Does the Cathelicidin/LL-37 Peptide Affect Amyloid-β Aggregation in a Mouse Model of Alzheimer's Disease?

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Alzheimer's, Amyloid β, and LL-37

- Amyloid β (Aβ) peptide deposits as clumps or plaques in the brain.
  - Causes inflammation, neuronal cell death, and eventually AD.
- In vitro evidence Human LL-37 can interact with and prevent clumping of Amyloid β (Aβ).
- Human Cathelicidin antimicrobial peptide LL-37 encoded by the *CAMP* gene is regulated by the vitamin D pathway

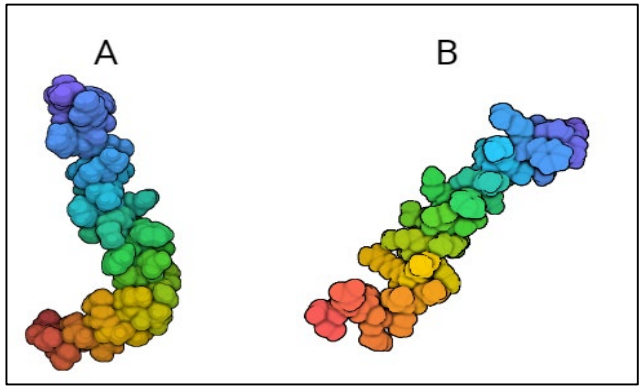


Figure 1. (A) Amyloid-β and (B) Cathelicidin LL-37 Peptide Structure derived from the Protein Data Bank and 3D NMR Structure Determinations (2,3)

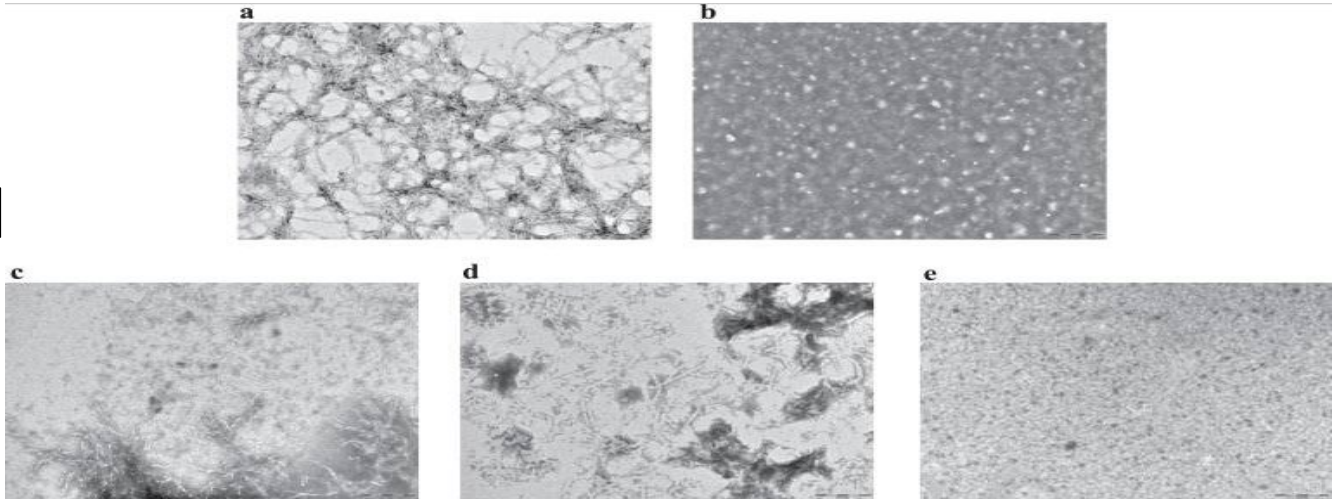


Figure 2. Transmission electron micrographs for (a) 50 µM Aβ42 at t = 0; (b) 100 µM LL-37 at t = 10 days; (c) equimolar mixtures of 50 µM Aβ42 and LL-37 at t = 0; (d) t = 3 days; (e) t = 9 days. Scale bar: 200 nm, magnification 60,000x. Ab42 alone forms fibrils and LL-37 alone forms globular, amorphous aggregates. In an equimolar mixture of both peptides, the formation of fibrils decreases over time (panels c-d). Adapted from De Lorenzi, et al., 2017 (1).

