Grey matter reduction in the occipitotemporal cortex in Spanish children with dyslexia: a voxel-based morphometry study

Jesús Adrián-Ventura[1*], Manuel Soriano-Ferrer[2], Paola Fuentes-Claramonte[1], Manuel Morte-Soriano[1], Maria Antònia Parcet[1], & César Ávila[1]

[1] *Department of Basic and Clinical Psychology and Psychobiology, Jaume I University, 12071 Castellón, Spain.*

[2] *Department of Developmental and Educational Psychology, University of Valencia, 46010 Valencia, Spain.*

Corresponding author:

Jesús Adrián-Ventura (jadrian@uji.es) / +34964387664

Basic and Clinical Psychology and Psychobiology, Jaume I University. Avda. Sos Baynat, s/n. E-12071 Castellón, Spain.

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Structural and functional neuroimaging studies have reported brain alterations in occipitotemporal, temporoparietal, and left frontal areas in dyslexic patients. These areas have been linked to reading skill impairments, due to their involvement in word recognition and processing. However, most of the patients in these studies were speakers of languages with a deep orthography. In this study, we used voxel-based morphometry (VBM) to investigate brain differences in grey matter volume associated with a transparent language in a sample of 25 native Spanish participants (13 dyslexic and 12 non-dyslexic children). Results revealed a volume reduction in the left occipitotemporal cortex and right cerebellum in dyslexics. Significantly, the reduction in occipitotemporal areas has been previously linked to reading in transparent languages. Our results support previous studies and are consistent with the idea that reading problems in languages with a shallow orthography are related to the ventral reading network.

Keywords: developmental dyslexia; voxel-based morphometry; occipitotemporal cortex; shallow orthography

Introduction

Developmental dyslexia is a reading disability of neurological origin with a prevalence of between 3.2% to 5.9% in Spain (Jiménez, Guzmán, Rodríguez, & Artiles, 2009). It persists throughout life despite adequate intelligence, education, and socioeconomic background to learn to read (Soriano-Ferrer, Echegaray-Bengoa, & Joshi, 2016). Individuals with dyslexia have difficulties with accurate or fluent word recognition and spelling (American Psychiatric Association, 2014; Peterson & Pennington, 2012; Snowling & Hulme, 2013). Specifically, studies relating to different orthographies suggest that reading accuracy problems are more pronounced in opaque languages, given that there are no regular grapheme-to-phoneme rules, and it is necessary to integrate more than grapheme-level information (specifically, phonological) to correctly read a word (Martin, Kronbichler, & Richlan, 2016; Price, 2012; Richlan, 2014). By contrast, in relation to transparent languages, the main difference between individuals with and without dyslexia is a reading speed deficit (Wimmer & Schurz, 2010). For this reason, reading speed/fluency is considered the main indicator of dyslexia for transparent languages such as Spanish (Jiménez, Rodríguez, & Ramírez, 2009; Serrano & Defior, 2008; Soriano-Ferrer & Miranda, 2010; Suárez-Coalla & Cuetos, 2012), whereas reading accuracy is less affected. Initially, dyslexic children present a deficit in the processing of phonological information, and so they cannot achieve an adequate level of phonological awareness (Melby-Lervåg, Lyster, & Hulme, 2012), this being essential for the posterior acquisition of automated grapheme-phoneme conversion skills (McCandliss & Noble, 2003; Norton, Beach, & Gabrieli; Peterson & Pennington, 2012; Richlan, 2012). As a result, children with poor phonological skills (i.e. lower phonological awareness) show deficits in learning to read in both shallow and deep orthographies (Peterson & Pennington, 2012). Thus, deficits in reading could be attributed to a dysfunction in phonological processing-related brain regions that alters the phonological representation of speech sounds.

Word recognition impairment is also a key aspect of reading disruption, which can produce complex, and slow, learning of the necessary skills for the integration of multiple visual, linguistic, cognitive, and attentional processes (Norton et al., 2015). Neuroimaging studies have revealed that the left occipitotemporal (OT) regions are the brain areas most involved in word reading (Fiez & Petersen, 1998; Price, 2012; Price & Devlin, 2011; Seghier & Price, 2011). Among them, the visual word form area (VWFA), located in the left fusiform gyrus (FG), plays a prominent role in the processing of written stimuli acquired through experience (Cohen et al., 2000, 2002; Dehaene & Cohen, 2011;

