**Title**: Myelin Water Fraction and Quantitative Relaxation Rate Differences in Orienting Attention-Related Regions in Autism

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**Introduction**: While increasing evidence suggests widespread white matter microstructure alterations in autism, there is limited research on direct assessments of myelin in this population. Multicomponent relaxometry may provide a more specific assessment of myelin content through the myelin water fraction (*VFM*). To date, only one study (Deoni et al., 2015) has examined *VFM* in autism via voxel-wise comparisons, revealing lower *VFM* in autistic individuals in comparison to neurotypical individuals. The present study aims to investigate *VFM*, along with quantitative longitudinal and transverse relaxation rates (*R1*, *R2*), in white matter tracts involved in orienting attention (the splenium of corpus callosum [SCC], superior longitudinal fasciculus [SLF], and uncinate fasciculus [UF]; e.g., Dowe et al., 2020; Elison et al., 2013; Murray et al., 2016; Niogi et al., 2010), which are closely linked to social communication difficulties in autism. Specifically, this study aimed to examine: (Aim 1) the effects of age-related maturation on *VFM*, *R1*, and *R2* in the SCC, SLF, and UF across autism and neurotypical groups; (Aim 2) whether *VFM*, *R1*, and *R2* in the SCC, SLF, and UF differ between groups, including age-related group differences; and (Aim 3) whether these alterations are associated with parent-reported social behaviors (Social Responsiveness Scale [SRS]; Constantino & Gruber, 2012) in autistic individuals.

**Methods**: Imaging data were acquired from 48 autistic (92% male; *Mage* = 16.3; 5 to 42 years) and 71 neurotypical individuals (94% male; *Mage* = 15.8; 5 to 33 years) using mcDESPOT, a multicomponent relaxometry technique (Dean et al., 2014, 2016; Deoni et al., 2008). Before the main analyses, a paired t-test assessed the mean differences in the left and right SLF and UF to determine the inclusion of SLF and UF as single or separate left and right regional values; and an intraclass correlation evaluated the agreement between mother and father reports on SRS to inform the inclusion of mother and father reports as separate predictors in the model. Subsequently, linear and logarithmic models were compared using the Akaike/Bayesian Information Criterion to identify the model best representing age-related effects; and non-parametric ANCOVA and multiple regression with permutation test were conducted to examine group differences and associations with SRS, respectively. For all analyses, age was controlled (the best-fit age model identified from the first aim), and a false discovery rate (FDR) correction (Benjamini & Hochberg, 1995) was applied to adjust for multiple comparisons.

**Results**: The paired t-test indicated distinctions between the left and right UF for *VFM* (*p* < .001), the left and right SLF and UF for *R1* (*p* = .004, *p* < .001, respectively), and the left and right SLF for *R2* (*p* < .001), leading to different region of interest (ROI) analyses for each measure under investigation: SCC, SLF, lUF, and rUF for *VFM*; SCC, lSLF, rSLF, lUF, and rUF for *R1*; and SCC, lSLF, rSLF, and UF for *R2*. For Aim 1, age effects were observed for *VFM*, *R1*, and *R2* in the relevant ROIs across autism and neurotypical groups at the FDR-corrected level of *p* < .05. Specifically, *VFM* showed a positive nonlinear association in the SCC (*p* = .006) and SLF (*p* < .001) and a positive linear association in the rUF (*p* < .001); *R1* showed a positive nonlinear association in all ROIs (SCC: *p* = .041; lSLF: *p* = .005; rSLF: *p* = .010; lUF: *p* = .016; rUF: *p* = .016); and *R2* showed a positive nonlinear association in the SCC (*p* < .001) and SLF (left: *p* < .001; right: *p* < .001) and a positive linear association in the UF (*p* < .001). For Aim 2, no group differences were found for *VFM*. However, in the uncorrected results, the autism group showed higher *VFM* in the SLF (*p* = .030) compared to the neurotypical group. In addition, *R1* was higher in the autism group compared to the neurotypical group in the SCC (*p* = .005) and SLF (left, *p* = .005; right, *p* = .003) at the FDR-corrected level of *p* < .05. No group differences for *R2* or age-by-group interactions across *VFM*, *R1*, and *R2* were found at either the uncorrected or corrected levels. Lastly, for Aim 3, intraclass correlation revealed a low agreement between mother and father reports of SRS, leading to the examination of mother-reported and father-reported SRS scores as separate predictors. No associations were found between SRS scores and *VFM*, *R1*, or *R2* in the relevant ROIs within the autism group at either the uncorrected or corrected levels.

**Discussion:** Findings indicate that as people age, the structural integrity of white matter in brain regions linked to orienting attention evolves in a complex, non-uniform way. In addition, findings suggest that observable differences in orienting attention skills between autistic and neurotypical individuals may not stem from variations in myelination but instead may reflect other tissue properties or microstructural changes captured by longitudinal relaxation rates. Lastly, considering existing evidence of an association between similar measures investigated in this study and social behaviors indexed by less subjective behavioral measures, myelination perhaps may not be universally predictive across different types of behavioral measures used to assess social behaviors in autistic individuals. It suggests incorporating other types of behavioral measures, particularly those that are less subjective, may be needed to better understand how white matter integrity in orienting attention connects to observable social behaviors in autistic individuals.

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