Characterizing the Effects of Glass Adhesives on Plated Fluorescent Beads

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Introduction and Background

This project sought to create a calibration tool for an automated electrophysiology system. This system utilizes a laser to locate fluorescence, and can be used to study deep-brain activity. To function, this system must be calibrated using fluorescent beads that mimic the appearance of fluorescent-dyed neurons. By understanding the glass adhesives and the fluorescence of the beads under various conditions, a sustainable, effective, and reusable tool can be developed. This research tested both the quality of curing and the extent of photobleaching that occurred when the adhesives and fluorescent beads were exposed to varying amounts of longwave UV light (320 nm-380 nm).

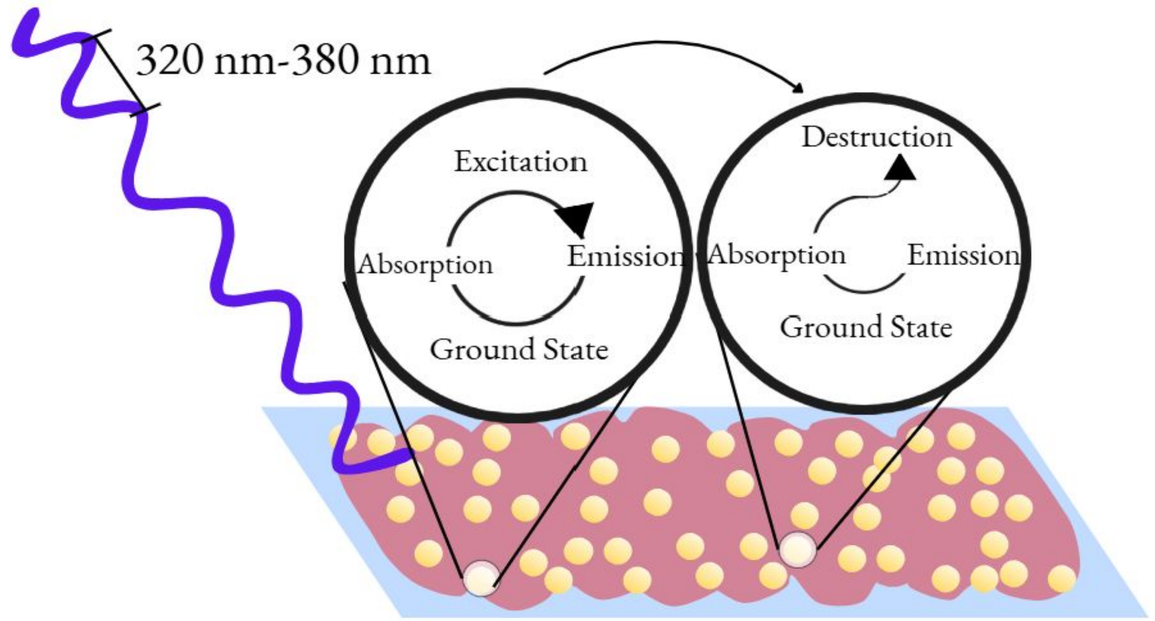


Fig. 1: Illustration of the cycle of fluorescence; destruction of this cycle is known as photobleaching

Materials and Methods

Fluorescent beads are polyethylene microspheres that emit photons when excited by UV light. They are used to calibrate and align laser systems. The adhesives used in these experiments were NOA 68, NOA 81, and Poly-L-Lysine. NOA 68 and NOA 81 are UV-curing adhesives used to bond glass to plastic, while Poly-L-Lysine is a water-soluble adhesive mainly used in cell culture and molecular biology. These materials were adhered to borosilicate microscope slides. Adhesive strength (Experiment **A**) was quantified by the ratio of beads adhered to beads lost after the slides were washed with distilled water. Similarly, the aqueous adhesive strength (Experiment **B**) was determined by the ratio of beads adhered to beads lost after slides were soaked in distilled water for ten minutes. To determine the brightness of the beads after various UV exposure times (Experiment **C**), beads were imaged under shortwave UV light (200 nm-280 nm). This light caused the beads to emit photons, which were then measured and compared to the brightness of fresh beads with no photobleaching.