McCandliss, Cohen, Dehaene, 2003); this is due to its involvement in rapid whole-word recognition processes. Furthermore, the neural reading system also involves the left superior (STG) and middle (MTG) temporal gyrus, the bilateral inferior parietal lobe (IPL), and the left inferior frontal (IFG) and precentral gyrus (Hadzibeganovic et al., 2010; Martin, Schurz, Kronbichler, & Richlan, 2015; Price, 2012). Functionally, this network has traditionally been divided into two different streams, as stated in the dual-route model (Coltheart, Rastle, Perry, Langdon, & Ziegler, 2001) of reading. This computational model proposes two distinct routes that work in parallel and are necessary for the correct integration of written or spoken stimuli (inputs) with appropriate responses (outputs). On one hand, the ventral, lexical route connects the left ventral OT cortex and MTG with the ventral IFG, and is associated with semantic processing. On the other hand, the dorsal, non-lexical phonological route links the STG and IPL with the dorsal precentral gyrus, and is involved in letter-to-sound and sensorimotor conversion (see review by Price, 2012). Moreover, in a previous study, Rueckl et al. (2015) demonstrated that these areas were also activated regardless of the orthographic depth, postulating a universal sign of reading-related activations across languages. However, as suggested in previous studies, the specificity of each route could be modulated by the orthographic depth, thus showing common but specific differences based on language transparency.

The most common functional brain underactivations in both child and adult dyslexics, compared to controls, are mainly localized in brain areas within this reading network (Maisog, Einbinder, Flowers, Turkeltaub, & Eden, 2008; Richlan, Kronbichler, & Wimmer, 2011). Specifically, in the meta-analysis by Richlan et al. (2011) comparing adult vs. children studies, the authors reported an underactivation of the ventral OT cortex in both adults and children with dyslexia, however, this underactivation was more prominent in children. By contrast, although adults with dyslexia also showed underactivations in the ventral OT cortex, predominant underactivation foci in TP areas were observed. Taken together, these lower activations in reading-related areas may reflect a lack of engagement in the demands of reading, leading to poor acquisition of reading-related skills (Norton et al., 2015; Richlan et al., 2011).

Previous meta-analyses of studies carried out in adults and children have shown that some anatomical deficits might underlie these functional differences in dyslexics (Eckert, Berninger, Vaden, Gebregziabher, & Tsu, 2016; Linkersdörfer, Lonnemann, Lindberg, Hasselhorn, & Fiebach, 2012; Richlan, Kronbichler, & Wimmer, 2013). All the studies included in these meta-analyses coincide in observing in dyslexics reduced grey matter volume (GMV) or density in the left supramarginal/posterior superior temporal sulcus (SMG/pSTS). However, they differ in finding reductions in GMV in other areas of the language network, including reading-related areas such as the left lateral prefrontal cortex (pars orbitalis), the fusiform, and the cerebellum (Eckert et al., 2016; Linkersdörfer et al., 2012). Correlational data have additionally shown that the GMV in these regions is associated with performance in language and reading tasks (Dole, Meunier, & Hoen, 2013; Jednoróg et al., 2015; Kronbichler et al., 2008; Pernet, Poline, Demonet, & Rousselet, 2009).

Different factors may contribute to explaining the variability in the observed results. A qualitative coordinated-based meta-analysis on functional studies compared meta-analytic maps from studies with participants from shallow orthographies to maps from studies with participants from opaque orthographies highlighting the relevance of orthographic depth (Martin et al., 2016). When studying the maps separately, a common pattern of underactivation is observed in the left IPL and left OT cortex in languages with both shallow (SO) and deep (DO) orthographies. These results support the idea of universal brain deficits in both transparent and opaque languages, as postulated in previous studies (Paulesu et al., 2001; Pugh, 2006). However, the analysis of SO specifically revealed that the left occipitotemporoparietal cortex (with the main underactivation in the FG) was the only relevant region, whereas in the analysis of DO, this region was less significant, and other frontal and temporoparietal (TP) areas were relevant. The direct comparison of the two orthographies revealed a similar pattern. The SO > DO contrast showed a higher convergence of underactivation mainly in the left FG, as well as other less relevant differences in other brain areas, whereas the opposite contrast revealed main underactivations in TP areas. These differences support the idea that specific brain alterations in SO may be more focused on the posterior occipital and fusiform areas compared to DO, and that previous meta-analyses may be biased by this factor.

In line with this perspective, we observed that almost all the voxel-based morphometry studies with dyslexic children were conducted in languages with a DO. As far as we know, three studies were the exception. In a sample with German speakers, Kronbichler et al. (2008) found that the dyslexic readers had less GMV in the left and right FG, the right SMG, and the bilateral anterior cerebellum, compared to healthy controls. These results were not replicated in the study by Jednoróg, Gawron, Marchewka, Heim, and Grabowska (2014) with Polish participants, as they only observed a GMV reduction in the left IFG. However, in the multicentre study by Jednoróg et al. (2015) with French, Polish and German speakers, the authors found a GMV decrease in the right cerebellum declive (for the German dyslexic sample) and in the left ITG (for the Polish sample). Thus, the possibility of a different pattern of structural deficits in dyslexic children using languages with a more transparent orthography remains open.

