FUNCTIONAL AND STRUCTURAL BRAIN IMAGING STUDIES OF DEVELOPMENTAL DYSLEXIA

A Dissertation
submitted to the Faculty of the
Graduate School of Arts and Sciences
of Georgetown University
in partial fulfillment of the requirements for the
degree of
Doctor of Philosophy
in Neuroscience

By

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Washington, DC
April 9, 2013
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ABSTRACT

Written language is relatively new from an evolutionary standpoint; however, reading has become an essential aspect of success in everyday life. As with other acquired skills, the ability to learn to read is variable amongst the population. Those with dyslexia struggle to acquire normal reading ability despite normal intelligence, access to education, and a motivation to learn. The brain basis of this neurobiological disorder has been studied at length, but questions remain as to the causality of the anatomical differences and how the brain changes with a reading intervention. This dissertation presents three studies addressing these questions. First, using a reading level matched design, reduction in gray matter volume (GMV) was observed in left middle/superior temporal gyrus when comparing dyslexic children to age-matched control children, consistent with prior studies. However, when compared to a group matched on reading level, these differences were no longer observed. Only right precentral gyrus showed differences compared to both control groups, calling into question the idea that left temporoparietal GMV is causal to the reading deficit in dyslexia. In the second study a reading intervention was given to a group of dyslexic children as well as either a developmental (no intervention) or active (math intervention) control. Here, the data showed an increase in activation in left inferior frontal gyrus that was specific to the reading intervention period and not driven by an underlying change in GMV. Also, bilateral activation of supramarginal/angular gyri prior to the intervention
predicted the amount of gain in single word reading standard score following the intervention suggesting functional integrity of this region represents a readiness for intervention. The final study used a subset of the subjects in the intervention study to examine changes in GMV following the intervention. The results showed changes bilaterally in the hippocampus, left precuneus, and right cerebellum, areas involved in more general learning and memory and mental imagery (a key component of the intervention). Together these studies provide insight into the dyslexic brain, and how reading experience (via reading ability or intervention) has an effect on both brain function and structure.
This dissertation is dedicated to my parents, Chip and Liz Krafnick, for always letting me pursue my interests without boundaries or judgment, and Amber Leaver for deciding to stay in D.C.
ACKNOWLEDGEMENTS

I would like to first and foremost thank Guinevere Eden for providing incredible mentorship in the completion of this work. I could not imagine a better relationship with a thesis mentor, and I am grateful for all of the help and insight in all aspects of the graduate experience. Thanks to Lynn Flowers for her expert insight and assistance with the work presented in this dissertation. My fellow lab mates past and present have also been a great help: Jeremy Purcell, Tanya Evans, Caitlin Taylor, Olumide Olulade, Megan Luetje, Eileen Napoliello, Shawn Nock, Ted Turesky, Justyna Mach, Allison Turza Bajger and Ashley Wall Piche. Not just for their help with the research but also for being good friends. Thanks to my thesis committee: Chandan Vaidya, Max Riesenhuber, Rachel Barr and Charles Perfetti for your all of your help, time and patience in helping me complete this work. I would also like to acknowledge and thank Daniel Walsh for helping forge my love of science. A special thanks to Andrew McElrone, my first science mentor, and the Saint Joseph’s University Biology Department, especially Karen Snetselaar and John Tudor for inspiring me to pursue research.

Finally I would like to thank my wife Amber, my parents Chip and Liz, my brothers Ryan and Trevor, and all of my friends for all of their love and support.

Acknowledgements for specific chapters are listed below:
Chapter I

This work was supported by the National Institutes of Child Health and Human Development (P50 HD40095, R01 HD056107). Thanks to Corina Moore, Emily Curran, Iain DeWitt, Alison Merikangas, Ashley Wall-Piche, Robert Twomey and Jenni Rosenberg, for their assistance in MRI data collection and Emma Cole, Martha Miranda and Gina Smith for collection of behavioral psychometric measures. We thank our subjects for their participation and the students, parents and staff at the Jemicy School, MD, for their involvement.