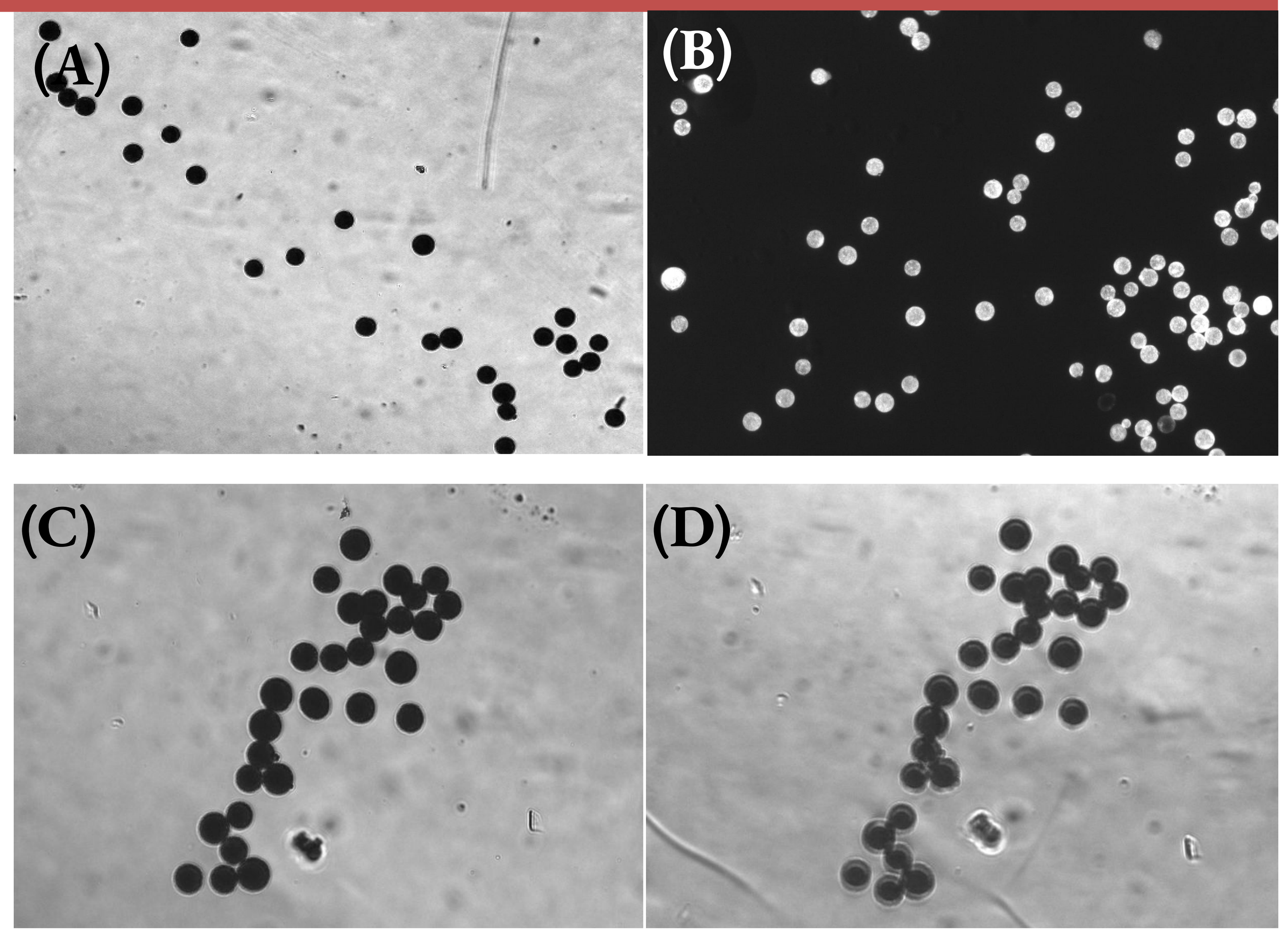
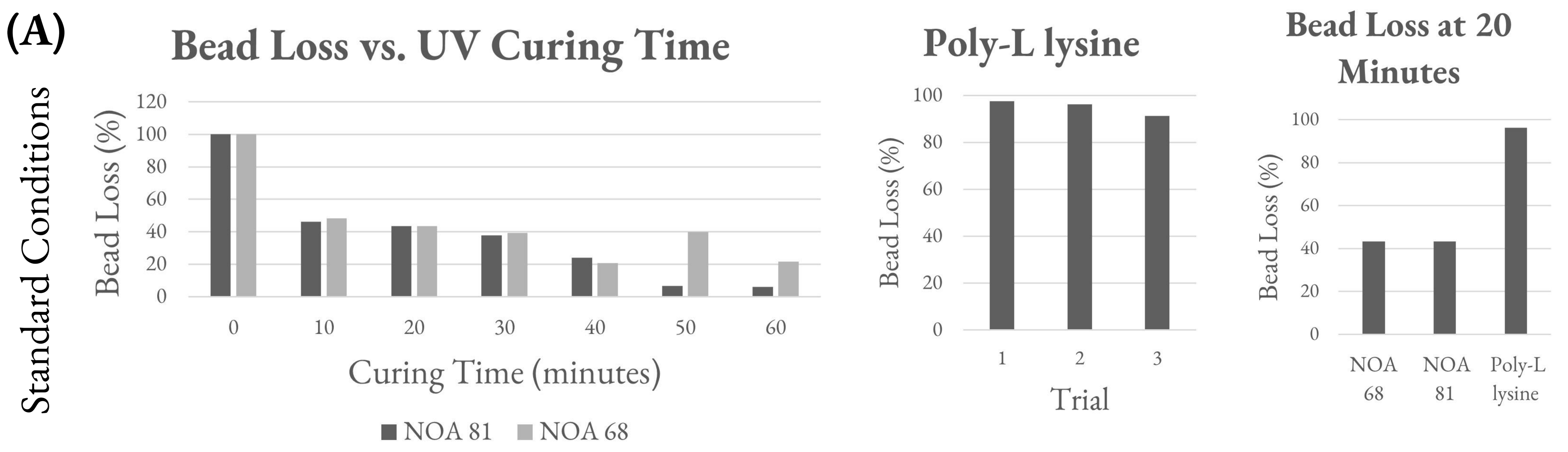