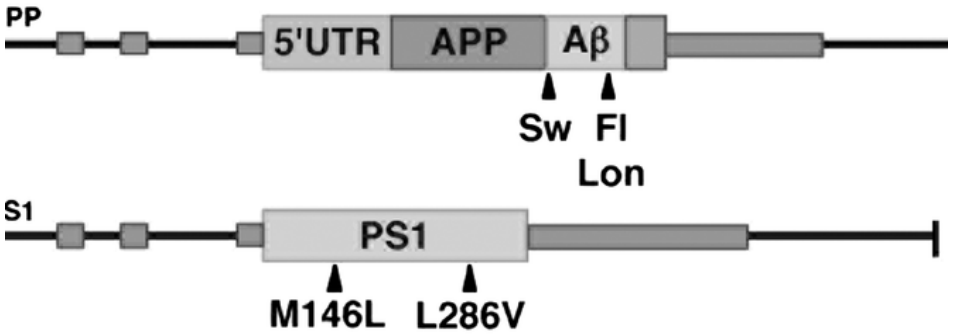
Hypothesis

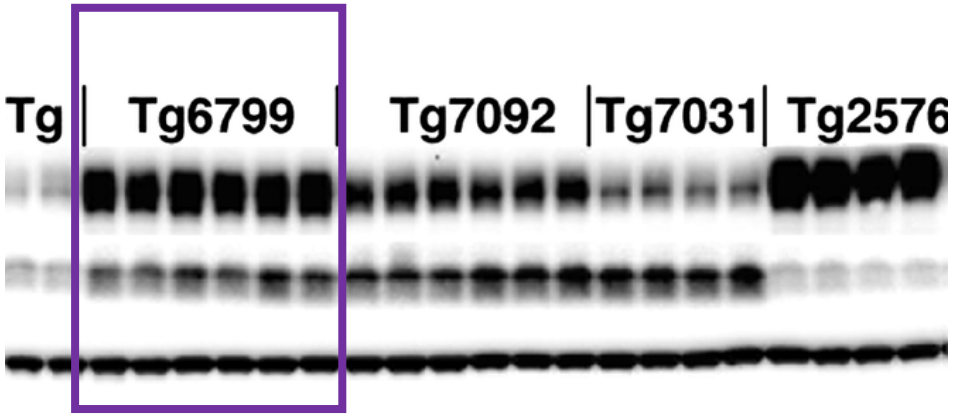
We hypothesize that *the absence of murine Camp will increase the aggregation of the Amyloid-β protein in vivo in a mouse model of AD.*

We expect the average plaque % area will be greater for the *Camp*-null:5xFAD-positive mice than the Camp-positive:5xFAD-positive mice.

