Visualizing Compound Action Potential Propagation With High Frequency Ultrasound

Emma M. Glass, Shani E. Ross, Parag V. Chitnis
George Mason University, Krasnow Institute, Department of Bioengineering, Fairfax, Virginia

Abstract

Current methods used in electrophysiological recordings either require invasive high resolution electrodes, or non-invasive low resolution electrodes. Therefore other direct, non-contact methods for determining neural activity are currently being explored. Previous research investigated methods using high resolution optics to determine neural activity, but these recordings are complex. This study aims to use high frequency ultrasound (HFU) as a simple, non-invasive, high resolution imaging method to visualize neural activity in the ventral and walking leg nerves of the lobster, *Homarus americanus*. Previous investigations have shown a possible correlation between action potential propagation and a physical change in neural diameter by several micrometers. By combining electrical stimulation with HFU and electrical recording, physical changes in the diameter of the lobster nerves should be observable. Ventral and walking leg nerves were exposed in an anesthetized lobster. Using stainless steel electrodes, the action potential propagation through the ventral cord in the anatomy could be recorded electrically and analyzed. These electrical recording methods will provide validation for action potential propagation when used in conjunction with HFU imaging. This procedure for visualizing neural activity could be used in the future on other in vitro and in vivo models, eventually leading to a better alternative to today's electrophysiological recordings.

Introduction

Background:

- Electrodes used in electrophysiological recordings currently are either invasive or low resolution.
- Optics with complex methods have proven to be a high resolution non-invasive method for determining neural activity.
- A previous study has determined that nerves change in diameter with propagating compound action potentials (CAP).
- Ultrasound could be used to image a nerve with a propagating CAP to visualize the physical diameter change of the nerve.

Objectives:

1) Stimulate a lobster nerve with an electrical pulse and record evidence of compound action potential propagation down the length of the nerve.
2) Characterize the conduction velocity of the lobster nerve being used.
3) Find an effective configuration for using a high frequency ultrasound (HFU) transducer to image a lobster nerve.
4) Use electrical stimulation and recording in conjunction with HFU imaging to visualize change in diameter of a lobster nerve, which suggests propagation of a CAP.

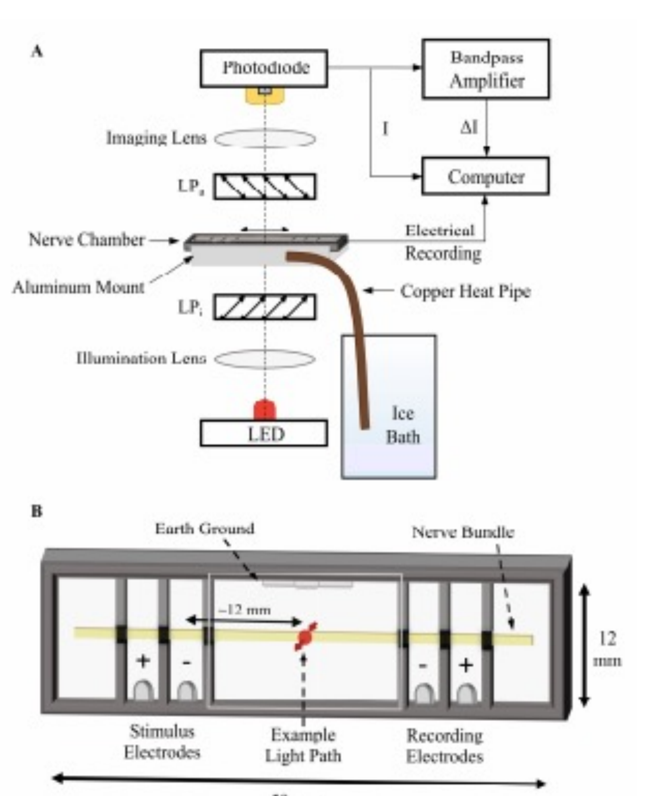


Figure 1. Complex Optical Recordings to Visualize Action Potential Propagation by Wininger et al. 2009.