Chapters II and III

This work was supported by NICHD grants: P50 HD40095 and RO1 HD056107. Thanks to Ashley Wall, Emma Cole, Corinna Moore, Jenni Rosenberg, Iain DeWitt and Alison Merikangas for aiding with the data acquisition. Also, thanks our participants and their families for volunteering their time, the Jemicy School for allowing us to conduct the intervention at their school and the staff from Lindamood Bell Learning Processes for providing the intervention.
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INTRODUCTION

The earliest known examples of written language are generally attributed to the Sumerians and Egyptians some five thousand years ago\(^a\). With the advent of a written form of communication and record keeping also came the necessity to be able to process (that is, to read) that information. Reading has become virtually essential to success in everyday life from the home, to school, and the workforce, as it is required for basic navigation, formal education and most forms of employment. Furthermore, it is only more recently that the majority of the general population sets out to learn this skill, as access to education has steadily become more available throughout the world and across social classes. Being such a new skill, from an evolutionary standpoint, learning to read requires the brain to execute a function which it was not specifically evolved to support (Dehaene and Cohen, 2007). As with any skill, there will be a range in the ability of individuals to master that task. Much like mastering a musical instrument is not something everyone can easily do, not everyone is easily able to master reading. While it is not necessary that everyone becomes a concert pianist, it is important in our current society for people to learn to read. Thus, reading may be considered of interest for two broad reasons: First, as an acquired skill, reading is an example of a cognitive ability learned by a high percentage of the population over a specific and protracted period of time. Second, due to the important nature of acquiring this skill in everyday life, studying how one learns to read and what goes wrong in those that struggle to learn is of importance to both basic science and education.

\(^a\) Sumerian cuneiform and Egyptian hieroglyphs are both dated \textit{circa} 3000 B.C.
Studying dyslexia not only provides insight into the disorder itself, but also informs us about reading in general. While dyslexia research has been ongoing for many years, continued improvements in human imaging technologies have allowed for the asking and answering of new and interesting questions in regards to both dyslexia and reading. In this dissertation, I will describe three studies utilizing magnetic resonance imaging (MRI) techniques to study the brains of children with developmental dyslexia. The first study is an anatomical comparison of dyslexic children to two groups of typical reading controls. One group matched on chronological age, and a second (younger) group matched on reading level (Krafnick et al., Submitted). The final two studies follow a group of dyslexic children who have undergone reading intervention in order to try and improve their reading skills. One study uses functional MRI (fMRI) to ask if activity in certain brain regions prior to the intervention can predict reading gain following the intervention, and whether those areas are the same as those brain areas that change following the intervention. The other study uses MRI to probe which areas of the brain show a change in gray matter volume (GMV) following reading intervention (Krafnick et al., 2011). Together these studies are aimed at investigating the effects of reading experience and reading intervention on the brain. In this section I will introduce a brief history of dyslexia, brain based differences between dyslexics and typical controls, studies examining cause vs. consequence of these brain based differences, and the framework for the studies presented in the following chapters.

**A Brief History of Dyslexia**
The term dyslexia was first coined by the German physician Rudolf Berlin in 1887 (Wagner, 1973), though another German physician, Oswald Berkhan, is believed to have first reported on individuals with dyslexia when he described students with a specific spelling disorder in 1885 (Warnke et al., 2012). In the United Kingdom, also at the end of the 19th century, W. Pringle Morgan documented a case of what he termed ‘congenital word blindness’ (Morgan, 1896). Here, he described a 14 year old who, “…has always been a bright and intelligent boy, quick at games, and in no way inferior to others of his age. His great difficulty has been—and is now—his inability to learn to read” (Morgan, 1896). Thirty years later, Samuel Orton of the United States also found himself evaluating children who other than struggling with reading seemed normal. Along with performing some of the early work in remediating children with reading disability, he also hypothesized that the struggle to attain reading was due to a lack of connection between the written word and the spoken word (Orton, 1925, 1937). These pioneers of dyslexia research worked a century ago, but the fundamental ideas are still at the core of the definition of dyslexia used today. The International Dyslexia Association (IDA) defines dyslexia as:

