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# Asymmetry Matters: Diffusion Tensor Tractography of the Uncinate Fasciculus in Children with Verbal Memory Deficits

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## ABSTRACT

**BACKGROUND AND PURPOSE:** Verbal declarative memory performance relies on frontotemporal connectivity. The uncinate fasciculus is a major association tract connecting the frontal and temporal lobes. Hemispheric asymmetries contribute to various cognitive and neurobehavioral abilities. Here we investigated microstructural alterations and hemispheric asymmetry of the uncinate fasciculus and their possible correlation to memory performance of children with learning disorders attributed to verbal memory deficits.

**MATERIALS AND METHODS:** Two groups of right-handed children with learning disorders attributed to verbal memory deficits and typically developing children ( $n = 20$  and  $22$ , respectively) underwent DTI on a 1.5T scanner. Tractography of the uncinate fasciculus in both hemispheres was performed, and fractional anisotropy and diffusivity indices (radial diffusivity, axial diffusivity, and trace) were obtained. The asymmetry index was calculated. Verbal memory was assessed using subsets of the Stanford Binet Intelligence Scale, 4th edition, a dyslexia assessment test, and the Illinois test of Psycholinguistic Abilities. Correlation between diffusion metrics and verbal memory performance was investigated in the learning disorders group. Also, hemispheric differences in each group were tested, and between-group comparisons were performed.

**RESULTS:** Children with learning disorders showed absence of the normal left-greater-than-right asymmetry of fractional anisotropy and the normal right-greater-than-left asymmetry of radial diffusivity seen in typically developing children. Correlation with verbal memory subsets revealed that the higher the fractional anisotropy and asymmetry index, the better the rapid naming performance ( $P < .05$ ) was.

**CONCLUSIONS:** These findings demonstrated microstructural aberrations with reduction of hemispheric asymmetry of the uncinate fasciculus, which could disrupt the normal frontotemporal connectivity in children with learning disorders attributed to verbal memory deficits. This outcome gives more understanding of pathologic mechanisms underlying this disorder.

**ABBREVIATIONS:** AD = axial diffusivity; AI = asymmetry index; FA = fractional anisotropy; LD = learning disorder; RD = radial diffusivity; UF = uncinate fasciculus; TDC = typically developing children; VMD = verbal memory deficits

Memory is considered a critical component of the learning process. Children rely on their memory for acquiring and processing new information, which is needed for proper school performance.<sup>1</sup> Verbal memory is concerned with the processing of language and verbally presented information. A higher proficiency at verbal memory-related tasks was found to be associated with


better educational outcomes. Verbal memory was reported to be defective in children with various types of learning disorders (LDs).<sup>2,3</sup>

Different brain systems have been found to control different types of memory. Connections between diverse brain regions are essential for the process of encoding memory, in addition to its consolidation and retrieval.<sup>1,4</sup> Verbal declarative memory is thought to rely primarily on medial temporal lobe structures including the hippocampus. Functional neuroimaging studies have found that the left medial temporal lobe supports the ability to encode verbal information with activation of the left hemispheric prefrontal regions during semantic retrieval.<sup>5,6</sup> Increased memory performance has been correlated with elevated functional connectivity between the temporal lobes and prefrontal cortex.<sup>7</sup> Moreover, the 2 cerebral hemispheres were found to have functional and anatomic asymmetries that contribute to cognitive and neurobehavioral abilities; this hemispheric lateralization of function was found to be associated with increased cognitive ability.

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Aberrations in hemispheric asymmetries have been reported in dyslexia, autism spectrum disorder, and schizophrenia.<sup>8-12</sup>

The uncinate fasciculus (UF) is a long association tract connecting the inferior frontal and mesial temporal lobes (regions that are implicated in encoding and retrieval of verbal memory). Left hemispheric dominance of the UF has been reported in healthy individuals.<sup>13-16</sup> In developing children and adolescents, the proficiency of verbal memory performance has been linked to white matter integrity of the left UF in DTI studies.<sup>17,18</sup>

Furthermore, DTI has been used to explore the correlations between white matter microstructure and different aptitudes reflecting cognitive performance, not only in typically developing children (TDC) but also in children with other neuropathologic conditions such as temporal lobe epilepsy and traumatic brain insult.<sup>12,19-22</sup> In addition, frontotemporal connectivity has been investigated using DTI in schizophrenia, dementia, and bipolar disorder, which are known to be associated with memory deficits.<sup>8,15,23</sup> However, to the best of our knowledge, there were no prior reports investigating frontotemporal connectivity and hemispheric asymmetry in children with LDs manifesting verbal memory deficits (VMD) without reading or writing disorders.