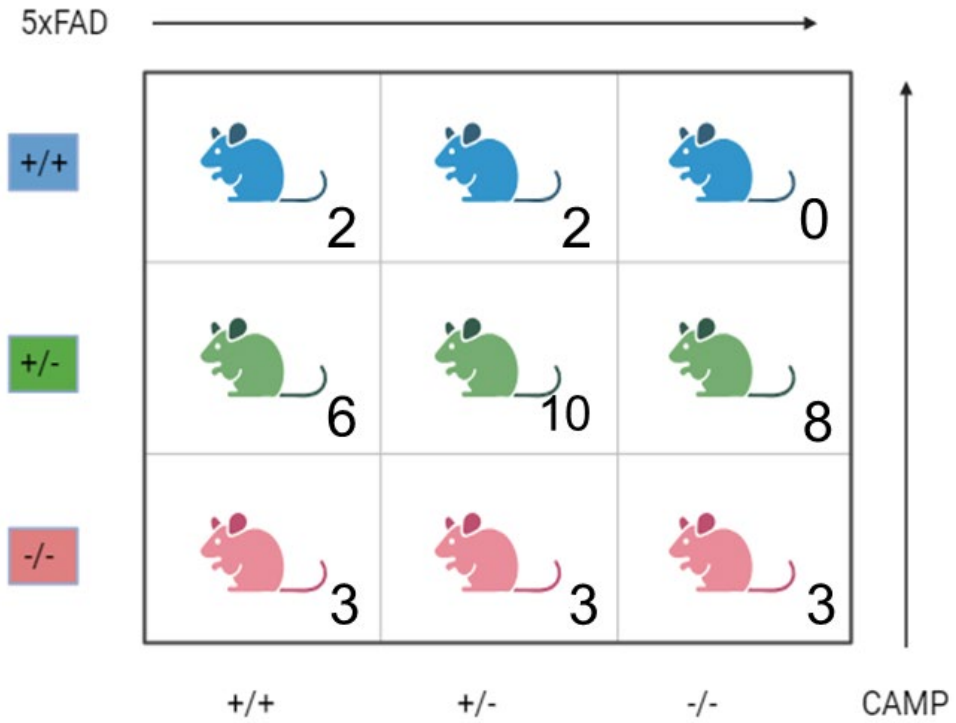
Mouse Model

5xFAD mouse





Camp$^{KO/KO}$ x 5xFAD$^{+/-}$:*Camp*+/+



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Plaque Quantification Methodology

