Diffusion tensor imaging metrics may be less sensitive than volumetry/morphology in measuring differences in mouse models related to autism.

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Background - Current research has shown volumetric differences in 26 different mouse models related to autism (1). In that work, we found strong differences in the white matter. Both smaller and larger white matter was found in two subsets of those 26 models, leading us to hypothesize about differences in the structural connectivity in autism.

Objectives - The purpose of this work is to use diffusion tensor imaging (DTI) to further examine the structural connectivity differences in 13 different mouse models related to autism to further investigate the white matter differences.

Methods – The mouse models were acquired either from the Jackson Laboratory (jax.org) or through collaboration with other research labs. Approximately 10 mice were included for each individual genotype and each model had its own corresponding wild type control. All scans were performed ex-vivo with the brain left in the skull.

MRI Acquisition - For the 13 mouse models of autism, 212 individual scans were acquired using previously detailed high-throughput techniques (2), which allow 16 mouse brains to be scanned at a time. A 7.0 Tesla MRI (Varian Inc., Palo Alto, CA) was used to acquire ex-vivo anatomical images of brains within skulls. DTI scan - A 3D Diffusion-Weighted fast spin-echo sequence was used, with a TR of 325 ms, a first TE of 30 ms and a TE of 6 ms for the remaining 5 echoes. A single average was used in each of 30 directions following the Jones30 scheme (3) for the high-bvalue images (b=1917 s/mm^2), 5 b=0 s/mm^2 images were also acquired. The field-of-view was 14 × 14 × 25 mm^3 with a matrix size of 180 x 180 x 324 giving an image with 0.078 mm isotropic voxels. Total imaging time was ~12 h.

Data Analysis – To visualize and compared the differences found the b=0 s/mm^2 images were registered together for each of the 13 models. Mean Diffusivity (MD) and Fractional Anisotropy (FA) maps were created using the FSL software package, and then transformed using the b=0 s/mm^2 transformation to bring the MD and FA maps into alignment. Voxelwise and Regional differences for both MD and FA were then calculated for each of the 13 models. Regional differences were calculated in 62 different regions encompassing the whole brain (4).

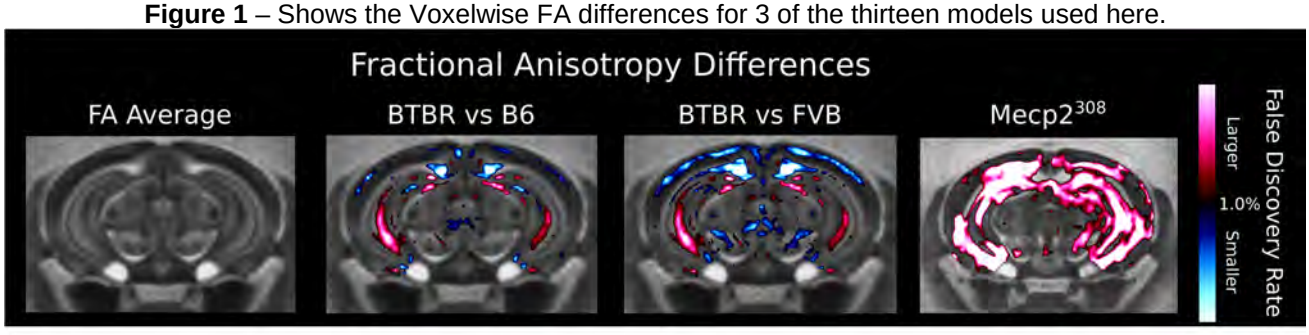
Table 1 – Lists the 13 models and the regional differences in FA

Model	# of Regions FA difference FDR of 5%
BTBR (B6)	15/62
BTBR (FVB)	23/62
FMR1 (-/y) (B6)	0/62
GTF2I (-/+)	0/62
GTF2I (dp/dp)	0/62
ITGβ3 (-/-)	0/62
MECP2^{308}	40/62
NLGN3 R451C KI	0/62
NRXN1α (+/-)	0/62
NRXN1α (-/-)	0/62
SLC6A4 KI (B6)	0/62
SLC6A4 KI (129)	0/62
SCL6A4 KO	0/62

Results – Table 1 highlights the regional differences in FA for each of the 13 models examined. Only 3 mouse models had FA differences: BTBR compared to both control strains, it should be noted that the BTBR does not have a crossing corpus callosum, and the Mecp2 (Rett Syndrome) model. The BTBR mouse did have several differences outside of the corpus callosum, however, including increases in FA in the hippocampus (Figure 1), and the Mecp2 mouse showed large scale FA increases throughout the brain. Outside of the differences shown in Fig. 1, no differences were found in FA or MD in the other 10 models, despite models like NL3 R451C KI (5) and ITGβ3 (6) which show large-scale white matter differences. In fact, the ITGβ3 mouse had a 7.1% and 12.3% smaller corpus callosum and fimbria, respectively.

Conclusions – While it was expected to not see FA or MD differences in some of the models listed here, for example the SLC6A4 models and the GTF2I models in which the white matter was not affected, we did expect to find FA differences in the NL3 R451C KI, ITGβ3 and NRXN1α models. A possible explanation for the relative lack of DTI findings may be that the volume differences are caused by fewer axons, but those remaining axons, however, remain as tightly packed with similar sizes as before (i.e. consistent density, organization, and size), thus not affecting the FA or MD. The fact that we are scanning ex-vivo tissue also might affect the DTI metrics. Overall, our results highlight that volumetry/morphology may be more sensitive than FA and MD when examining subtle difference brought on by genetic manipulations.

Figure 1 – Shows the Voxelwise FA differences for 3 of the thirteen models used here.



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