

Sequential language learning versus language immersion in bilingualism: Diffusion MRI connectometry reveals microstructural evidence

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Background: Bilingualism is a universal phenomenon. Study of bilingual brain has provided evidence to probable advantageous outcomes of early second language learning and brain structural correlates to these outcomes. Preservation of cognitive function with aging and executive dexterity are amongst proposed benefits. **Method:** Using the data deposited by Pliatsikas we analyzed through diffusion MRI connectometry structural difference in white matter tracts in 20 healthy sequential bilingual adults, who used English as a second language on a daily basis, and 25 controls in fiber differentiation analyses. Significant tracts were extracted with and without regression against language immersion period. **Results:** Connectometry results revealed increased connectivity in corpus callosum (CC), bilateral cingulum, arcuate fasciculus (AF), and left Inferior fronto occipital fasciculus (IFOF), of sequential bilingual adults. Also bilateral IFOF, AF as well as body and genu of corpus callosum were positively correlated with language immersion time. **Conclusion:** Mentioned white matter tracts with diffusion structural significance in young adults with long immersion into the second language, confirm results of previous results. These are also in consort with known pathways involved in neurophysiological processes in speech stream and in cognitive performance. Insertion of immersion time as a variable in the model, yielded the same results with higher FDR values, except for arcuate fasciculus. Future studies are warranted to address structural differences with larger samples, investigating the effects of early language immersion on white matter tracts versus individual and social variables that interfere with white matter maturation and integrity.

Sequential Language Learning versus. Language Immersion in Bilingualism: 1

Diffusion MRI Connectometry Reveals Microstructural Evidence 2

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Running Title: Diffusion MRI Connectometry in Bilingualism 15

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Abstract 22

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fasciculus. Future studies are warranted to address structural differences with larger 41
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versus individual and social variables that interfere with white matter maturation and integrity. 43
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Keywords: *Bilingualism; Immersion; Connectometry; Diffusion MRI* 45

Introduction 46

Active use of two or more languages in daily life, or bilingualism is a global phenomenon, which 47
has a known role in "cognitive reserve" or preservation of cognitive function. Bilinguals are more 48
successful in resolving nonlinguistic cognitive conflicts (1), are less prone to degenerative brain 49
processes with aging (2), and have better executive control (3). Neuroimaging studies have opened 50
a window into the underlying structural changes in "bilingual brain", especially structural changes 51
affecting integrity of the white matter. 52

One unresolved question is whether critical periods exist for the developing brain to make 53
microstructural adaptations while learning a new language? It appears that white matter 54
microstructural changes associated with beneficial cognitive characteristics and increased white 55
matter integrity in terms of significantly increased fractional anisotropy (FA), occur in both early 56
and simultaneous learning of a second language (L2) (4). Increased FA in the inferior fronto 57
occipital fasciculus (IFOF), and decreased FA in genu and anterior corpus callosum (CC), are 58
amongst reported changes in early L2 learners among school aged children (5). Pliatsikasa and his 59
colleagues provided insights into white matter structural differences in young bilingual adults, who 60
were are sequential learners of English as second language and were highly immersed, meaning 61
that they had been using L2 on a daily basis for a long period (6). Through tract based spatial 62
statistics, they reported increased white matter integrity, in bilateral IFOF, uncinated fasciculi 63

(UF), and superior longitudinal fasciculus. They failed however to demonstrate a correlation 64
between the degree of immersion in months and white matter changes. 65

Grey matter change in volume has been also proposed in bilinguals. While no statistically 66
meaningful difference was apparent in point analyses by voxel based morphometry, longitudinal 67
data supported grey matter increase in size and volume in bilateral hippocampal areas (7), left 68
inferior parietal lobule (8), left anterior temporal gyrus (8), and bilateral anterior cingulate cortices 69
(1). Grey matter volume in these areas positively correlated with lexical efficiency and negatively 70
correlated with the age at which bilinguals had started learning a new language (7). 71

Using the data deposited at <https://central.xnat.org/> the by Pliatsikas (9), we analyzed through 72
diffusion MRI connectometry structural difference in white matter tracts in 20 healthy sequential 73
bilingual adults, and further investigated the probable correlation of diffusion metrics against 74
immersion period of the second language. 75

2. Materials and methods 76

2.1 Participants 77

Twenty healthy young adults who all used English as second proficient language and had lived in 78
the United Kingdom for at least 13 months, with various ethnic and mother languages backgrounds 79
formed the study group. Control group consisted of 25 age, sex and education matched native 80
speakers of English. All the subjects were taken the Quick Placement Test (QPT) for assessment 81
of their proficiency in English. Both groups scored at least 80 %. Detailed demographics of both 82
groups can be reached at the <http://xnat.org> depository (9). 83

