Mechanical Properties in Radial Bones of Polypterus senegalus

Sakib Kazi, Emily Standen

University of Ottawa, Faculty of Science, Department of Biology

Introduction

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Polypterus senegalus are an air breathing fish with the ability to leave water and walk between pools. When walking on land, the fin rays and radial bones in the pectoral fins experience drastically different loading conditions compared with swimming; leading to the question of how these changes will influence the bone structure in radials.

The **mechanical properties** of bone result from the mineral and collagen components.

The **hardness** of a material is defined as its resistance to penetration by an indenter. Microhardness refers to indentations less than 30µm and 200g of load.

Purpose

By testing the microhardness values along the length of the radial bones present in the pectoral fins, the goal of this study is to determine if there is a correlation between the hardness values and the position on the length of the radial. We hypothesize that there will be variation in hardness along the radial bone and that there will be a correlation between radial position and microhardness.

Results

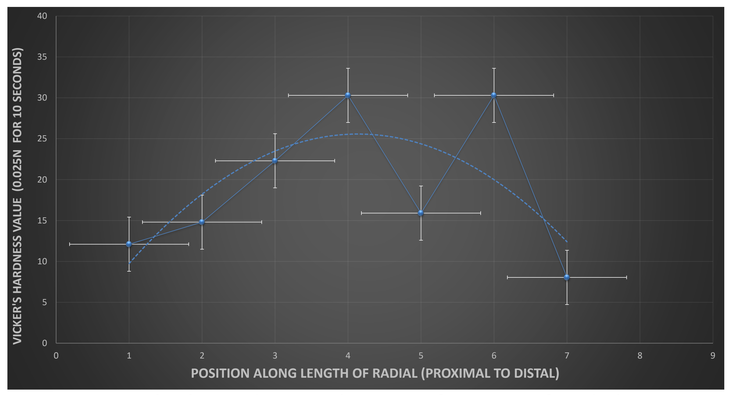

Figure 3. Vicker's Hardness Values as a function of Radial Position

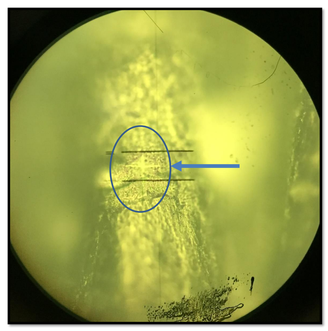

Figure 4. + shaped Microindentation mark on surface of radial bone.

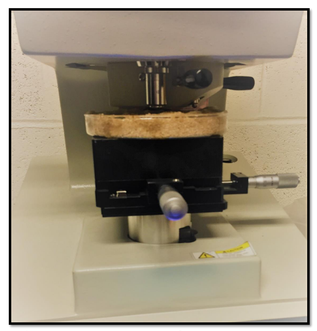

Figure 5. Microindentation Set up in process of taking measurement

Conclusion

Based on the results, given the limitations of the of the experimental design, the preliminary data shows decreasing micro hardness values towards the proximal and distal ends of the radial bone, with the highest micro hardness values present in the central region.

The limitations of this experimental design are primarily due to the miniscule size of the radial bones. Laying the fin upon the resin does not present if as a sufficiently flat planar surface for microindentation measurements. Furthermore, some portions of the bone were coated with thin layers of resin, thus affecting the microhardness readings obtained.

Future Experiments

In the future, experimental setup will be redesigned in order to cater more to the size of the radial bones. This will be done by embedding the fin in LR White resin between two microscope slides. This will ensure a planar fin which will allow for more accurate micro indentation measurements. Furthermore, as the sample will be coated in resin, it will be polished to go past the ossified surface and into the cartilage which makes up the majority of the radial bone. As seen in the image below, the ossified regions make up a very small portion of the overall bone, leading us to believe that majority of the material properties are a result of the are a result of the cartilage. This allow for more extensive data collection to further investigate variation in hardness across radial bones in a large sample.

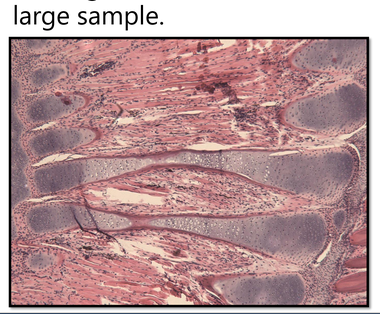

Figure 6. Section of Polypterus senegalus fin depicting cartilage interior of radial bones

Methods

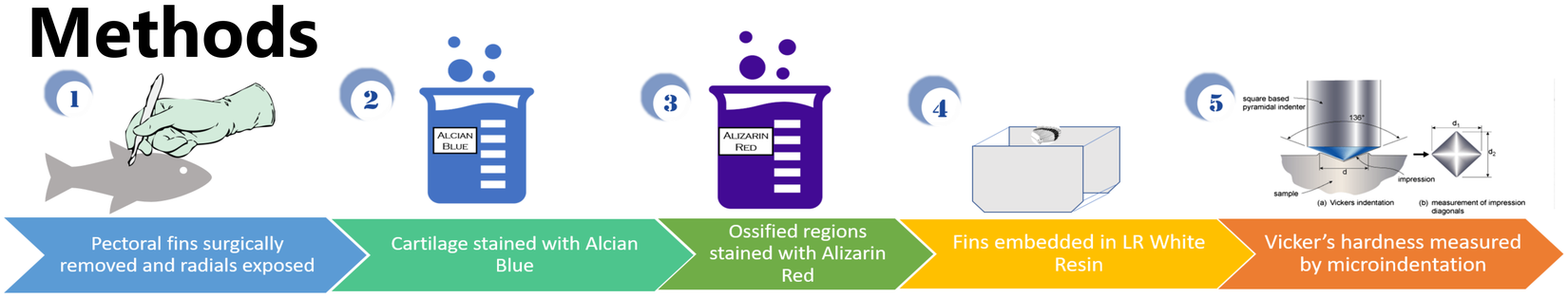

Figure 1. Steps in Experimental Procedure

Protocol Design

Step in Methodology	Development of Methodology Through Trials
Step 1	Trial 1: In order to place the fins in the resin block, they needed to first be carefully dissected from the fish without damaging the bones.
Step 2	Trial 2: Due to uniform colouration, the position of the radials within the fin was difficult to determine. Staining with Alcian blue highlighted the cartilage on both ends of the radials.
Step 3	Trial 3: The translucent fins were not visible once placed in the resin. Alizarin red was used to stain the bones making them easy to visualize under the scope of the microindenter
Step 4	Trial 4: Due to the thin ossified surface, polishing the resin would run through the bone. A platform was built to lay the fins upon the resin thus no longer requiring polishing.
See Future Experiments	Trial 5: As the samples were placed onto the resin, the radial bones were not perfectly in plane, increasing the difficulty of micro hardness measurements.

Figure 2. Development of Experimental Procedure Through Trial Tests

Bibliography

1. Song, J. and L.R. Parenti, 1995. Clearing and staining whole fish specimens for simultaneous demonstration of bone, cartilage, and nerves. Copeia 1995(1):114-118.
2. Evans, G.P., Behiri, J.C., Currey, J.D. et al. J Mater Sci: Mater Med (1990) 1: 38. doi:10.1007/BF00705352
3. Avci, Utku, and Jin Nakashima. "A Flat Embedding Method to Orient Thin Biological Samples for Sectioning." *Methods in Molecular Biology Plant Gravitropism* (2015): 13-22. Web.

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