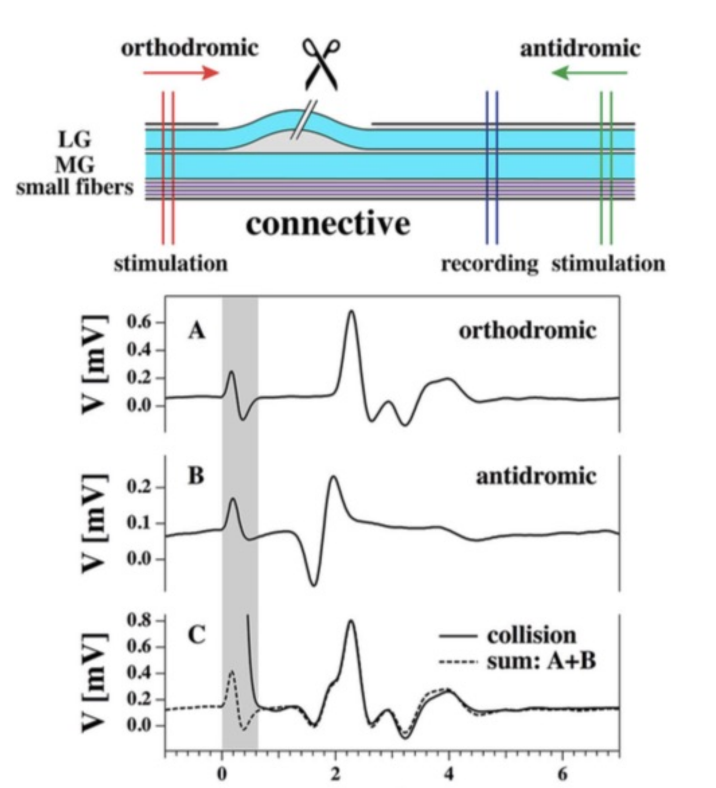


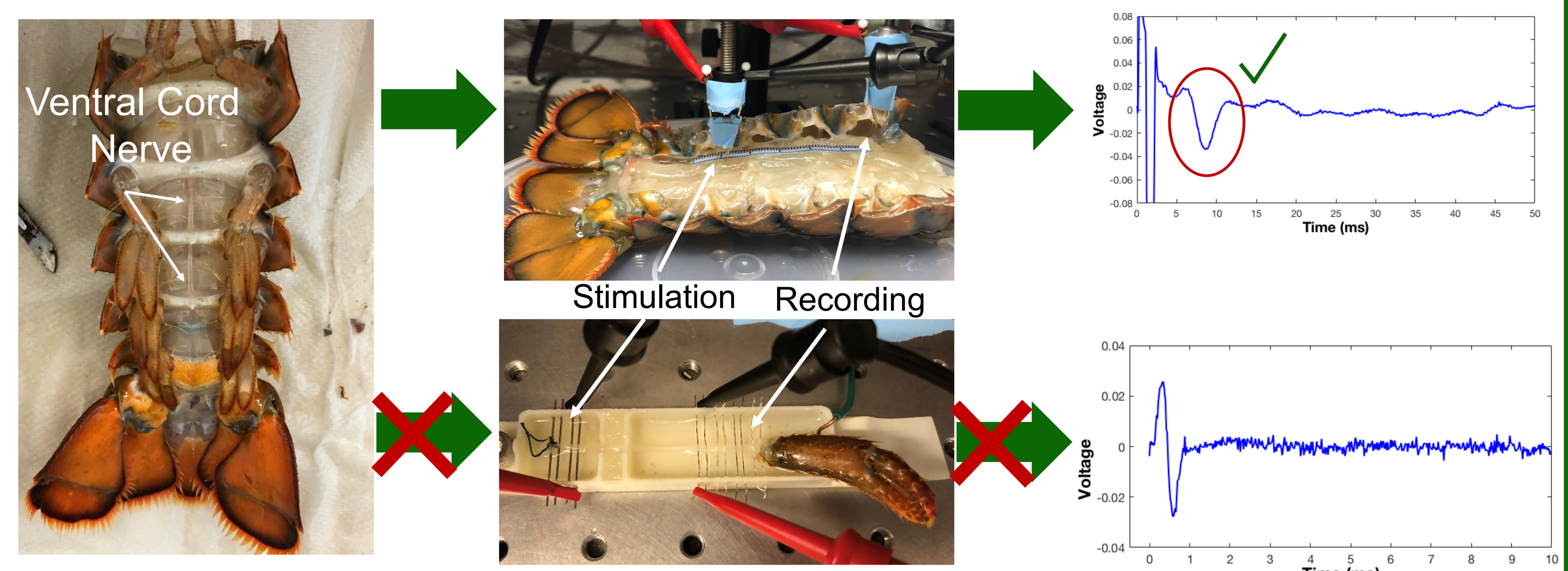
Figure 2. Antidromic and Orthodromic Action Potentials Recorded Electrically by Gonzalez-Perez et al. 2016.