“…a specific learning disability that is neurobiological in origin. It is characterized by difficulties with accurate and/or fluent word recognition and by poor spelling and decoding abilities. These difficulties typically result from a deficit in the phonological component of language that is often unexpected in relation to other cognitive abilities and the provision of effective classroom instruction. Secondary consequences may include problems in reading comprehension and reduced reading experience that can impede growth of vocabulary and background knowledge.” (Lyon et al., 2003)

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b From Berlin’s monograph *Eine besondere Art der Wortblindheit (Dyslexie)*. (Wagner, 1973)
The core of this definition centers on the idea that the difficulty with reading stems from a
deficit in phonological skill (Lyon et al., 2003). In addition to being suggested by Samuel Orton
in the 1920’s, behavioral and neuropsychological research over the past several decades also
contributed to the phonological deficit hypothesis becoming the primary theory in dyslexia
research (For review see: Vellutino et al., 2004; Peterson and Pennington, 2012). This work in
part stems from studies in both typical and dyslexic readers in regards to the relationship
between phonological skills and reading in general and as a deficit in dyslexia specifically,
including the benefits of remediation aimed at phonological skill (Vellutino and Scanlon, 1987;
Wagner and Torgesen, 1987; Torgesen et al., 1994; Wagner et al., 1997).

Other major theories of dyslexia focus on additional behavioral abnormalities observed in
dyslexics, typically within the sensory-motor domain, including: the cerebellar deficit hypothesis
focused on deficits in automaticity (Nicolson et al., 2001), the magnocellular deficit hypothesis
which suggests magnocellular dysfunction across sensory, motor, and language pathways,
affecting processing speed (Stein and Walsh, 1997), and the double deficit hypothesis looking at
rapid naming speed and phonological skill as separate impairments in dyslexics (Wolf and
Bowers, 1999). Evidence has also been found concerning implicit learning problems (Vicari et
al., 2003; Howard et al., 2006; Menghini et al., 2006, 2008) and temporal processing deficits
within the auditory domain (Tallal, 1980; Temple et al., 2000). The evidence for many of these
theories has been discussed in various reviews (Ramus, 2003, 2004; Vellutino et al., 2004;
Peterson and Pennington, 2012), and while the general consensus is the phonological deficit
Brain Based Differences in Dyslexia

In addition to stating the primary and secondary deficits ascribed to those with developmental dyslexia, the IDA’s inclusion that dyslexia is neurobiological in origin is an essential aspect of the current theoretical framework. This comes from years of research into the differences between the brains of dyslexics and typically reading controls. Post-mortem work from Galaburda and colleagues found cortical ectopias and differences in planum temporale symmetry between dyslexics and controls (Galaburda and Kemper, 1979; Galaburda et al., 1985; Humphreys et al., 1990). These ectopias were found to represent a disruption of neuronal migration during cortical development, primarily within the perisylvian regions of the left hemisphere (Galaburda and Kemper, 1979; Galaburda et al., 1985; Humphreys et al., 1990). The fact that the ectopias represent disruptions in cortical development and later genetic work linking genes involved in this process to families with dyslexia further cemented the neurobiological basis of the disorder (Galaburda et al., 2006).

These gross anatomical findings led to a greater interest in determining the brain basis of dyslexia and with the advent of MRI, researchers gained access to a greater amount of data and were able to image the brain in vivo. Much of the early anatomical neuroimaging work built logically upon the post-mortem work from Galaburda. Studies employing manual tracing of MRI images focused on investigating the lack of leftward planum temporale asymmetry in
dyslexic subjects compared to typical controls (Hynd et al., 1990; Larsen et al., 1990; Leonard et al., 1993; Rumsey et al., 1997a; Dalby et al., 1998; Preis et al., 1998; Robichon et al., 2000), with inconsistent results.