The aim of this study was to investigate brain structural deficits in a group of Spanish dyslexic children. This study contributes to the characterisation of anatomical differences in children who use a language with a SO by analysing differences in GMV between dyslexic and non-dyslexic children. Previous functional and structural meta-analyses reported alterations in both TP and OT areas in patients with dyslexia. Nevertheless, these brain abnormalities vary depending on the orthographic depth (Richlan, 2014). Specifically, a functional underactivation in the left anterior ventral OT cortex has been found only in SO (Martin et al., 2016). Thus, in this study we expected to find GMV alterations in brain areas within the reading network in dyslexics, compared to controls, especially in left ventral OT regions.

Materials and methods

Participants

A total of 25 volunteers participated in the study. All the children were students from different schools in Valencia (Spain) and had average socio-economic backgrounds. All the subjects were Caucasian and spoke Spanish as their native language. They ranged in age from 9 to 14 years (mean age = 12.18; SD = 1.68). There were 15 boys and 10 girls in the sample. These children were classified into two groups: (a) the developmentally dyslexic group, which was made up of 13 students (mean age = 11.91; SD = 1.63; 7 males); and (b) the control group, which consisted of 12 normal readers (mean age = 12.48; SD = 1.76; 8 males). The average intelligence quotient (IQ) was 109 (SD = 9.70; range 97 to 130) for the dyslexic group and 118.25 (SD = 13.69; range 91 to 137) for the control group. Additionally, both groups were balanced according to participants' handedness (5 left-handed participants took part in the study; 3 in the dyslexic group and 2 in the control group). Two-sample t-tests showed no significant differences between the two groups in: age, $t = -0.85$, $p = 0.405$, $d = -0.34$; IQ, $t = -1.96$, $p = 0.062$, $d = -0.78$;

gender, $\chi^2 = 0.43$, $p = 0.513$, $\eta^2 = 0.02$; or handedness, $\chi^2 = 0.16$, $p = 0.689$, $\eta^2 < 0.01$.

Finally, in relation to parental occupations, all fathers in both groups had a paid job, whereas the number of mothers with a paid job was lower (69% for the dyslexic group and 50% for the control group, with no differences between groups ($p > 0.05$)). Occupations of fathers and mothers were distributed into occupations with four levels of skills, in accordance with the international standard classification of occupations (ILO, 2012). There were no significant differences in the fathers' occupations, $\chi_{(3)}^2 = 2.56$, $p = 0.464$, nor in mothers' occupations, $\chi_{(4)}^2 = 5.37$, $p = 0.252$, between both groups. Also, around 60% of fathers in both groups had a skill 2 occupation.

Tasks

Culture fair (or free) intelligence test (Scale 2, Form A, Cattell & Cattell, 1950/1989): this test measures general mental capacity without the interference of cultural basis. We employed the Spanish version of this test (Seisdedos, de la Cruz, Cordero, & González, 1991).

Word and Pseudoword Reading Skills (PROLEC-R, Cuetos, Rodríguez, Ruano, & Arribas, 2007; PROLEC-SE, Ramos & Cuetos, 2003): these tests require the correct identification of 40 words that vary greatly in frequency, length and linguistic structure (CCV, CVV, CVC, CCVC, CVVC and VC, where C = consonant and V = vowel). Pseudoword reading consists of pronouncing 40 pseudowords constructed by changing or adding one or two letters to each of the 40 words on the reading test. In both cases, the child's score consists of an accuracy measure (correct words and pseudowords read) and the total time used to finish the task.

Rapid Automatized Naming-Rapid Alternating Stimulus (RAN/RAS, Wolf & Denckla, 2005). The RAN test consists of four rapid automatized naming subtests (letters, numbers, colours, and objects), whereas the RAS test is divided into two subtests: the 2-set (letters and numbers) and the 3-set (letters, numbers, and colours). Each RAN subtest is comprised of five stimuli randomly repeated ten times, arranged in 5 rows of 10 items, for a total of fifty items. RAS subtests are displayed in the same way as RAN subtests, but with 10 (2-set) and 15 (3-set) token items. For both the RAN and RAS tests, children are asked to name each stimulus accurately and as fast as possible. Scores are based on the total naming time required on each subtest.

Verbal fluency test (FAS; Benton & Hamsher, 1989): this test evaluates phonetic and semantic fluency. For phonetic fluency, children are asked to evoke as many words starting with the letter "F", "A" or "S" as they can in one minute on each of three trials. The global score is obtained by computing the total number of words for the three trials, excluding repetitions, proper names, and numbers. For semantic fluency, children have to say as many words as they can within a given semantic category, in this case "animals", in one minute, excluding repetitions and errors.

Clinical diagnosis

Information on the children's academic and developmental history was obtained from the school records available to the school's psycho-educational support services. The presence of developmental dyslexia was determined by using the DSM-5 criteria (American Psychiatric Association, 2014). Specifically, the criteria for the assessment were: (a) poor academic performance on reading, according to a teacher's rating report, and average achievement in other academic areas (e.g. arithmetic); (b) scores of 80 or higher on an intelligence test (Cattell & Cattell, 1950/1989), in order to exclude students with intellectual deficits; and (c) no evidence or history of neurological damage, environmental disadvantage, emotional disturbance, hearing or vision abnormalities, or any other major handicapping condition (exclusion criteria); (d) the reading achievement criteria adopted in this study have been commonly used in the learning disabilities literature.