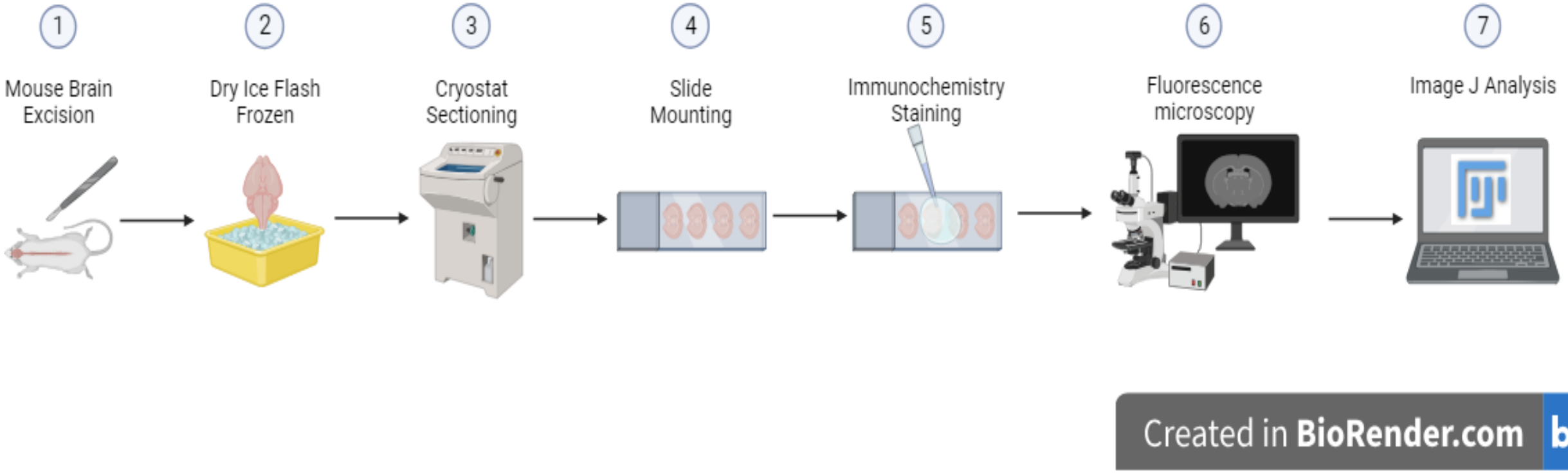
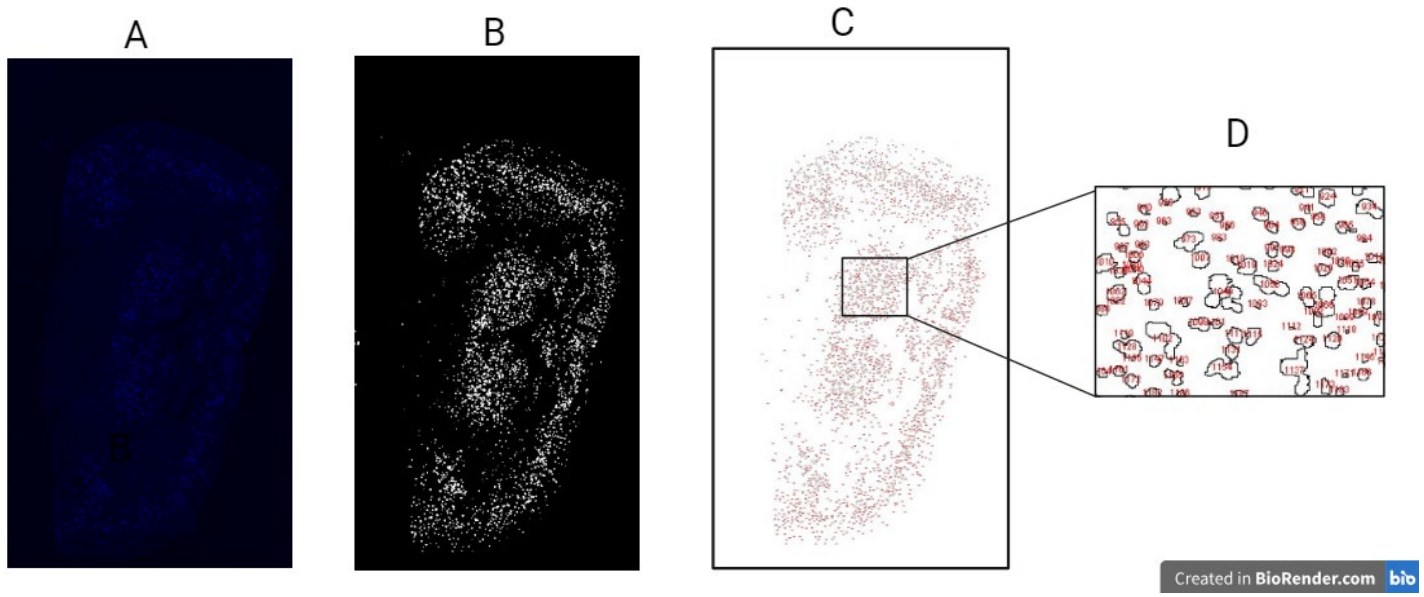