Methodology

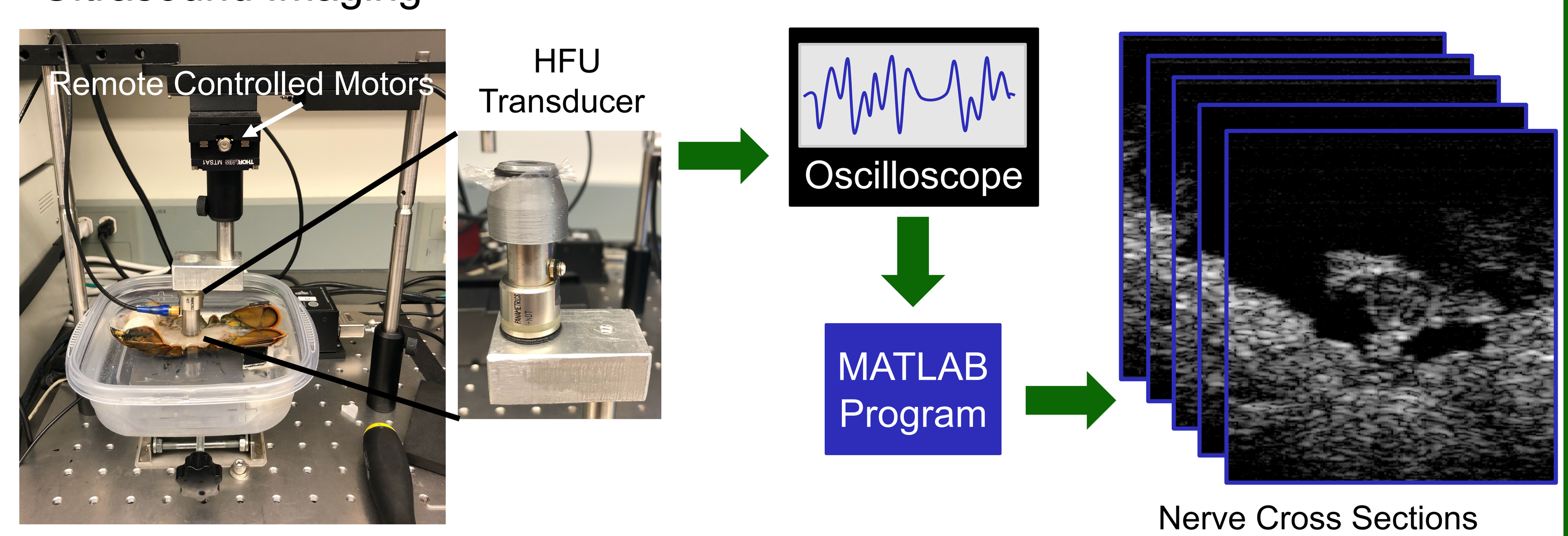
Equipment Configuration

Pulse Stimulator → Nerve → CAP Recording → 1000x gain, Band Pass Filter → Oscilloscope

Stimulating and Recording CAP



Ultrasound Imaging



Signal Recording Parameters

Table 1. Compound Action Potential Recording Parameters

Nerve Type	State	Filter	Stimulation Pulse Amplitude	Stimulation Pulse Width	CAP Recorded***
V.C.*	Chamber	10-3kHz	1, 5, 10 mA	0.2, 1 ms	No
V.C.	Anatomy	5-3kHz	1.5, 4, 5 mA	1 ms	**Yes**
W.L.**	Chamber	10-3kHz	10 mA	0.25, 1 ms	No
W.L.	Anatomy	5-3kHz	1.5, 4, 5, 10 mA	1 ms	No