Specifically, children with developmental dyslexia were defined as those who had reading scores of 1.5SD below the mean of the standardization sample on at least one subtest (word accuracy, pseudoword accuracy, word reading time, or pseudoword reading time) on the Reading Processes Assessment Battery (PROLEC-R, Cuetos et al., 2007; PROLEC-SE, Ramos & Cuetos, 2003). In order to clarify the differences between the groups, we selected our control group as the reference (all of them obtained normal scores on all the subtests compared to the standardization sample). Thus, all the dyslexic children obtained scores of 1.5SD below the mean of the control group in at least two areas (two participants failed on two skills, two participants failed on three skills, and the rest failed on all four skills). Finally, we decided to add the accuracy scores (word accuracy plus pseudoword accuracy) and the reading time scores (word reading time plus pseudoword reading time) in two separate variables. With these new variables, we observed that all the children, except two, scored higher (1.5SD) than the control group on reading time, but all the children, without exception, showed a lower score (1.5SD) than the control group on reading accuracy. Differences between groups on these two variables can be seen in Figure 1. All children with developmental dyslexia, as stipulated in the current regulations in Spain for students with learning disabilities, attended special education classes (i.e. resource rooms) three hours a week in their respective schools, while the non-impaired children (as normal readers) attended regular classes.

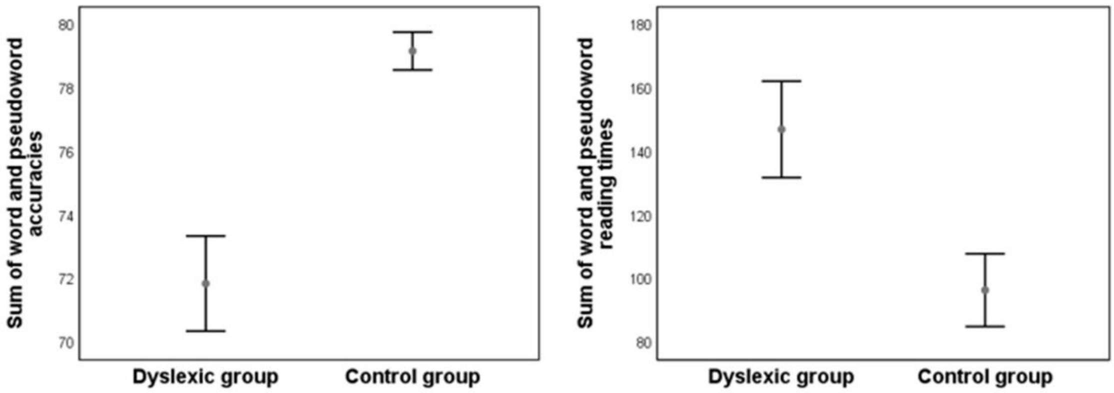

Figure 1. Differences in reading performance between the dyslexic and control groups. Error bars indicate 95% confidence intervals.

Structural magnetic resonance imaging

Data acquisition

Before formal MRI scanning, all the children were familiarized with the experimental procedure and the noise made by the scanner. They were also informed about the duration of each session and setting details. All images were acquired on a 3T Philips Achieva scanner. A high-resolution structural T1-weighted sequence was obtained (TR/TE = 8.4/3.8 ms, matrix = 320 x 320 x 250, voxel size = 0.75 x 0.75 x 0.8 mm^3).

Preprocessing

VBM was performed with the VBM8 toolbox for the SPM8 package (Wellcome Department of Imaging Neuroscience, London, UK; http://www.fil.ion.ucl.ac.uk/spm/software/spm8/). First, a customized tissue probability map (TPM) template was created based on the paediatric sample of the Template-O-Matic (TOM8) toolbox (http://dbm.neuro.uni-jena.de/software/tom/), including age and gender to obtain a more accurate template (according to our sample). The resulting TPM template was then resliced with trilinear interpolation to a final voxel size of 1 x 1 x 1 mm^3. After that, images were segmented into grey matter, white matter, and cerebrospinal fluid, and they were registered through affine regularization to the average size template. Subsequently, as suggested in the manual when studying samples from children, images were normalized to the low-dimensional SPM default template (MNI152) and modulated by the non-linear components derived from this spatial normalization. Thus, the volume variations resulting from the normalization were corrected in the resulting grey matter volume maps. Additionally, the “non-linear only” modulation applies the correction for individual brain sizes directly to the images, so it is not necessary to introduce total intracranial volume as nuisance covariate in statistical models. Finally, images were smoothed with a 10-mm Gaussian kernel.