Image J Area Quantification



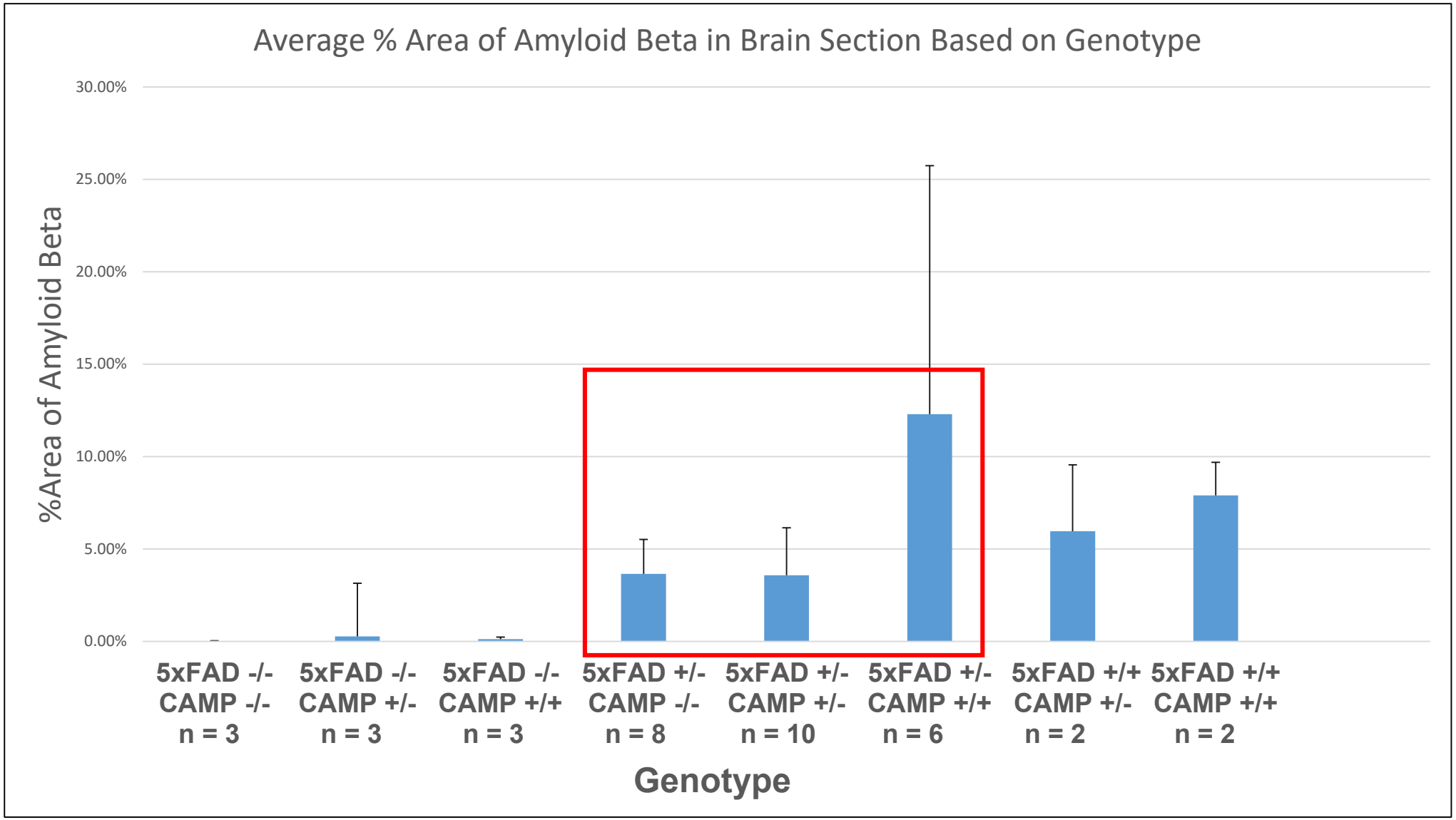
Images of mouse 402-3 **A**) DAPI Fluorescence Microscope Image **B**) Image J Thresholded DAPI Image **C**) Image J Analyze Particle Outline Map **D**) Close in on C looking at individual plaque numbers

Slice	Count	Total Area	Average Si	%Area	Mean
5xFAD_40	2626	23.453	0.009	4.922	255