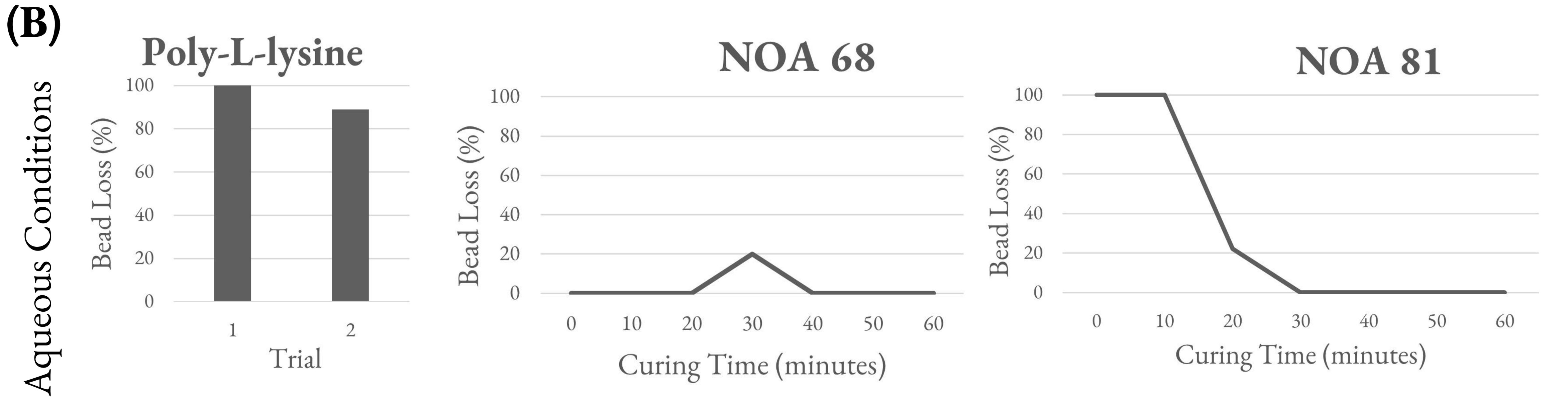