Statistical analyses

Group differences in grey matter volume were evaluated with a whole-brain two-sample t-test. We also added age, gender, and IQ as nuisance covariates in order to control possible effects on brain volume. Participants’ handedness was considered as nuisance covariate, however this variable was not included in the model given that showed multicollinearity with the IQ and sex covariates (0.37 and 0.39, respectively). Permutation-based nonparametric testing (5000 permutations) was carried out using the TFCE (threshold-free cluster enhancement) Toolbox (Smith & Nichols, 2009). The statistical threshold to control for multiple comparisons was set at $p < 0.05$ familywise error (FWE) corrected. In addition, due to our small sample size, we applied the T statistic rather than the cluster enhancement, as recommended for distributions with low degrees of freedom (Nichols & Hayasaka, 2003). This permutation-based approach (without cluster enhancement) is very similar to the SnPM approach (Statistical Nonparametric Mapping; Nichols & Holmes, 2002), however, with the TFCE toolbox there is no need to define an initial cluster-forming threshold.

Results

Behavioural results

Means and standard deviations for each variable appear in Table 1. We observed significant differences between groups in word reading accuracy ($t = -8.55$, $p = 0.000$, $d = -3.37$) and pseudoword reading accuracy ($t = -6.78$, $p = 0.000$, $d = -2.67$), with large effect sizes in both cases. In addition, t-tests also showed significant differences between the two groups in word reading time ($t = 5.15$, $p = 0.000$, $d = 2.08$) and pseudoword reading time ($t = 5.15$, $p = 0.000$, $d = 2.07$), also with large effect sizes. In regard to verbal phonetic fluency, children with developmental dyslexia obtained significantly lower scores (FAS; $t = -3.22$, $p = 0.004$, $d = -1.28$). However, there were no significant differences between groups in verbal semantic fluency ($t = -1.84$, $p = 0.084$, $d = -0.74$). In addition, children with developmental dyslexia took longer to name familiar stimuli, showing lower naming speed with alphanumeric stimuli: RAN letters, $t = 4.04$, $p = 0.001$, $d = 1.62$, and RAN numbers, $t = 4.18$, $p = 0.001$, $d = 1.65$; with non-alphanumeric stimuli: RAN object, $t = 4.18$, $p = 0.001$, $d = 1.65$, and RAN colour, $t = 3.80$, $p = 0.001$, $d = 1.54$; and even with alternating stimuli: RAS letter/number, $t = 3.09$, $p = 0.005$, $d = 1.23$, and RAS letter/number/colour, $t = 3.78$, $p = 0.001$, $d = 1.53$, also showing large effect sizes.

Table 1. Two-sample t-tests between the dyslexic and control groups in reading performance

	Dyslexic group M (SD)	**Control group M (SD)**	**t-value**	**p**	**Cohen's d**
Word reading accuracy	37.23 (1.09)	39.92 (0.29)	-8.55	0.000	-3.37
Word reading time	64.54 (14.13)	39.25 (9.84)	5.15	0.000	2.08
Pseudoword reading accuracy	34.62 (2.29)	39.25 (0.87)	-6.78	0.000	-2.67
Pseudoword reading time	82.46 (13.65)	57.14 (10.56)	5.15	0.000	2.07
Phonetic fluency (FAS)	19.54 (5.81)	29.00 (8.72)	-3.22	0.004	-1.28
Semantic fluency ("animals")	15.77 (2.39)	18.50 (4.60)	-1.84	0.084	-0.74
RAN (object)	50.77 (11.96)	36.00 (4.26)	4.18	0.001	1.65
RAN (colour)	51.08 (11.42)	37.25 (5.51)	3.80	0.001	1.54
RAN (number)	30.23 (6.02)	22.42 (2.94)	4.18	0.001	1.65
RAN (letter)	29.54 (4.63)	22.58 (3.92)	4.04	0.001	1.62
RAS (letter/number)	34.85 (6.67)	26.00 (7.64)	3.09	0.005	1.23
RAN (letter/number/colour)	40.92 (9.68)	28.08 (6.93)	3.78	0.001	1.53

VBM results

Results of the two-sample t-test comparison appear in Table 2 and Figure 2. The dyslexic group showed a lower GMV than the control group mainly in the bilateral occipital cortex (cuneus, lingual gyrus, and calcarine sulcus), the bilateral precentral gyrus, and the inferior occipital/fusiform gyrus (especially in the left hemisphere). The correction for multiple comparisons was set at the pre-established threshold ($p < 0.05$ FWE, $k > 750$), using a non-parametric analysis method. The opposite contrast, Dyslexics > Controls, did not yield any significant differences.

Additionally, we performed further analysis in order to observe the power of our results. These results are shown in the Supplementary material.