We hypothesized that frontotemporal connectivity expressed by UF integrity and hemispheric asymmetry might be altered in children with LDs manifesting VMD compared with TDC. To test this hypothesis, we investigated microstructural alterations of the UF in terms of fractional anisotropy (FA) and diffusivity indices using DTI. In addition, we tested the correlation between DTI markers and verbal memory performance in children with LDs.

## MATERIALS AND METHODS

### Participants

The study was conducted after approval of the ethics committee. A cross-sectional study included a convenient sample of children with poor scholastic achievement, which was related only to VMD. They were diagnosed as having an LD with executive functioning deficits, which included memory performance.<sup>24</sup> They were visiting the Learning Disability and Neurorehabilitation Research Clinic at the Medical Research Center of Excellence, National Research Center. In addition, control subjects were recruited from the patients' relatives. They included age- and sex-matched TDC, having the same social and ethnic origins. All participants were right-handed, native Arabic speakers, enrolled in the national educational system. Children with impairment in reading and writing and dyscalculia, sensory deficits, intellectual disability, associated neuropsychiatric disorders, or abnormalities on electroencephalography were excluded from the study group. Control subjects with a history of developmental language disorders, delayed developmental milestones, or having received language therapy sessions in early childhood were excluded as well. Also, any participant with contraindications to MR imaging was excluded.

A total of 45 subjects were initially included. Three subjects were excluded due to poor image quality. Finally, 20 children with LDs related to VMD and 22 TDC (matched in age, sex, and handedness) were enrolled in the study. The age of children in both groups ranged from 7 to 11 years; the mean ages were 9.3 (SD, 1.3) years and 9.1 (SD, 1.3) years for cases and control groups, respectively.

### Clinical Measures

The aptitudes of the children with LDs were evaluated by the Arabic version of the Stanford-Binet Intelligence Scale, 4th edition, for intelligence quotient assessment,<sup>25</sup> the Arabic Dyslexia Assessment Test,<sup>26</sup> and the Arabic version of the Illinois Test of Psycholinguistic Abilities.<sup>27</sup> The items that represented verbal memory in these tests were rapid naming, semantic fluency of the Arabic Dyslexia Assessment Test (representing long-term memory), the Digit Span Backward (representing working memory), and the Auditory Sequential Memory Test of the Illinois Test of Psycholinguistic Abilities (representing short-term memory). The higher the scores of rapid naming, the worse the performance of this subtest was. The opposite applied to the other subtests in which higher scores were associated with better performance.

### MR Imaging Protocol

All children were scanned with a 1.5T scanner (Achieva; Philips Healthcare) using an 8-channel sensitivity encoding head coil (sensitivity encoding acceleration factor of 8). The DTIs were acquired in 32 noncollinear directions along with baseline  $B_0$  images using a single-shot echo-planar sequence. Axial images were acquired parallel to the anterior/posterior commissure line with a 2-mm section thickness. The FOV was  $230 \times 230$  mm, and in-plane resolution was  $2.5 \times 2.5$  mm<sup>2</sup>. The head position was maintained using padding.

