Analysis of Dendritic Spine Density in Hippocampus Following Cardiac Arrest

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Background

- **Cardiac Arrest:** Cardiac arrest (CA) is a condition in which the heart's electrical system malfunctions, leading to inadequate blood flow to the body. Cardiopulmonary resuscitation (CPR) is performed to reverse CA.
- **Anatomy:** The hippocampus, specifically CA1 neurons, is the area that is affected during hypoxic events (fig. 1). Memory and learning occur here.

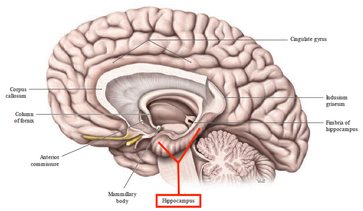
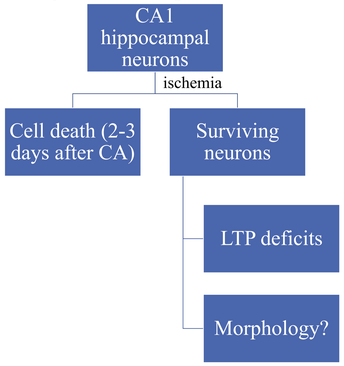


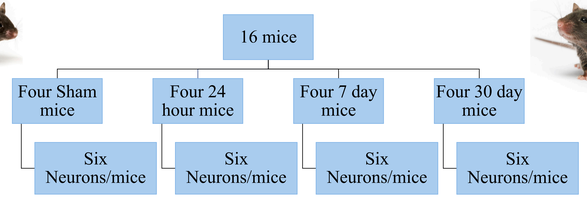
Fig. 1: Lateral view of the brain. The hippocampus (red lines) is located in the medial temporal lobe.[1]

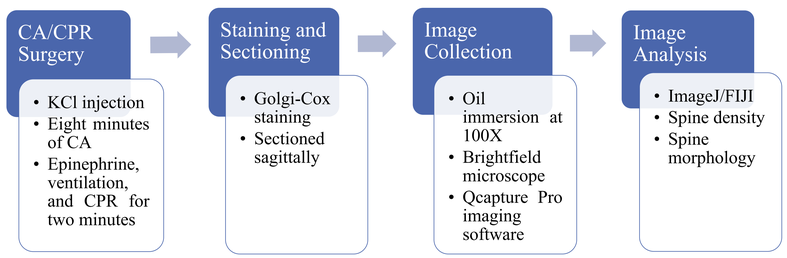


- **Rationale:** Reduced dendritic branching is shown at two to three days after an ischemic event (acute time points), yet little is known about the morphology of surviving CA1 neurons at later time points.
- **Goal:** To determine the structural plasticity of CA1 hippocampal neurons at longer time points after cardiac arrest.

Hypothesis: CA/CPR induces structural changes to dendrite morphology in acute and longer time points that correlate with hippocampal LTP deficits.

Project Approach





Dendritic Spine Analysis

- Neurons were chosen for their pyramidal soma shape, continuous dendrites, and clear imaging (fig. 2). Apical (both proximal and distal) and basal dendrites were chosen (fig. 3), and their spines were categorized (fig. 4).

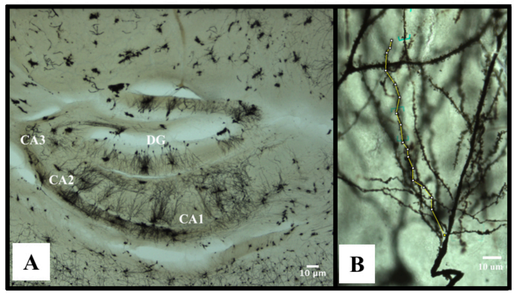


Fig. 2: A Golgi-Cox staining of the hippocampus of a mouse brain. (A) shows the Golgi-Cox stained hippocampus at 40X with all the areas of the hippocampus, including cornu ammonis (CA)1, CA2, CA3, and dentate gyrus (DG). (B) shows an outlined CA1 neuron dendrite (in yellow).

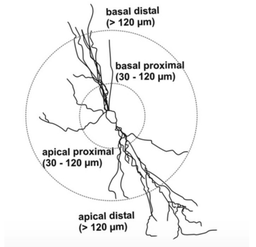
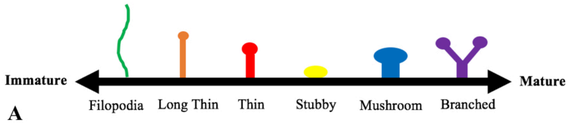


Fig. 3: Proximal and distal apical dendrites. The proximal apical dendrites are found 30µm-120µm from the soma, while the distal apical dendrites are found greater than 120µm from the soma.[2]



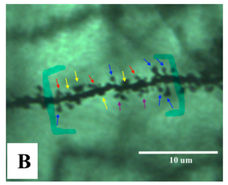


Fig. 4: Characteristics and morphology of dendritic spines. **(A)** shows a cartoon of the dendritic spine morphology and maturity of the spines. **(B)** shows the sham dendritic spines and the characterization of the spines (shown in colored arrows) using figure **(3A)**.