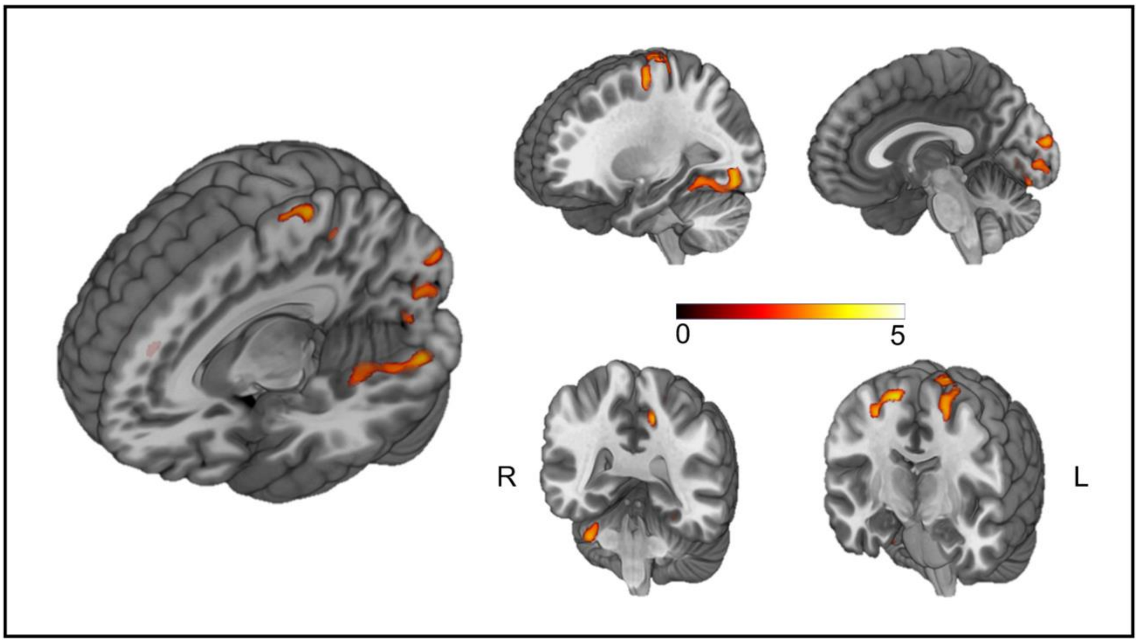

Figure 2. Whole-brain grey matter volume reduction in brain areas in the dyslexic group compared to the control group (Control > Dyslexic). R: right; L: left. Bar colour represents T values.

Table 2. Brain areas showing a grey matter volume reduction in the dyslexic group compared to the control group (Control > Dyslexic)

Region	**Hemisphere**	**Coordinates MNI x, y, z**	**t-scores**	**k-voxels**
Cuneus	R	10, -94, 23	4.08	6071
Cuneus[a]	L	-7, -97, 15		
Lingual[a]	R	7, -86, -8		
Calcarine[a]	L	-13, -96, -6		
Lingual[a]	L	-9, -84, -14		
Calcarine[a]	R	11, -78, 4		
Paracentral lobule	L	-15, -23, 77	3.72	2208
Precentral[a]	L	-28, -14, 72		
Frontal superior[a]	L	-23, -10, 59		
Inferior occipital	L	-33, -77, -12	3.55	2131
Fusiform[a]	L	-31, -51, -10		

Fusiform	R	30, -54, -14	2.95	1152
Cerebellum lobe 6[a]	R	32, -65, -20		
Superior frontal	R	17, -11, 62	3.99	1138
Precentral[a]	R	29, -12, 51		
Middle frontal	L	-29, 44, 21	4.15	857
Precuneus	L	-17, -46, 56	4.58	847
Cerebellum lobe 6	R	33, -39, -37	3.29	838
Cerebellum lobe 4-5[a]	R	26, -34, -25		
Superior frontal medial	L	-6, 46, 35	3.64	807

R: right; L: left; [a] subpeak within cluster; $p < 0.05$, FWE corrected.

Discussion

To the best of our knowledge, our study is the first to investigate alterations in GMV in Spanish children with dyslexia compared to a healthy control group. Considering the previous literature, Spanish is one of the shallowest languages used to investigate these differences. We found that the dyslexic children had less GMV principally in the left occipitotemporal cortex, the right cerebellum and the bilateral precentral gyrus than the healthy control group. Our results raise the possibility that language transparency may be a relevant factor in dyslexia research and that previous meta-analyses on this topic may be biased by the high percentage of studies conducted in opaque languages.