*V.C. = Ventral Cord
**W.L. = Walking Leg
***CAP = Compound Action Potential

- All signals amplified 1000x (60 Db gain)
- Stainless steel needle electrodes to penetrate nerve
- Stimulation pulse amplitude was increased if muscle contractions in tail anatomy became weaker
- CAP signal recorded when ventral cord nerve was not dissected

Results

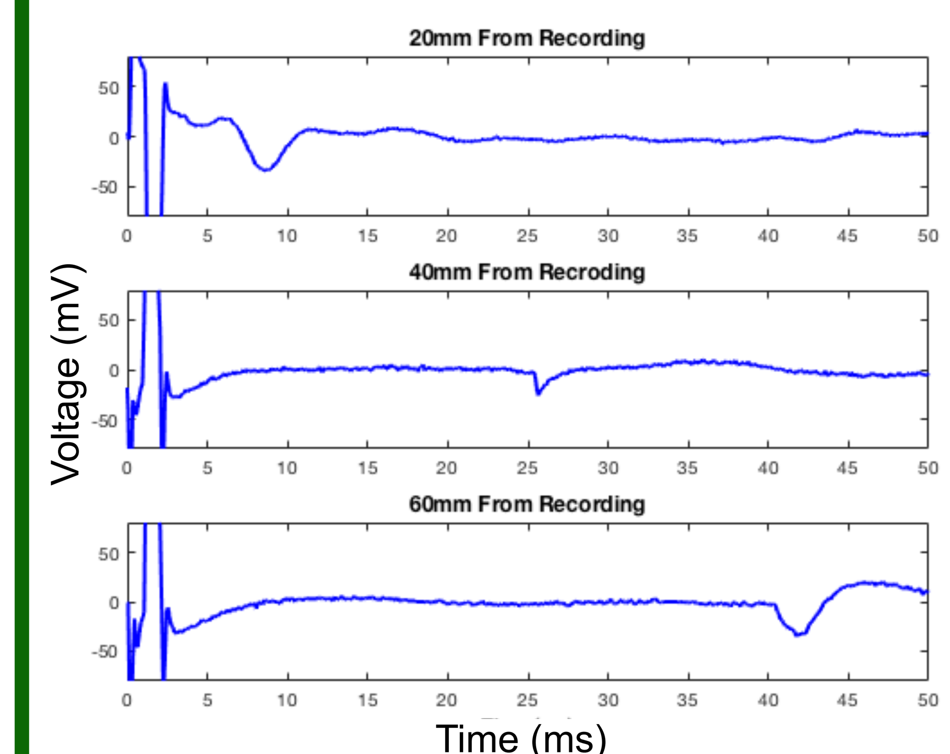


Figure 3. Propagating CAP Signals

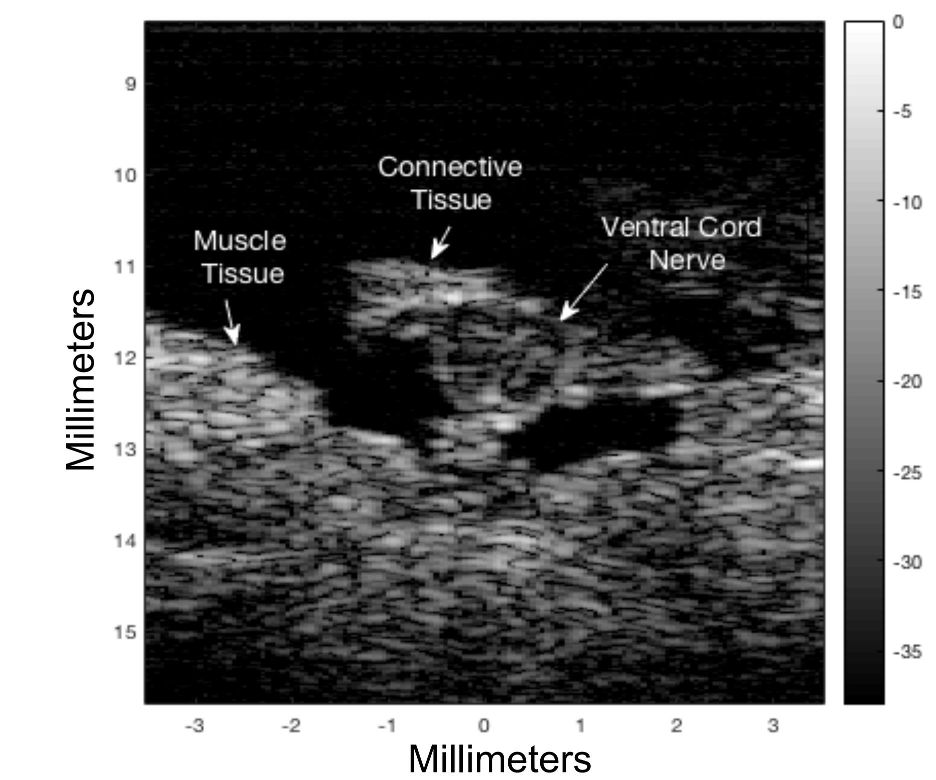


Figure 4. Cross Sectional Nerve Image

Table 2. Conduction Velocities of *Homarus americanus* Ventral Cord

Distance from Recording Electrode	Trials Averaged	Conduction Velocity
20 mm	5	2.35 m/s
40 mm	5	1.56 m/s
60 mm	5	1.43 m/s

Results:

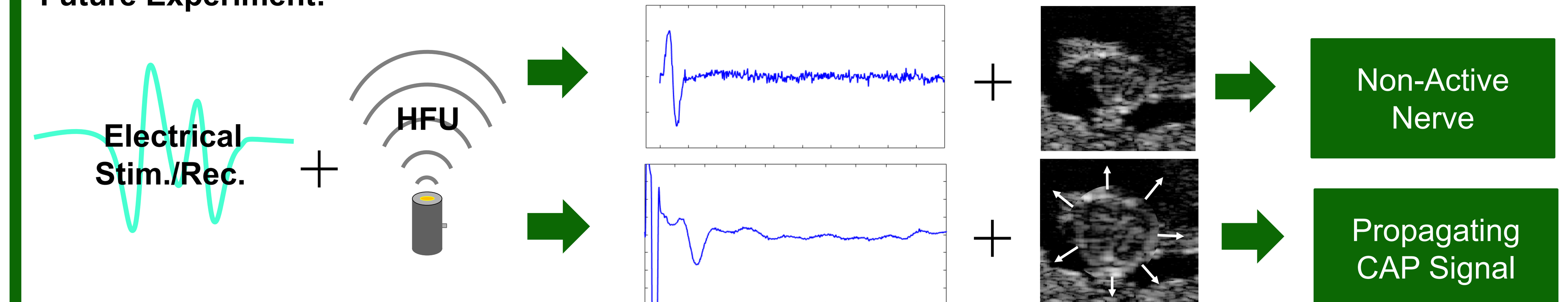
- CAP signals recorded at different distances away from recording electrodes
- Conduction velocities calculated using $\frac{\textit{Disance traveled between stim. and rec.(mm)}}{\textit{Time taken to travel between stim and rec.(ms)}}$
- High resolution HFU image of nerve created

Conclusions & Implications

The **purpose** of this study was to use high frequency ultrasound (HFU) recordings as a high resolution, non-invasive method to visualize neural activity in a lobster model. The **conclusions** of this study are as follows:

1) We were able to successfully stimulate and record a propagating action potential down the length of the ventral cord nerve. Ultimately in the next phase of this study, these electrical recording methods will be used in conjunction with ultrasound imaging to visualize neural activity.
2) The conduction velocity of the lobster nerves were characterized, which will be useful when pairing HFU imaging with electrical recording to verify neural activity temporally.
3) A lobster ventral cord nerve was imaged successfully using HFU, revealing a detailed cross-sectional image. A change in diameter of the nerve due to CAP propagation could be detectable by such a high resolution cross-sectional image.

Future Experiment:



Acknowledgments

We would like to thank Dr. John R. Cressman for providing access to the electrical stimulation and recording apparatus and salts to create a saline solution. We would also like to thank the ASSIP program for providing us with general lab supplies used in this study.