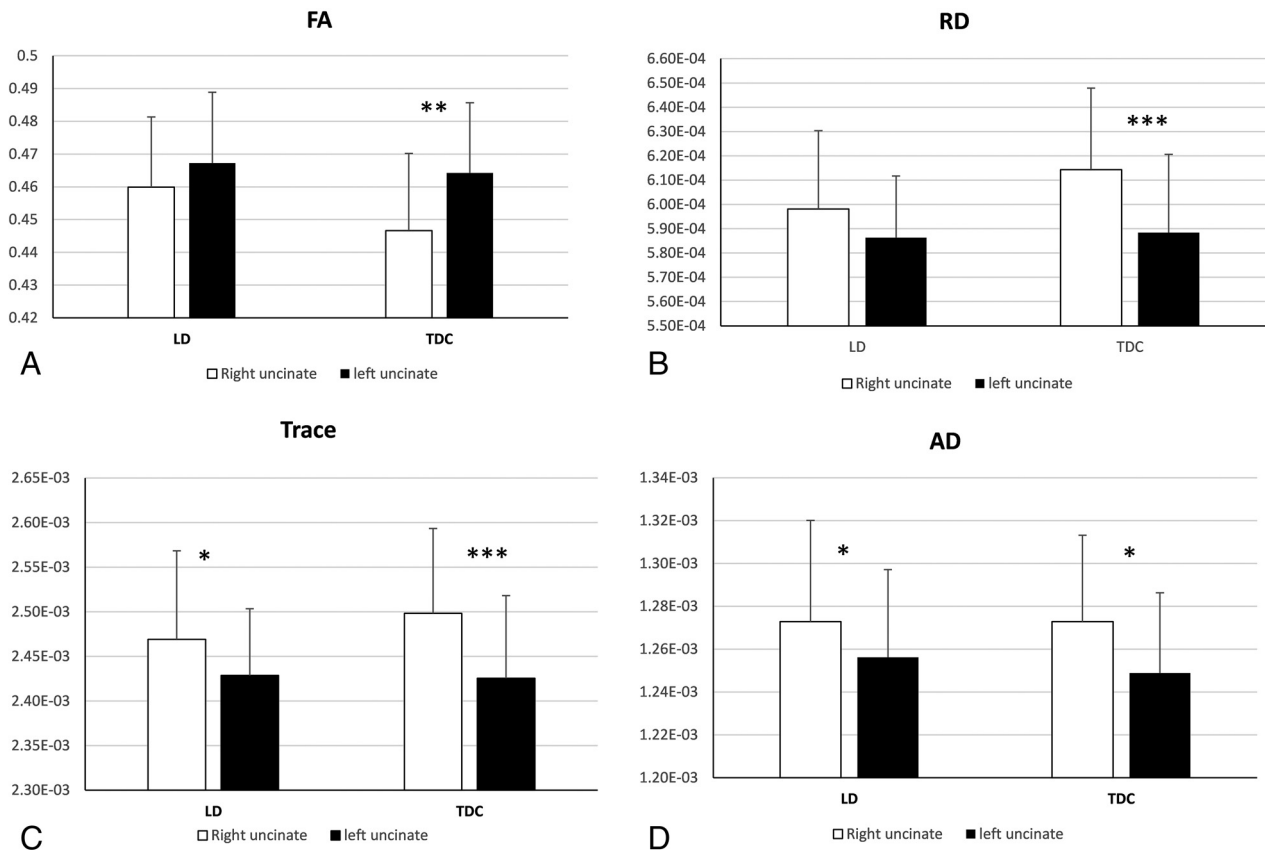
### Image Analysis

DTI images were analyzed in Windows on a PC using DTIStudio software (Version 3.0.3; Johns Hopkins University), produced by this laboratory (H. Jiang and S. Mori, the Johns Hopkins Medical Institute [<http://lbam.med.jhmi.edu>]). Participants with scans that showed obvious head-motion artifacts were excluded from the study. The raw DWIs were then coregistered to  $B_0$  images using the Automatic Image Registration tool (AIR; <https://www.nitrc.org/projects/air/>) with affine transformation and trilinear interpolation. FA and color FA and different diffusivity indices, radial diffusivity (RD), axial diffusivity (AD), and trace maps, were calculated in native space.<sup>28</sup>

### Tractography

Tractography was performed using the fiber assignment by continuous tracking method following the previously described high-reproducibility protocol described by Wakana et al.<sup>29</sup> The UF was extracted from both hemispheres using 2 manually drawn ROIs (Online Supplemental Data).<sup>29</sup> Tractography started at FA = 0.25. It ended at FA = 0.25 and a turning angle = 70°. The average FA, RD, AD, and trace were then calculated.