In relation to behaviour, our data showed that children with developmental dyslexia were slower and less accurate in performing reading tasks. These results agree with findings from other studies conducted in the Spanish language with a reading level match design (Jiménez, Rodríguez, et al., 2009; Serrano & Defior, 2008). The lower word and pseudoword accuracy results suggest that a phonological deficit exists in children with developmental dyslexia who learn in a SO. However, the fact that these children were significantly slower on word and pseudoword reading is an indication that phonological processing is not automatic, as shown in studies carried out in Spanish (Jiménez, Rodríguez, et al., 2009; Rodrigo & Jiménez, 1999; Serrano & Defior, 2008) and other shallow orthographies (Tressoldi, Stella, & Faggella, 2001; Wimmer, Mayringer, & Landerl, 1998). Moreover, our results show reading-related cognitive deficits in verbal phonetic fluency, but not in verbal semantic fluency – consistent with other studies carried out in different orthographies (e.g., Frith, Landerl, & Frith, 1995; Reiter, Tucha, & Lange, 2005). Furthermore, our findings show that naming speed is also a reading-related cognitive deficit in a SO, which is consistent with results from studies carried out in Spanish (Jiménez, Rodríguez, et al., 2009; Lopez Escribano, 2007) and in other orthographies (see review in Norton & Wolf, 2012).

Regarding VBM analysis, our results comparing dyslexic and control children are partially consistent with previous VBM studies. According to different meta-analyses, the main differences between dyslexics and controls are located in left OT (i.e. the VWFA, within the FG) and bilateral TP areas (STG and SMG/pSTS), left IFG (pars orbitalis) regions, and in the cerebellum bilaterally (Eckert et al., 2016; Linkersdörfer et al., 2012; Richlan et al., 2011). Our data did not confirm GMV reductions in TP and IFG regions, as reported by previous studies (Eckert et al., 2016; Richlan et al., 2011). However, our results found reductions in GMV in other areas of the language network related to dyslexia, such as the FG (reported in Linkersdörfer et al., 2012) and the right cerebellum (reported in Eckert et al., 2016; Linkersdörfer et al., 2012). Importantly, although the GMV reduction in the FG was found bilaterally, in line with previous reports (Brambati et al., 2004; Kronbichler et al., 2008; Tamboer et al. 2016), a certain degree of asymmetry is observed in the results, showing a more left-lateralized pattern. A further issue is the functional correlates of this reduced volume. For instance, Tamboer et al. (2016)

demonstrated that the functional correlates with reading performance were observed only for the left hemisphere.

The discrepancy between our results and previous reports may stem from the fact that most of the previous studies included in meta-analyses were carried out in languages with different orthographic depth and combining children and adult populations. As far as we know, only four morphometric studies (voxel- and surfaced-based) form the exception as they examined German (Jednoróg et al., 2015; Kraft et al., 2015; Kronbichler et al., 2008) and Polish (Jednoróg et al., 2015, 2014) children (both languages are considered transparent; see Kusiak, 2013; Liu & Cao, 2016; Martin et al., 2016). The study by Jednoróg et al. (2014) only reported differences in the IFG, whereas the study by Kronbichler et al. (2008) and Kraft et al. (2015) reported differences in the cerebellum, the FG and the SMG (as well as other areas; see Kronbichler et al., 2008). Nonetheless, Jednoróg et al. (2015) identified grey matter reductions in the right cerebellum (in the German sample) and in the left ITG (in the Polish sample). All this evidence converges in locating the main differences in brain volume in the inferior temporal lobe, the cerebellum and the inferior frontal cortex more than in other regions (i.e. TP areas), especially in children who are native speakers of transparent languages. This pattern has also been shown in studies with dyslexic adults that used transparent languages (Brambati et al., 2004; Tamboer, Vorst, Ghebreab, & Scholte, 2016); however, results have also been reported showing alterations in TP areas (Steinbrink et al., 2008; Tamboer et al., 2016). So, these previous reports with SO are in line with our results, particularly with the brain alterations observed in the OT cortex, thus adding further evidence about the implication of this area in reading processes in SO. Consequently, the brain differences between adults and children with dyslexia speaking a transparent language may show a different pattern more focused on posterior ventral areas (FG and OT cortex) and left IFG regions.

The data of our study were also coincident with the meta-analysis of functional studies of reading in dyslexics and controls (Martin et al., 2016). In this study, a common pattern of underactivation was observed in the left posterior ventral occipitotemporal (vOT) cortex independently of the language transparency. Thus, the underactivation in this region might be considered as a universal neurobiological hallmark of dyslexia (Martin et al., 2016; Paulesu et al., 2001; Pugh, 2006). Also, the dysfunction in this area for both DO and SO is compatible with the idea that the left vOT is responsible for lexical (fast visual-orthographic whole-word recognition) and sublexical (serial grapheme-phoneme conversion) processes (Martin et al., 2016; Richlan, 2012, 2014; Richlan et al., 2010). However, when the BOLD response was compared according to the transparency of the languages, the higher convergence of underactivation in dyslexic patients when using a DO was mainly located in the intraparietal sulcus (IPS), whereas this underactivation corresponded to the FG when using a SO (see Martin et al., 2016). This pattern aligns with the results obtained in the present study which shows a GMV reduction in the same area with a transparent language, like Spanish, contrasting with the distributed pattern observed in opaque languages (Martin et al., 2016). One interpretation of these differences relates to the cognitive processes involved in each case. Hence, although the brain areas involved in reading are similar in both SO and DO, some differences could appear based on specific psycholinguistic aspects (see the grain size theory; Ziegler & Goswami, 2005). Particularly, SO are characterised by single letter or letter-cluster conversion to single phonemes (i.e. small grain size) because the grapheme-phoneme rules are regular. By contrast, grapheme-phoneme conversion in DO is more complex because grapheme-phoneme rules are not consistent, and more letter units (from syllables to whole words) are needed (i.e. large grain size) (Richlan, 2014). Based on these differences, one might assume that grey matter reductions in dyslexics using a SO would be primarily linked to the FG (within the vOT cortex) because this region is involved in both lexical and sublexical processes (Martin et al., 2016; see Richlan et al., 2010 for a review). Additionally, given that the greater involvement of this region was evident on tasks requiring non-word processing in transparent languages (Martin et al., 2016), we could speculate that the reduced grey matter in this area might affect more sublexical processing (regular grapheme-phoneme conversion rules).