2.2 Data Acquisition 84

A 3.0-Tesla Siemens MAGNETOM Trio MRI scanner was used with Syngo software and 36- 85
channel Head Matrix coil to acquire a whole-brain diffusion-weighted Echo-Planar Imaging 86
image (Two averages, 30 directions, 60 axial slices; slice thickness, 2 mm, no interslice gap; field 87
of view, 256×256 mm; acquisition matrix, 128×128; voxel size, 2 mm isotropic; echo time, 93 ms; 88
repetition time, 8,200 ms; b-value, 1,000 s/mm²). 89

2.3 Diffusion MRI Data Processing 90

Diffusion MRI data were corrected for subject motion, eddy current distortions, and susceptibility 91
artefacts due to the magnetic field inhomogeneity using ExploreDTI toolbox. 92

2.4 Comparison Analysis 93

The diffusion data were reconstructed in the MNI space using q-space diffeomorphic 94
reconstruction to obtain the spin distribution function. A diffusion sampling length ratio of 1.25 95
was used, and the output resolution was 1 mm. 96

Diffusion MRI connectometry was conducted to compare group differences in a total of 44 97
subjects. The group difference was quantified using percentage measurement. A threshold of 25% 98
difference was used to select local connectomes that had substantial difference. A deterministic 99
fiber tracking algorithm was conducted to connect the selected local connectomes. A length 100
threshold of 50 mm was used to select tracks. The seeding density was 20 seeds per mm³. The 101
analysis was conducted using DSI Studio (<http://dsi-studio.labsolver.org>). 102

2.5 Correlation Analysis with Immersion 103

In this part, Diffusion MRI connectometry was conducted in a total of 17 subjects using a multiple 104
regression model considering immersion. A percentage threshold of 50% was used to select local 105

connectomes correlated with immersion. A deterministic fiber tracking algorithm was conducted 106
to connect the selected local connectomes. A length threshold of 65 mm were used to select tracks. 107
The seeding density was 20 seeds per mm³. To estimate the false discovery rate, a total of 500 108
randomized permutations were applied to the group label to obtain the null distribution of the track 109
length. 110

3. Results 111

Our case-control study was carried out on 20 healthy "sequential" bilinguals, who spoke English 112
as their second language (original language varied across the participants). The case group 113
consisted of 7 (55%) male and 13 (65%) female bilinguals living part of their lifetime in United 114
Kingdom (with a mean of 91 months of UK residency). The mean age of bilinguals was $31.85 \pm$ 115
 8.07 (ranging from 20 to 47), and the age of L2 learning differed from 4 to 18 years among 116
bilingual adults. 117

Imaging data was analysed for 11 (44%) males and 14 (56%) females, who were matched with 118
respect to age, sex and English proficiency (measured through QTP criteria, scoring a minimum 119
of 80%), as control group. 120

3.1 Comparison Analysis 121

The connectometry analysis results showed tracks with increased quantitative anisotropy in 122
bilingual group with an FDR of 0.043. These tracks include corpus callosum (CC), bilateral 123
cingulum, arcuate fasciculus (AF) and left IFOF. Significant tracks are showed in Figure 1. 124

3.2 Correlation Analysis 125

Analysis of diffusion parameters against immersion period revealed that anisotropy values of IFOF bilaterally, AF bilaterally, genu and body of CC, are positively associated with immersion with FDR of 0.075, showing a trend toward significance (Figure 2). Higher FDR values mean higher probability of variation in brain connection patterns, and the observed FDR value is predictable due to low case numbers of this study.

4. Discussion

Bilingualism has been linked to a variety of cognitive benefits, including delayed brain aging, in terms of better executive performance in childhood, higher ability to resolve verbal and non-verbal conflicts and cognitive preservation of cognitive function with aging (10). The beneficial effects of bilingualism in cognition is still under debate, while measurements of executive function can give inconsistent reports even in a single population based on their current social, and educational status. The wide range of executive functions and the wide range of the tasks measuring one or two of the executive functions, along with different mechanisms by which an individual might have learned second language, further complicates an answer to this question (11).

There are different conditions through which someone might be exposed to a new language. This ranges from individuals being exposed to both languages since or early after birth, and thus are naturally immersed in the context of both languages, to those learning a second language in a sequential, not parallel, manner e.g. through classroom instruction etc. These cognitive benefits and aging preservation of bilinguals appear to be more prominent in the former, long immersed, natural/simultaneous learners (3). Finally, while executive and cognitive measurements are inaccurate, brain structural changes certify significant developmental or structural differences in bilingual versus monolingual individuals at least.