Moreover, the asymmetry index (AI) was calculated using the formula  $(2 \times [\text{Right} - \text{Left}] / [\text{Right} + \text{Left}]) \times 100$  to further quantify the differences between the measured variables of both hemispheres. A positive AI value indicated that the measured variable of the right hemisphere was greater than the corresponding left variable (rightward asymmetry), while a negative value corresponded to the opposite (leftward asymmetry).<sup>11,12</sup>



**FIG 1.** The UF asymmetries were calculated by comparing FA, RD ( $\text{mm}^2\text{s}^{-1}$ ), AD ( $\text{mm}^2\text{s}^{-1}$ ), and trace ( $\text{mm}^2\text{s}^{-1}$ ) in both hemispheres. Rightward asymmetry was defined as having a higher value in the right brain than in the left, while leftward asymmetry was defined as left-greater-than-right values. The asterisk indicates  $P < .05$ ; double asterisks,  $P < .01$ ; triple asterisks,  $P < .001$ .

### Statistical Analysis

FA, diffusivity indices (RD, AD, trace), and AI were correlated to the raw scores of rapid naming, semantic fluency, Digit Span Backward, and the Auditory Sequential Memory Test using the Spearman rank correlation test. To test hemispheric asymmetry in each group, we compared each tract value between both hemispheres using a paired Student  $t$  test. All the measured FA, diffusivity indices, and AI for each tract were compared between both groups using the Student  $t$  test.

## RESULTS

All participants with LDs manifested rapid naming and semantic fluency deficits (mean raw scores: 66.2 [SD, 24.3] and 9.3 [SD, 0.9], respectively) with intelligent quotient ranges of 89–107 (mean, 96.7 [SD, 51]). The percentage of children with deficits in the Digit Span Backward was 65%, and in the Auditory Sequential Memory Test, it was 35%. Correlation with clinical tests revealed a significant negative correlation between rapid naming and both FA of the right uncinate fasciculus and its AI ( $r = -0.5$ ,  $P = .02$  and  $r = -0.52$ ,  $P = .01$ , respectively). In other words, the higher the FA and AI, the lower the rapid naming score was with better performance. Otherwise, no significant statistical correlations were detected.

In TDC, the mean FA of the left UF was significantly higher than that of the right UF ( $P = .004$ ), while RD, trace, and AD were found to be significantly lower ( $P < .001$ ,  $P < .001$ , and  $P =$

.016, respectively). However, in children with LDs, there was no significant statistical difference between the right and left hemispheres regarding FA or RD. On the other hand, hemispheric differences regarding the AD and trace were still preserved ( $P = .04$  and .03, respectively) (Fig 1 and the Table).

Compared with TDC, the AI was found to be lower in children with LDs (Fig 2). Also, the UF of both cerebral hemispheres of the LD group showed higher FA, higher AD, but lower trace and RD. However, these differences failed to reach statistical significance (Table).