All this evidence is consistent with the idea that both the ventral and dorsal routes are involved in reading for transparent and opaque languages. However, when direct comparisons are made between both, the ventral route (lexico-semantic system, including the OT cortex) is more related to transparent languages, whereas the dorsal route (phonological system, including the TP cortex) is especially related to opaque languages (Martin et al., 2016). Thus, the different brain areas involved in languages with shallow and deep orthographies may be related to these different cognitive processes. However, it is important to note that many of the previous studies -and also meta-analyses- could be biased by their anglocentricity (Share, 2008), and more studies with transparent languages will result in better knowledge about the neurobiological basis of developmental dyslexia.

Additionally, we observed a GMV reduction in occipital areas (i.e. the cuneus, the calcarine sulcus and the lingual gyrus) in the dyslexic group that could explain some differences in reading processing. Interestingly, in a recent research by Jagger-Rickels, Kibby, & Constance (2018) with American children, the authors found a volume reduction in a large cluster, comprising the calcarine sulcus and lingual gyrus bilaterally. Furthermore, these results were similar with those reported in Jednoróg et al. (2014) with regard to Polish children, and in Eckert et al. (2005) with regard to English speaking children. Thus, these brain regions within the occipital cortex would be linked to deficits in reading; in particular, the lingual gyrus participates in visual rapid letter processing (Jagger-Rickels et al., 2018; Richlan et al., 2013), although unlike the FG (i.e. the VWFA), the lingual gyrus is not directly involved in letter processing, but in general shape processing (Mechelli, Humphreys, Mayall, Olson, & Price, 2000). Therefore, alterations in this area could lead to early deficits in letter/character recognition, thus affecting the information flow through the ventral reading stream.

Our results also show a reduced GMV in the right cerebellum in the dyslexic group. This reduction has been reported in previous meta-analyses (Eckert et al., 2016; Linkersdörfer et al., 2012). The cerebellum is involved in different skills needed for reading, including eye fixation, eye-voice coordination, and phonological and articulatory skills (Mariën et al., 2014; Stoodley & Stein, 2013). Thus, a reduced GMV in this area may affect the development of these skills via a deterioration in the cerebro-cerebellar loops (Stoodley & Stein, 2013), which can affect reading-related processes, such as word generation and speech production (Mariën et al., 2014; Stoodley & Schmahmann, 2009). Specifically, we found a grey matter reduction in the right lobule VI, a region previously found to be involved in language- and motor-related domains (Mariën & Beaton, 2014; Stoodley & Schmahmann, 2009), and therefore an alteration in this area might cause poor performance on reading tasks. The implication of the right cerebellum declive (lobule VI) in dyslexia has also been suggested in other studies (Jednoróg et al., 2015; Pernet et al., 2009), supporting the involvement of the right cerebellum in reading.

Regarding the aim of this study, we found grey matter alterations in Spanish dyslexics, when compared to a healthy control group. The most relevant differences are located within the occipitotemporal cortex and cerebellum. However, it is also important to note the limitations of our research – mainly the sample size and the differences in IQ (almost significant). We addressed this latter limitation by covarying IQ in the VBM model, along with age and gender, but still the balance between groups should be more accurate in future studies. In relation to the sample size, even though we used an appropriate non-parametric approach to our data (< 25 degrees of freedom), larger samples would be needed to better explore the brain alterations in patients with dyslexia. Also, using large samples would beneficially increase the power of the results, thus allowing a better identification - and higher sensitivity - of underlying effects (i.e. effects in TP or IFG regions in our study, see Supplementary material). Even so, the results of this study contribute to the characterization of the brain structure deficits in dyslexic children in a language with a shallow orthography, in this case Spanish, and it is the first study to compare dyslexic and non-dyslexic children in this language.

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Disclosure statement

The authors report no conflicts of interest.

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