Also, apart from many factors that have been noticed apart from age, brain plasticity and perhaps 148
social and ethnic background that might affect white matter integrity in bilinguals, is the 149
“immersion”, or the time interval individual has been exposed to both languages 150

Luk and colleagues (12) performed one of the first DTI studies on bilingual subjects to extract 151
WM microstructural features and compare brain architecture in bilingual individuals. Their sample 152
consisted of lifelong bilingual individuals with English as first language and age-matched English 153
monolinguals. TBSS analysis revealed higher values of FA in bilingual group particularly in CC, 154
bilateral superior longitudinal fasciculi, right ILOF and uncinate fasciculus (UF). Pliasticas and 155
his colleagues (6) reported almost the same correlations on DTI scan in 20 young “sequential” 156
proficient English learners. 157

These fibers are parts of brain semantic processing as ventral and dorsal speech streams (13). Gold 158
(2), Cummine (5) and their colleagues, reported FA decrease in CC, Inferior longitudinal fasciculi 159
(ILF), IFOF and fornix in English sequential bilinguals, and in the right ILF and anterior thalamic 160
radiation in young Chinese–English late bilinguals respectively. IFOF and ILF share mutual 161
projections to and from the occipital and temporal lobes. Integrity of the ILF correlates well with 162
object recognition with lexical labels in children and increases sequentially with elaboration of the 163
skill. 164

These results were enough to propose the hypothesis that not necessarily bilingualism relies on 165
critical periods in life, but particular conditions are warranted to strongly induce it and that weather 166
by increase or decrease of anisotropy, white matter major associational fibers undergo structural 167
adaptations in response to new language learning. 168

Mohades et al. used ROI FA measurements assessed fractional anisotropic values in a sample 169 including 3 subgroups: 15 simultaneous bilingual children, 15 age-matched sequential bilingual 170 participants and 10 monolinguals. They obtained FA values for the left IFOF and four other 171 language related projections, and the right IFOF as control. In a longitudinal assessment, sequential 172 learners' depicted highest degree of change in FA values in the IFOF, and the mean FA value in 173 this region was able to distinguish simultaneous bilinguals from monolinguals (14). The left 174 inferior fronto-occipital fasciculus is primarily involved in semantic processing and integrative 175 functions of the ventral stream of speech formation, connecting the frontal and dorsolateral 176 prefrontal cortices to occipital and temporal gyri (15). Further from semantic processing, IFOF 177 actively participates in verbal-visual incongruence recognition and also in word recognition (14). 178 In a maturation point of view, these results confirm a temporal significance for critical changes in 179 microstructure of the IFOF as previously reported, and suggest that the degree of structural 180 maturity in fibers is determined by the duration of being bilingual (16). In another study Mohades 181 et al. revealed that maximal anisotropic values changes in the left IFOF occur with simultaneous 182 bilinguals or those using a merging of both languages (14). 183

There are also longitudinal reports studying the process of brain structural and functional changes 184 during L2 acquisition. One striking result was obtained from a cohort study by Schlegel et al (17), 185 who followed two groups of English monolinguals with the experiment group receiving intensive 186 Chinese language acquisition. Even as early as nine months of learning, higher FA values in "late" 187 new language learners was obvious, especially in the genu of CC and left frontal language 188 associative areas. This is in consort with advantageous effects of second language (L2) learning in 189 brain microstructural changes and improvements in connectivity, emerging even after short term 190 intervals in sequential bilinguals. More interesting was that with expansion of vocabulary domain 191

i.e. the ability to remember and use new words in new learners of a foreign language, FA values 192
of the mentioned fibers changed respectively. 193

Hosoda and his colleagues (18) report results of a cross-sectional study followed by their 194
longitudinal follow up on bilinguals after they had received 4 months of intensive language 195
training. Not only the baseline FA of Japanese-English proficient individuals was higher in the 196
arcuate fasciculus, inferior frontal gyrus (IFG), caudate nucleus and superior temporal gyrus, but 197
also these values tended to increase proportionately with L2 learning. Finally, when L2 training 198
was halted for one year, DTI findings certified a regression to start point values, the same as their 199
matched counterpart monolinguals. This brings us to realize the immense potential of brain 200
plasticity in language learning, compatible with constant active use of the second language in daily 201
life (19). 202

