Studying Alzheimer's Disease Preclinically in More than Just "Bob": A Multi-Strain Study of 5xFAD Gene in Aged Mice

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Introduction/Background

Alzheimer's disease is a neurodegenerative disease that commonly affects older adults [1]. Advanced age is the greatest risk factor, but genetics plays a big role [1]. The 5xFAD mouse model expresses five genetic mutations commonly associated with familial Alzheimer's disease [2]. Mouse research is commonly done with one inbred mouse strain, the C57BL/6. Because of the inbred nature, it is akin to doing research with genetic variation of N=1, i.e. only in "Bob". The BXD family are a genetic reference population derived from crossing the C57BL/6 and DBA/2J strains. The resulting offspring have genetic variation comparable to that of the human population [3]. In this study, we investigate the brains of four strains in the BXD family crossed with the 5xFAD gene using high-resolution diffusion MRI. This framework allows us to study the impact of the underlying genetic background on the expression of Alzheimer's disease.

Methods

Four BXD strains (32, 65, 77, and 101) of age 438.4 +/- 31.6 days with two different genotypes (**Tg**: 5xFAD expressed, **nTg**: no-transgene) were perfusion fixed with a contrast agent to enhance the MR signal. The excised heads were scanned in a 9.4T MRI scanner using an imaging sequence to encode diffusion. The distribution of specimens is indicated in **Figure 1**. Labels were placed on 231 regions and parent structures using SAMBA [4]. Diffusion metrics of axial diffusivity (AD), radial diffusivity (RD), mean diffusivity (MD), and fractional anisotropy (FA) were extracted for each region. Statistical processing (N-Way ANOVA for each contrast type ~ strain + sex + scanner + genotype) was performed in MATLAB using custom pipelines. BH FDR correction was performed. Follow up ANOVA tests were performed with strain stratification (~ sex + scanner + genotype) for regions identified by the overall ANOVA.

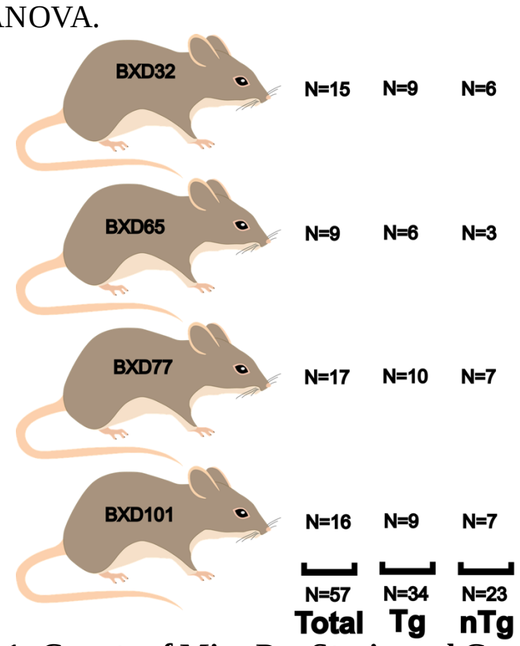


Figure 1: Counts of Mice Per Strain and Genotype.

Results

The overall ANOVA results, **Figure 2**, are illustrated in an ontology ordered from front to back of brain. The levels left to right indicate parentage of structures. Full name and abbreviations of all regions included here is in [5].

Most of the significant changes indicated in the overall ANOVA are in the FA contrast (**Figure 2 Right Most Column**). Filtering to the most highly significant regions (green) in the FA, we highlight the difference in significance by strain, **Figure 3**.