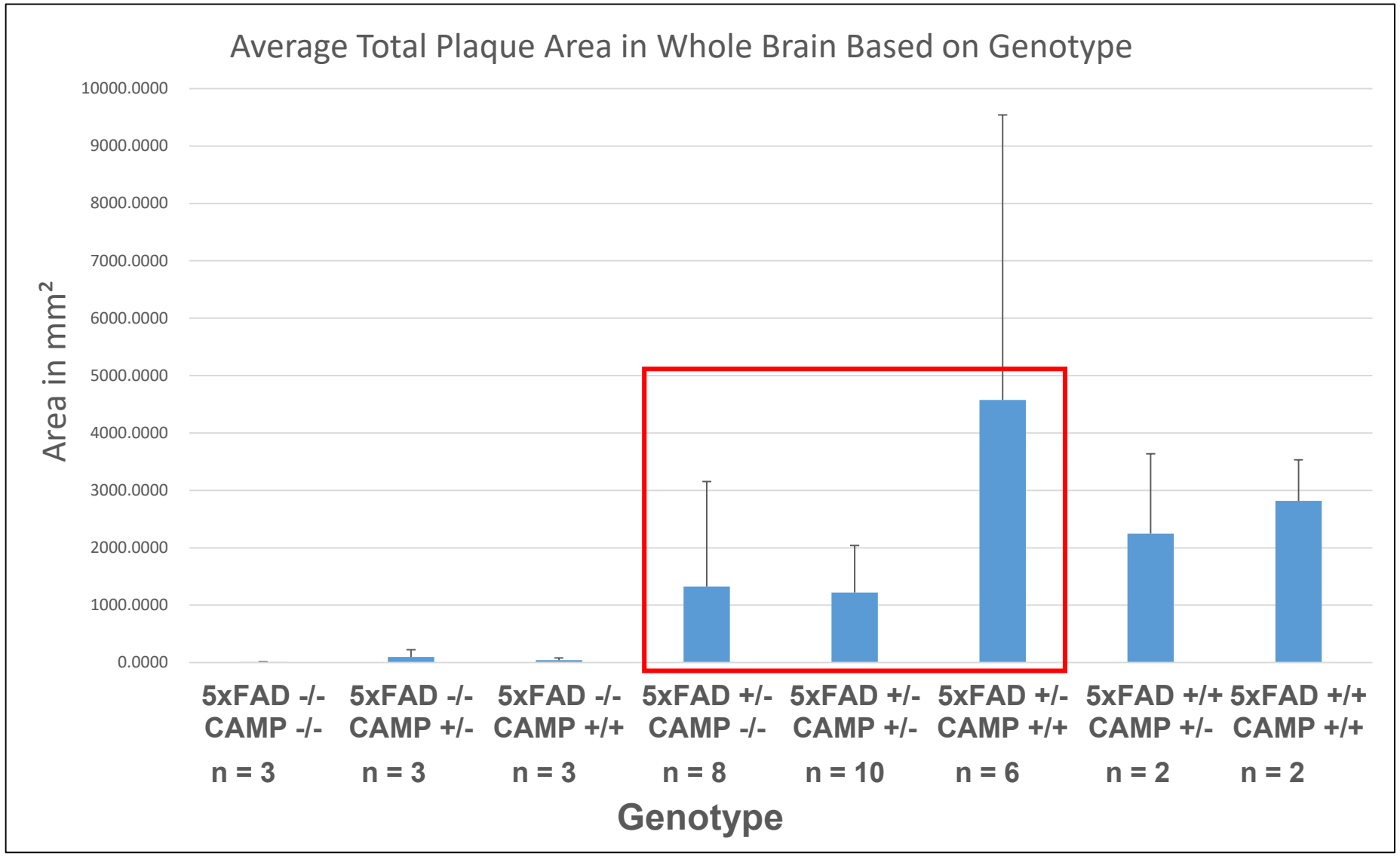
	Label	Area	Mean	%Area
1		0.002	255	100
2		0.008	255	100
3		0.009	255	100
4		0.001	255	100
5		0.004	255	100
6		1.09E-04	255	100
7		0.003	255	100
8		0.009	255	100
9		6.51E-04	255	100
10		0.005	255	100
11		0.003	255	100
12		0.01	255	100
13		0.005	255	100
14		4.34E-04	255	100
15		0.02	255	100
16		0.009	255	100
17		0.011	255	100
18		0.006	255	100
19		0.005	255	100
20		0.002	255	100
21		0.005	255	100
22		0.004	255	100

Image J Measurement Data

Average % Area in Whole Brain



Average Total Area in Whole Brain



Conclusions

1. Findings indicate the reliability and efficacy of our methodology
2. Did not detect a statistically significant difference in plaque burden in mice lacking the *Camp* gene.
3. Limitations include the number of mice for each genotype and the human/murine differences

Future Directions

1. Collect, process & analyze additional brains to increase the numbers of the different genotypes.

2. Determine if expression of human LL-37 in 5xFAD mice reduces plaque levels.

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