Arcuate fasciculus is part of the dorsal speech stream, mapping the heard language onto motor 203
areas for speech. This area is a network of three distinct cortical areas, the left inferior frontal 204
gyrus, left middle temporal gyrus, and left inferior parietal lobe (20). Together these results 205
underscore the bilingual advantage theory (3), connecting the neuroanatomical changes in fibers 206
to physiological function in bilingual brain. 207

Through Diffusion MRI connectometry, we identified white matter areas with increased 208
quantitative anisotropy in bilingual adults with long immersion period, compared to healthy age- 209
sex matched individuals. According to Yeh et al. (21) quantitative anisotropy (qa), is a measure 210
water density in a voxel or tract, rather than diffusivity, as measured by other diffusion variables 211
such as fractional anisotropy or mean diffusivity. Indeed, qa gives more accurate information about 212
normal/healthy distribution of water along the length of a fiber or within a voxel, and thus is a 213

diffusivity marker of a healthy fiber. QA is less sensitive to axonal degeneration than FA and is hence a marker of choice in comparing the two groups of healthy adults in our population.

Our results on significant increased qa in bilateral cingulum, inferior front occipital fasciculi, arcuate fasciculi, and corpus callosum, are in consort with previously reported areas. Apart from Aboutaleb et al. who suggested a role for ACC cortex in language switching and ability in non-verbal conflict, nothing was previously reported on cingulum structural adaptations in learning a new language. The cingulum is critical for emotion formation, executive function, visuospatial skills and attention. As a whole, cingulum is suggested to play a multimodal role in cognitive function and is highly active while performing cognitive tasks, especially those demanding a complex of executive and intellectual function (22). On the contrary, posterior cingulate areas are in a relative deactive state during cognitive tasks and accordingly regulate "internally directed cognition" or default mode self-awareness function of the brain (23).

Immersion period analysis revealed almost the same results of differentiation analysis, except for the AF, that did not show significant difference in qa value when regressed against immersion period, even with an FDR of 0.075. This might suggest that the aforementioned white matter changes in our sample of young healthy bilingual individuals, who had commenced their language learning no earlier than 4 years old, are mostly results of brain plasticity and active use of a second language rather than a developmental phenomenon of early L2 learning.

Structural connectivity in developing brain and brain structural maturation occurs in consort with normal developmental stages, aging process, and learning new skills. One major limitation of this study was the small number of cases in both groups, yielding to high FDR values. It is justifiable to repeat this study with larger sample group while taking into account the individual and social

variables, most importantly language proficiency, use of language, and immersion period as 236
probable confounding developmental factors. 237

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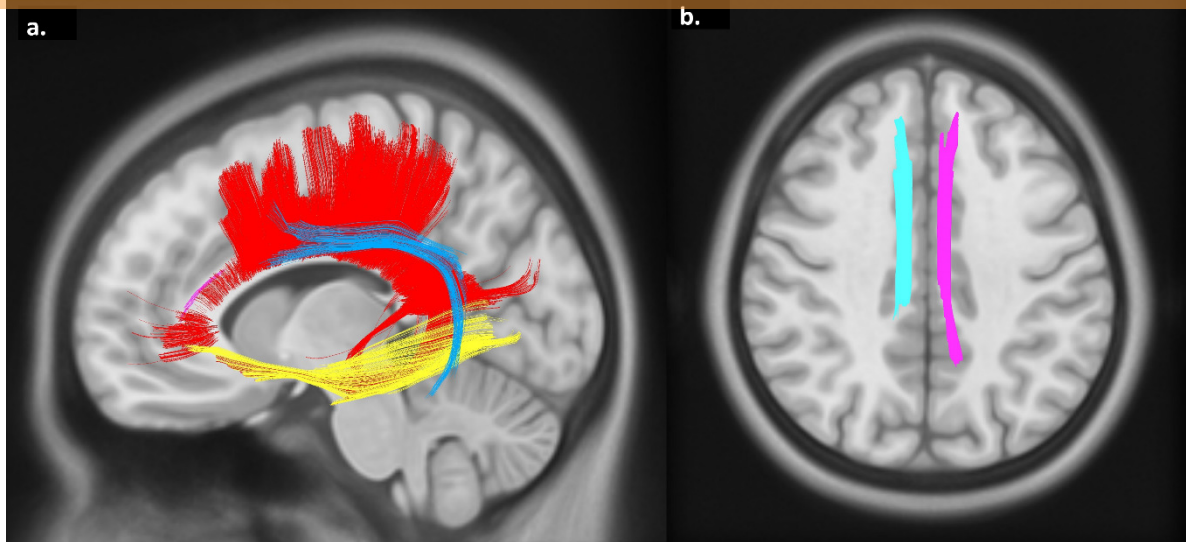