As new techniques were developed, questions were asked about specific brain tissues. Voxel based morphometry (VBM) and diffusion tensor imaging (DTI) for example, have allowed researchers to interrogate gray and white matter volume or white matter integrity respectively. Whole brain VBM studies examining gray matter volume (GMV) have shown less GMV in dyslexics compared to controls in bilateral temporoparietal regions, bilateral cerebellum and left frontal cortex (Brown et al., 2001; Eckert et al., 2003; Brambati et al., 2004; Silani et al., 2005; Vinckenbosch et al., 2005; Kronbichler et al., 2008; Steinbrink et al., 2008). Recent meta-analyses have found consistent differences either confined to right superior temporal gyrus and left superior temporal sulcus (Richlan et al., 2012) or bilateral supramarginal gyrus, bilateral cerebellum, and left fusiform/inferior temporal gyrus (Linkersdörfer et al., 2012) using slightly different methodology. DTI studies have shown results of lesser fractional anisotropy (FA) in dyslexics compared to controls bilaterally across the brain, though primarily in temporoparietal and frontal areas (Klingberg et al., 2000; Richards et al., 2008; Steinbrink et al., 2008; Carter et al., 2009; Keller and Just, 2009; Rollins et al., 2009; Rimrodt et al., 2010; Hasan et al., 2012; Vandermosten et al., 2012a), with a recent meta-analysis localizing the most common finding to a left temporoparietal area containing portions of the left arcuate fasciculus and corona radiata (Vandermosten et al., 2012b).
Functional neuroimaging (fMRI and positron emission tomography; PET) studies of dyslexia indicate functional differences akin to those areas of anatomical differences between dyslexics and controls. Dyslexics have shown lower activation in left hemisphere posterior regions (temporoparietal and/or inferior temporal) during various reading and phonological skill tasks (Shaywitz et al., 1998, 2002; Brunswick et al., 1999; Paulesu et al., 2001; Hoeft et al., 2006, 2007). Studies have also found disruption of the connectivity of left angular gyrus during tasks of phonological assembly (Pugh et al., 2000b), left angular gyrus blood flow predicts severity of dyslexia (Rumsey et al., 1999), dyslexic children do not show specific activity for real words compared to false fonts within the visual word form system (Van der Mark et al., 2009), and activity during sentence comprehension tasks may also be disrupted in dyslexics (Rimrodt et al., 2009). Meta-analyses of single word reading in dyslexics generally confirm the consensus of these studies, finding decreased activity in left posterior regions (Maisog et al., 2008; Linkersdörfer et al., 2012); though, another meta-analysis found decreases in bilateral inferior parietal lobule and fusiform gyrus in children, and left middle/superior temporal gyrus, inferior temporal/fusiform gyrus and inferior frontal gyrus in adults (Richlan et al., 2011). These brain based differences (both anatomical and functional) primarily indicate differences in the left hemisphere language network (with some involvement of right hemisphere homologous regions) believed to be involved in reading (Pugh et al., 2000a, 2001), fitting well with the described behavioral deficits.

Remediation and Compensation
While questions of general anatomical and functional differences between dyslexics and controls have been studied in detail, fewer studies have attempted to study how intervention may change the brains of dyslexic subjects. Remediation studies examining the functional changes following reading interventions have shown increases in activation during phonological tasks in areas that were deficient before intervention in dyslexic children (Aylward et al., 2003; Temple et al., 2003; Shaywitz et al., 2004) and adults (Eden et al., 2004), and during a sentence comprehension task in children (Meyler et al., 2008). These regions include the left hemisphere reading network as well as some right hemisphere homologous regions. The only anatomical study examining remediation to date showed increased FA in the left anterior centrum semiovale following reading intervention (which showed a difference between dyslexics and controls prior to intervention), though this is not the most obvious white matter tract involved in reading, and not the location most commonly identified in DTI studies of dyslexia (Vandermosten et al., 2012b). To date, there have been no studies investigating GMV changes with a reading intervention. While these studies do show that disrupted functional and anatomical regions respond to phonologically based instruction, it is worth noting that the methodologies employed in these studies (specifically, the use of control groups and contrast thresholds) are variable and inconsistent.