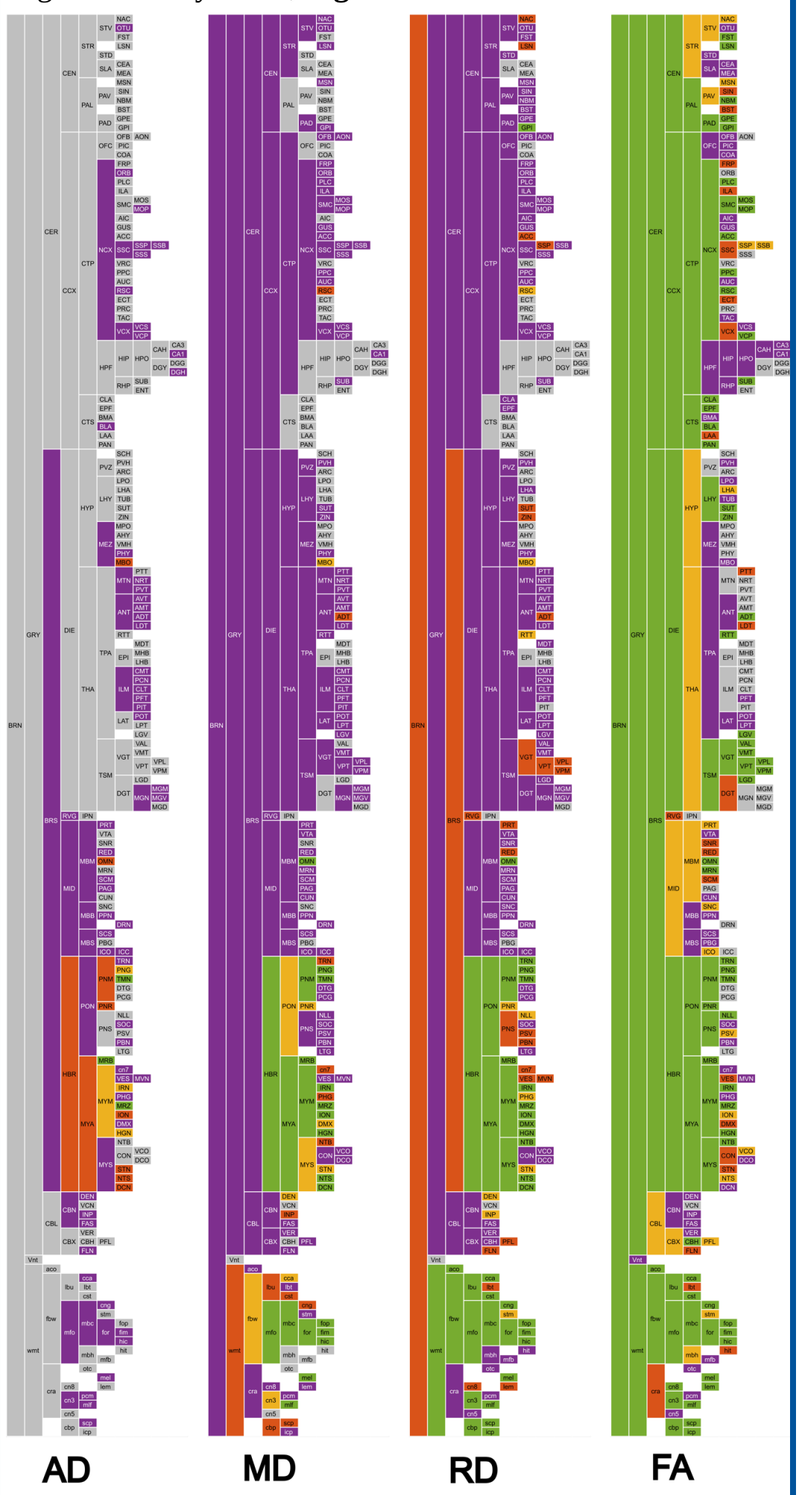


Figure 2: Significance of diffusivity metrics in all regions of the ontology for the overall ANOVA model.

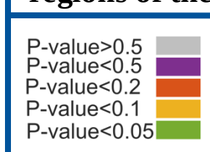
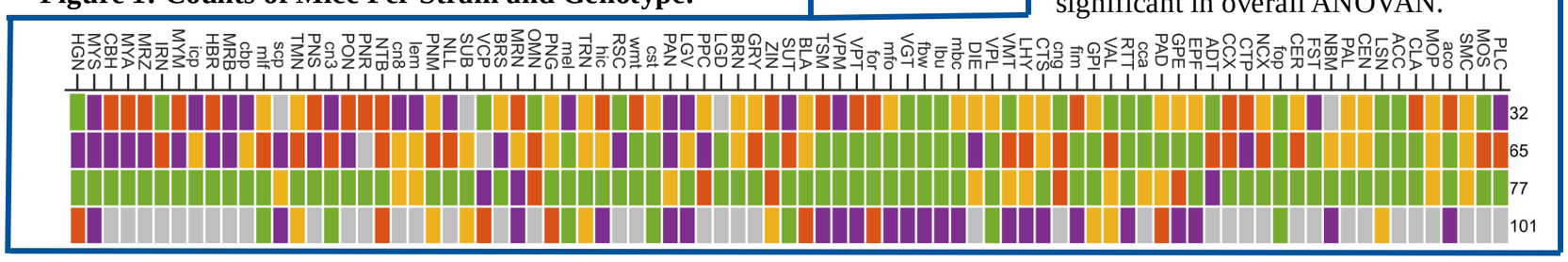


Figure 3: Significance of FA in separate strain stratified ANOVAs. Regions used here are most highly significant in overall ANOVAN.



Conclusion/Discussion

There are highly significant changes (corrected p-value <0.05) of diffusion metrics driven by the transgene across all strains. FA changes are seen in the largest number of structures (85 hits). MD (22 hits) and RD (39 hits) are primarily localized in forebrain white matter and hindbrain. There are limited changes in AD (3 hits). The changes in FA are variable in significance related to the strain. The strain with the most highly significant (p-value <0.05) regions is BXD77 (65 hits), followed by BXD65 (28 hits), BXD32 (18 hits), and BXD101 (7 hits). All strains are significantly changed in FA for the postcommissural fornix. This region is noted for links to memory and in human literature as potentially being disrupted with Alzheimer's [6].

Since the sampling of each group is not identical, the effect sizes needed to maintain sufficient statistical power varies. For future work, we are adding more BXD65. We plan to investigate younger mice with the same genotypes to consider age related differences.

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