Figure 1. Brain pathways with increased QA in sequential bilingual adults compared to healthy monolingual controls a) corpus callosum, bilateral arcuate fasciculi and left inferior fronto occipital fasciculus, b) bilateral cingulum

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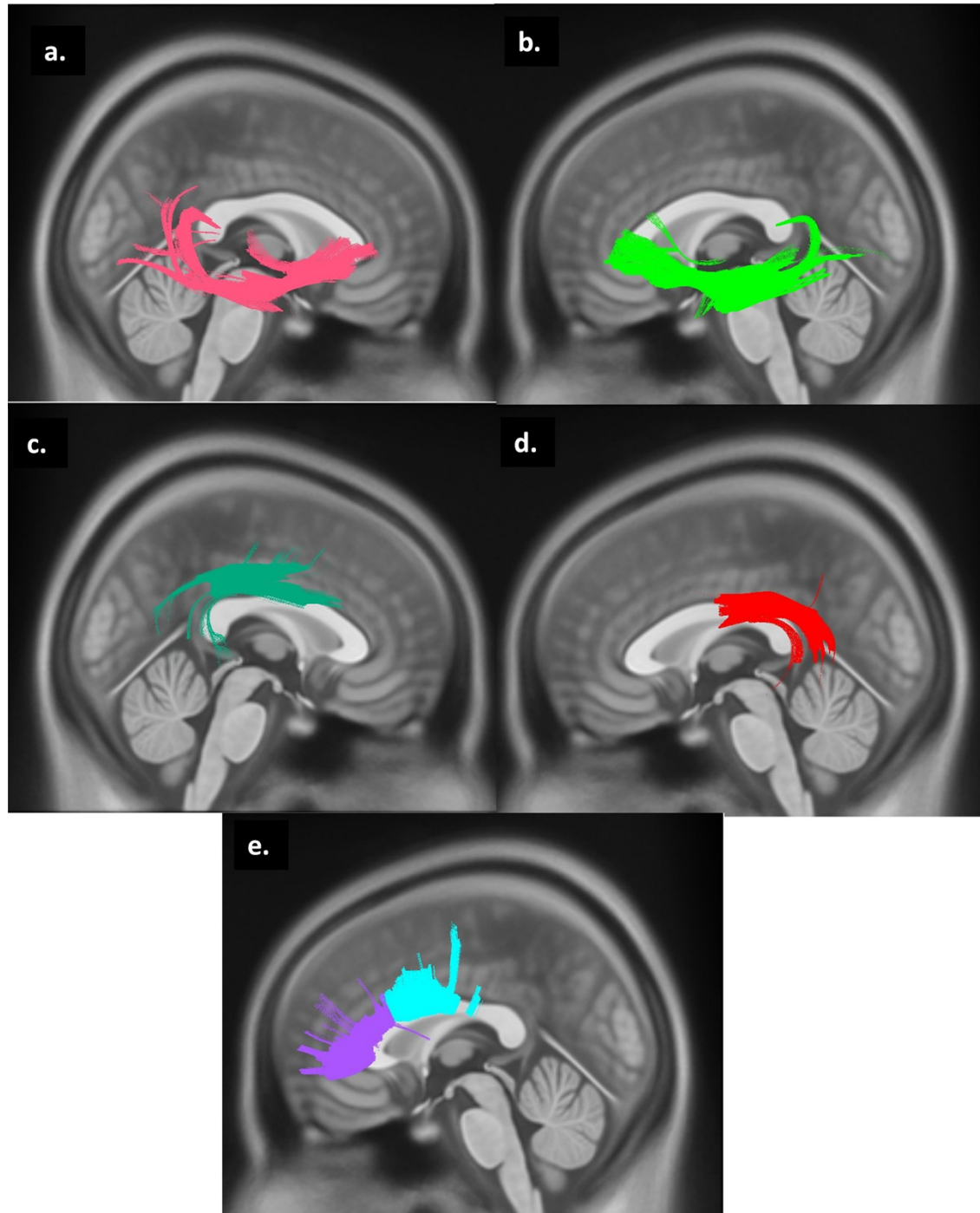
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Figure 2. Brain pathways with positive correlation with immersion period in a group of sequential bilingual adults a & b) right and left inferior fronto occipital fasciculi, c & d) right and left arcuate fasciculi and e) genu and body of corpus callosum