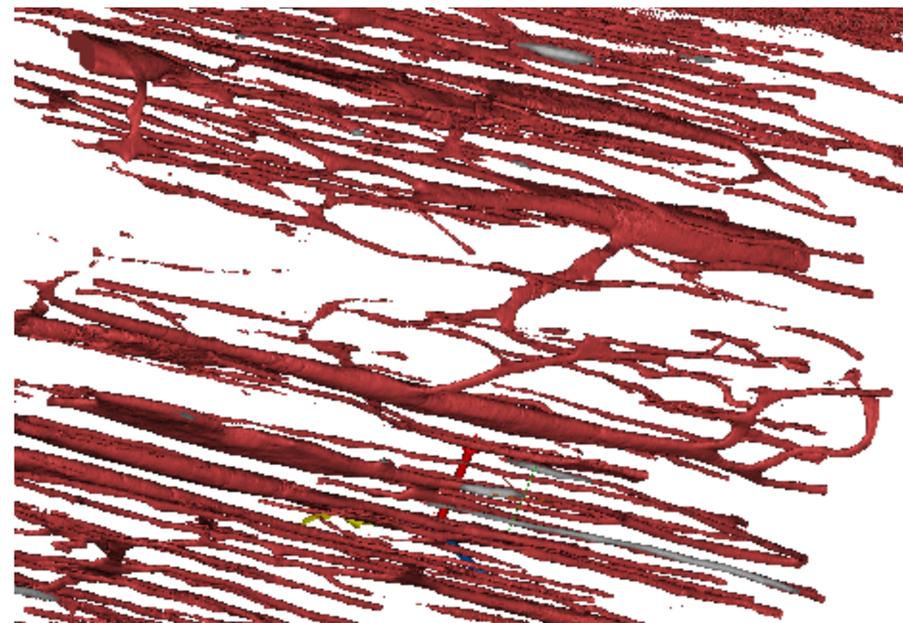
In this study, nano-CT scans of herring bone at the 100nm and 30nm level were done and the output of the scans was in the form of .VOL files. This is a compressed volume file extension that stored 33GB of data for each scan. The first images were acquired and analyzed by importing the .VOL files into the image processing software ImageJ. Then, the files opened in ImageJ were exported as a stack of 2048 slices across the bone. Using ImageJ the x,y,z and pixel values at different regions on each slice were identified. The pixel values represent the density value at a specific region. Bone has the maximum density value, while pores and air have the minimum density values. The values were mapped using a formula relating attenuation to bone density and were used to convert the TIFFs to DICOMs. The DICOMs were imported into MIMICS and were processed using morphological operations and segmentation techniques to separate the pores from the bone and create a 3D model of the pores.



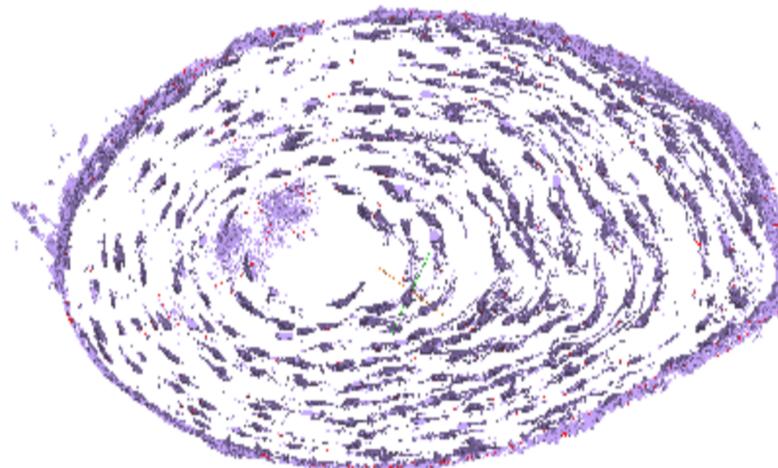
**Figure 1:** First image acquired of the nano-CT bone scans. The dark colors indicate bone and the white colors between the dark colors indicates pores.

## Results

The result of the image processing was multiple three dimensional models generated for a significantly large set of digital images. This enabled the visualization of the intramuscular bone structure and the analysis of the pores and the patterns created by the pores in the different bones. Each network of pores is unique and can be analyzed to show how it affects the material properties of the bone. The images generated are at a scale that has not been seen before and show the specific pores left by the cells, and not just the structural pores, at an incredibly detailed level.



**Figure 2:** A cross-sectional view of the 3D model generated of the pores within the bone.



**Figure 3:** An axial view showing a 3D model generated of the pores within the bone.

## Discussion

### Application of Generated Results

The models generated by this study are significant as they can help in looking at the microstructure of bone. At the bulk level, which is the centimeter scale, the elastic modulus is related to density but at the tissue level, which is the micrometer to millimeter scale, the relationship between elastic modulus and density is less significant. On the other hand, structural components in the microstructure of the bone, such as porosity and mineral crystal organization, significantly impact the mechanical properties of the bone (1). Additionally, diseases like chronic kidney disease (CKD) impact the microstructure and material properties of bone resulting in reduced bone quality (2). Therefore, the techniques in this study could be applied by trying the newly established nano-CT image processing framework on CKD and other bone diseases that cause micro-porosities.

### Current Data and Future Studies

This project challenges and seeks to shift current research by applying micromechanical modeling to a large data set of nano-CT scan data and in addition to quantifying bone density also analyzing the relationship between density and material properties such as elastic modulus and looking deeper at other structural components in bone. These models can be exported as voxel meshes and can be opened in Finite Element Analysis (FEA) software such as ANSYS and can be analyzed using FEA or more advanced micromechanical modeling techniques. Using this analysis, a relationship between pixel value and elastic modulus can be established. Then, the geometries and properties of the individual components can be used to predict the properties of the overall heterogenous bone material.

## Conclusions

The scanned images and .VOL files were opened and used to generate TIFFs and DICOMs. An image of the bone was visualized using ImageJ. A relationship was established between density and attenuation based on the raw data, and the large data sets of pixel values in ImageJ were converted to attenuation values and imported into MIMICS. A 3D model of the image stacks was created and processed to visualize the appearance of the pores and microstructure of bone. Overall, this study established the image processing work flow for nano-CT scans.

## Acknowledgements

Thank you to The College of New Jersey's Mentored Undergraduate Summer Experience (MUSE) program for providing the opportunity to perform this study.

## References

- [1] Nobakhti, S., & Shefelbine, S. J. (2018). On the Relation of Bone Mineral Density and the Elastic Modulus in Healthy and Pathologic Bone. In *Current Osteoporosis Reports* (Vol. 16, Issue 4, pp. 404–410). Current Medicine Group LLC 1. <https://doi.org/10.1007/s11914-018-0449-5>
- [2] Heveran, CM, Schurman, C, Acevedo, C, Livingston, EW, Howe D, Schaible, EG, Hunt, H, Rauff, A, Donnelly, E, Carpenter, RD, Levi, M, Lau, AG, Bateman, TA, Allison, T, King, KB, Ferguson, VL (2019) Chronic Kidney Disease and Aging Differentially Diminish Bone Material and Microarchitecture in C57Bl/6 mice. *Bone* 127:91-103.