Results- Reconstructed Neurons

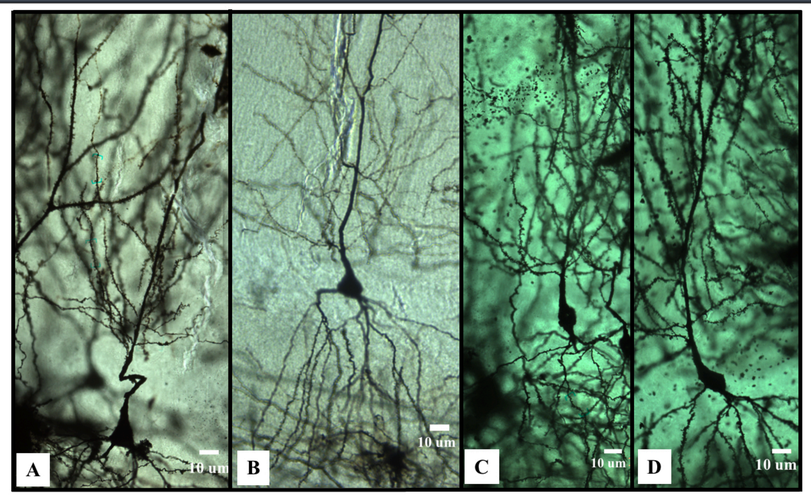


Fig. 5: Full neuron images. **(A)**, **(B)**, **(C)**, and **(D)** all show the full neurons from sham, 24 hours, 7 day, and 30 day groups, respectively.

Results- Spine Density and Morphology

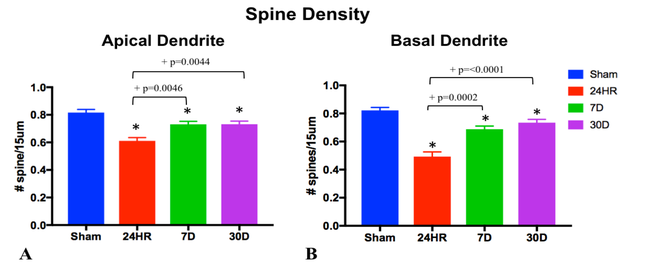


Fig. 6: Bar graph representation of spine density for apical and basal dendrites. **(A)** shows the spine density for the apical dendrites. The 24 hour (24HR), 7 day (7D), and 30 day (30D) spine densities showed statistically significant differences ($p<0.05$) when compared to the sham (shown with *). Further, there were differences between the experimental groups (shown with +). **(B)** shows the spine

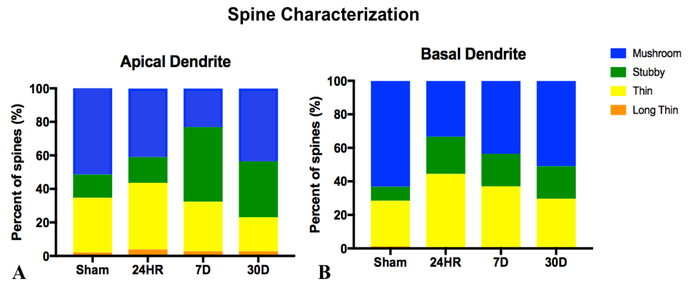


Fig. 7: A stacked bar graph showing the spine characterization and the percentage of spines found at each group for apical and basal dendrites. In **(A)**, the sham apical dendrites showed more mushroom spines. There was a shift towards more immature spines at 24HR. At 7D and 30D, there was an increased percentage of stubby spines. In **(B)**, the basal dendrites showed similar trends to the apical dendrites. Spine maturity was observed at 30D.

Conclusions

- The spine density at acute time points after CA/CPR were significantly lower compared to the sham. By 30 days, spine density and morphology mostly recovered to sham levels, though stubby spines were present in apical dendrites. These data indicate that the acute loss of spines seen at 24 hours do not fully account for LTP deficits seen at 30 days after CA/CPR.
- The spine density and morphology indicate the structural plasticity of spines, which show that neurons can strengthen and weaken synapses and potentially regain memory by increasing spine density and creating more spines that are mature in morphology.

References and Acknowledgements

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[2]Perez-Cruz C, Nolte MW, van Gaalen MM, Rustay NR, Termont A, Tanghe A, Kirchhoff F, Ebert U. 2011. Reduced spine density in specific regions of CA1 pyramidal neurons in two transgenic mouse models of Alzheimer's disease. J Neurosci 31:3926-3934.
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