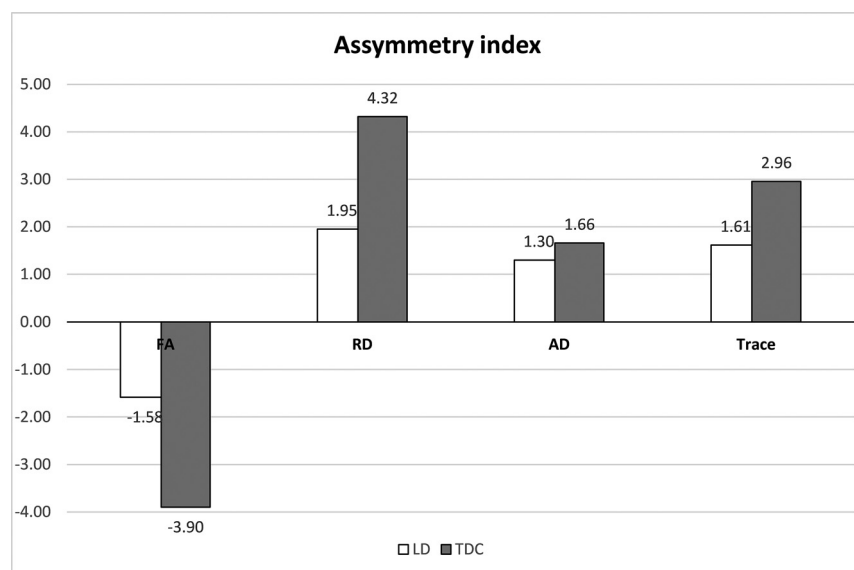
## DISCUSSION

Despite the existence of growing literature exploring the relation between frontotemporal connectivity and cognitive aptitudes in healthy subjects and individuals with neuropsychiatric abnormalities,<sup>8–12</sup> microstructural aberrations of the UF and hemispheric asymmetry in children with LDs related to VMD have not yet been comprehensively investigated. Our findings support the hypothesis of the existence of white matter microstructural alterations in terms of reduced hemispheric asymmetry in right-handed children with LDs related to VMD, in addition to significant statistical correlations with certain verbal memory subsets. These alterations in hemispheric asymmetry together with the correlation results might be useful in understanding the pathogenesis of VMD in children with LDs.

# Diffusion metrics of the uncinate fasciculus<sup>a</sup>

	LD				TDC			
	Right (mean) (SD)	Left (mean) (SD)	P Value	AI (mean) (SD)	Right (mean) (SD)	Left (mean) (SD)	P Value	AI (mean) (SD)
FA	0.46 (0.02)	0.47 (0.02)	.09	-1.58 (4.02)	0.45 (0.02)	0.46 (0.02)	.004	-3.9 (5.69)
RD	5.98E-04 (3.2E-05)	5.86E-04 (2.5E-05)	.06	1.95 (4.54)	6.14E-04 (3.35E-05)	5.88E-04 (3.22E-05)	<.001	4.32 (4.85)
AD	1.27E-03 (4.73 E-05)	1.26E-03 (4.09-05)	.04	1.3 (2.64)	1.27E-03 (4.03E-05)	1.25E-03 (3.74E-05)	.016	1.66 (3.00)
Trace	2.47E-03 (9.9E-05)	2.43E-03 (7.5E-05)	.03	1.61 (3.19)	2.50E-03 (9.50E-05)	2.43E-03 (9.25E-05)	<.001	2.96 (3.24)

<sup>a</sup> AD (mm<sup>2</sup>s<sup>-1</sup>), RD (mm<sup>2</sup>s<sup>-1</sup>), trace (mm<sup>2</sup>s<sup>-1</sup>).



**FIG 2.** AIs of the UF in both groups (FA, AD, RD, and trace).

All participants of the LD group manifested deficits in subtests related to long-term memory, highlighting its role in the performance of these children and necessitating its targeting in the rehabilitation programs designed to increase performance in executive functions. Furthermore, the percentage of participants showing deficits in verbal working memory was more than in those having deficits in short-term memory. Nonetheless, similar memory deficits were previously reported in children with dyslexia,<sup>3</sup> underscoring the contribution of memory performance in all kinds of LDs.

The rapid naming performance showed a significant correlation with the FA of the right UF and its AI, indicating the reliance of participants with LDs in this study on the right-sided connections. The more the FA and AI, the better was the performance of rapid naming. Learning and memory are complex interacting cognitive functions. Declarative memory, which is a form of long-term memory, incorporates semantic and episodic memory. Semantic memory is considered a child's database of knowledge about the world.<sup>30</sup> Data in formal educational situations in school-aged children are acquired through direct experience and are further extended through several processes. The latter include self-generation of new factual knowledge via integration of information acquired in separate-yet-related episodes of new learning. These operations are related to working memory integrity. Therefore, brain structures sharing in verbal memory processing are essential for the proficiency of learning.<sup>2</sup>

The uncinate fasciculus can be divided into 3 segments (temporal, insular segment, and frontal) (Online Supplemental Data). The temporal segment originates partly from the entorhinal cortex, perirhinal cortex, and anterior temporal lobe. The entorhinal and perirhinal cortices are believed to be related to episodic memory function, object memory, and perception, respectively, while the anterior temporal lobe has been linked to semantic memory.<sup>15</sup> While the role of the UF in object naming and semantic processing has been proposed; more recently, its role in object naming has been suggested to be the most relevant function.<sup>31</sup> Furthermore, verbal memory (as evaluated by list learning) was found to be instantly impaired following left UF resection in 18 right-handed individuals.<sup>32</sup> Similarly, a decrease in UF integrity has been associated with

semantic dementia and memory impairment in temporal lobe epilepsy.<sup>12,23</sup> Our finding of the significant correlation between the UF and rapid naming scores enforces the previously suggested link between them.