Fig. 2: Shown are four images of fluorescent beads. **(A)** Image taken without UV light. **(B)** Image taken under shortwave UV light. Images **(C)** and **(D)** show the same slide before and after soaking in distilled water.

Results





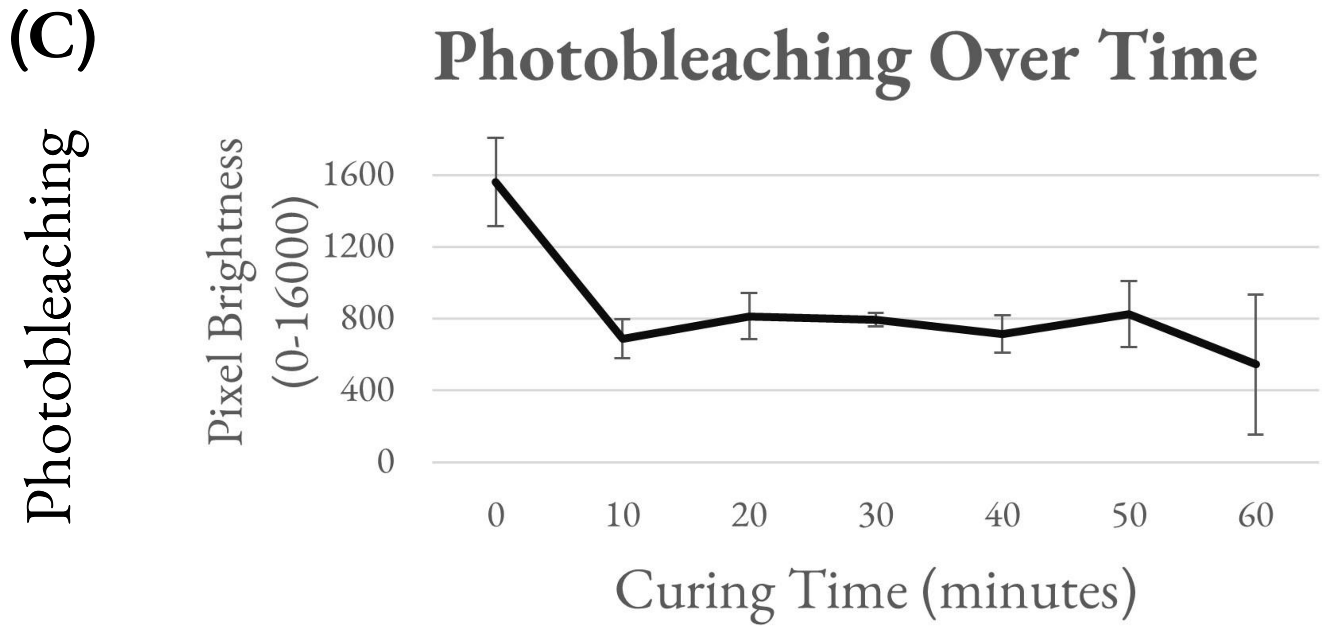


Fig. 3: Experiment **(A)** shows that bead loss decreases with increasing curing time. Experiment **(B)** suggests that bead loss in aqueous conditions decreases with increasing curing time. Experiment **(C)** shows that the majority of photobleaching occurs in the first 10 minutes of curing, then remains relatively constant with further exposure.

Conclusion and Future Work

The data presented suggest a variety of strengths and weaknesses in each adhesive. NOA 68 retained the highest percent of beads when exposed to aqueous conditions, while NOA 81 had the highest retention rate in standard conditions. Although Poly-L-Lysine was the weakest adhesive in both conditions, its ability to cure without UV exposure means that beads can be adhered to borosilicate slides without undergoing photobleaching. Further experimentation should be conducted to observe adherence under other variables in order to determine the optimal structure for the calibration tool.

References:

[1] "Poly-L-Lysine Cell Attachment Protocol." (2025) .sigmaaldrich.com.

[2] "UV-Curing Optical Adhesives." (2024) Thorlabs.com.

[3] "Fluorescence Fundamentals - US." (n.d.) www.thermofisher.com.

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