By means of DTI voxel-based morphometry, the UF was dissected into a longer superior segment and a shorter inferior segment. The former showed a higher FA in the left hemisphere, while the latter showed a greater FA in the right.<sup>20,33</sup> Using diffusion tensor tractography in our study, we averaged FA and diffusivity indices from all voxels occupied by this tract. Consequently, the mean values were influenced by the longer superior portion, which was found to be left-lateralized. Leftward asymmetry of the UF was also documented by other research groups.<sup>8,16,20</sup> Higher FA could reflect higher myelination and higher fiber numbers and/or density in the left hemisphere in TDC. Moreover, we have found that the left hemisphere had lower diffusivity values in terms of RD, AD, and trace. RD was found to reflect myelination, while AD reflects axonal integrity.<sup>34</sup>

In the control group (TDC), the hemispheric asymmetries of FA and the diffusivity indices of the UF may reflect structural differences between cerebral hemispheres. In the second and third trimesters of gestation, structural asymmetries have been found in the Sylvian fissure, the surrounding frontal operculum, and the planum temporal.<sup>33,35,36</sup> These structural asymmetries were found to be associated



with functional asymmetries as well. Production and processing of language are predominantly controlled by the left hemisphere, while visuospatial processing has right-hemispheric dominance in most individuals.<sup>37</sup> Decreased hemispheric asymmetry has been linked to autism spectrum disorder and developmental dyslexia.<sup>38</sup> Moreover, the lateralization hypothesis of schizophrenia signifying developmental aberrations in brain lateralization has been developed.<sup>36</sup> Similarly, the reduced asymmetry in the UF in children with LDs related to VMD can suggest an analogous mechanism with significant structural and functional aberrations that could be developmental in origin. Children with LDs in our study were found to have lost hemispheric asymmetry of the UF in terms of FA and RD, while the differences between UFs of both hemispheres in terms of AD and trace were preserved. To the best of our knowledge, this pattern of alterations of diffusion metrics has not been previously published.

Compared with the control group (TDC), UFs of both cerebral hemispheres in children with LDs showed higher FA, higher AD, but lower RD and trace. However, the difference was statistically insignificant. These possible aberrations in anisotropy and diffusivity could be attributed to an increase in myelination, axonal diameter, packing density, or branching. More FA and less diffusivity are not always indications of better function. These changes could represent compensatory mechanisms secondary to the dysfunction of memory-related cortical areas. Similarly, FA was found to be abnormally higher in the right superior longitudinal fasciculus of patients with Williams syndrome compared with control subjects.<sup>39</sup>

The study is possibly limited by the relatively small number of participants; however, to the best of our knowledge, this is the first study to investigate UF hemispheric asymmetry in children with LDs related to VMD. We recruited children who were perfectly matched in age, sex, and handedness to ensure that the DTI metrics were truly reflecting alterations in brain structure related to the disease rather than just differences in brain structure due to language hemispheric dominance. Also, a considerable percentage of included children with LDs had long-term memory impairment in addition to other types of verbal memory deficits. Nevertheless, memory performance and tasks interlace even in the models describing the memory.<sup>1</sup> Thus, it would be difficult to recruit children with only 1 type of memory deficit.

## CONCLUSIONS

The present findings indicated microstructural aberrations of the UF with reduction of hemispheric asymmetry in right-handed children with LDs attributed to memory deficits. These changes could influence the normal frontotemporal connectivity and, consequently, disrupt the proper functioning of memory performance of such children. The results of this study give more understanding of neuropathologic mechanisms underlying this disorder.

Disclosure forms provided by the authors are available with the full text and PDF of this article at [www.ajnr.org](http://www.ajnr.org).

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