In a related line of research, some studies have asked the question of what differentiates those individuals with dyslexia who are able to compensate and improve their reading ability compared to those who cannot. One study in adults looked at two groups of adult dyslexics, ‘compensated’ and ‘persistently poor’, compared to a group of adult typical readers (Shaywitz et
The two groups of dyslexics engaged different patterns of activation during both pseudoword and real word tasks. The ‘compensated’ readers engaged right middle temporal and superior frontal and anterior cingulate during the pseudoword task more than the ‘persistently poor’ readers. Conversely, during the real word task the ‘persistently poor’ readers showed greater activation of left occipitotemporal areas, although connectivity analyses showed this area was connected more with right frontal memory systems in the ‘persistently poor’ group as opposed to left frontal reading areas in the typical readers (Shaywitz et al., 2003). In a group of adolescent dyslexics, right inferior frontal gyrus activity during a reading task and right superior longitudinal fasciculus FA at one time point predicted the amount of gain in single word reading scores two and a half years later (Hoef et al., 2011). However, it is important to note that there was no intervention given in this study, as subjects were free to seek instruction or not during the two and a half year period.

Both the remediation studies and the studies looking at outcome are of value, but as of yet, no one has looked at intervention and outcome in the same group of participants. Therefore, it is unclear what the relationship is between brain areas that might predict outcome and those areas that are mobilized by a specific intervention. Understanding this relationship could have implications for intervention strategies as well as our understanding of the dyslexic brain.

**Cause vs. Consequence**

Since the findings of Galaburda and colleagues, the dyslexia field has moved forward under the impression that these anatomical differences are causal to the reading deficits observed in
dyslexia (Galaburda and Kemper, 1979; Galaburda et al., 1985; Humphreys et al., 1990). However, it is possible that some of the brain based differences observed in dyslexia are a consequence of the difference in reading experience. Some more recent work from two different research groups tries to address this question of causality of the functional and anatomical differences between dyslexics and controls. One group examines functional and GMV differences between pre-readers with a family history of dyslexia compared to pre-readers without a family history of dyslexia (Raschle et al., 2011, 2012). The GMV study found less GMV in several posterior regions implicated in the adult and pediatric dyslexia studies including bilateral temporoparietal cortex, left occipitotemporal and fusiform gyrus, and right lingual gyrus (Raschle et al., 2011). The functional study examined group differences in activity on a phonological processing task and found less activation in the group with a family history of dyslexia in bilateral occipitotemporal regions as well as temporoparietal and bilateral cerebellum (Raschle et al., 2012). The results of these studies suggest posterior reading network disruption is evident prior to learning to read, and argue in favor of the idea that these regions are causal to the behavioral deficits observed in dyslexia.

Another group utilized the concept of a reading matched design to address the issue of causality (Goswami and Bryant, 1989), comparing children with dyslexia with typically reading children (Hoeft et al., 2006, 2007). This design incorporates two control groups, one matched on age, and a second matched on reading level. If differences are seen between the dyslexic group and the traditional age matched control group, it is uncertain whether those differences are the cause of the deficits observed in dyslexia or a consequence of the reduced reading experience
(Goswami and Bryant, 1989). If the differences observed in the age-matched comparison are also found in the reading-matched comparison, those differences are more likely causal, as controlling for reading skill (in addition to the benefit of a higher age and probably cognitive skill in the dyslexic group) did not eliminate the difference (Goswami and Bryant, 1989). Hoeft and colleagues find less activation in left inferior parietal lobule and bilateral fusiform/lingual gyrus in their age-matched comparison, and replicate the results using a region of interest (ROI) analysis of these regions in their reading-matched group comparison (Hoeft et al., 2007). Conversely, hyperactivation in frontal areas for the dyslexic group in the age-matched comparison are not replicated in the reading-matched comparison, suggesting those differences are more likely due to reading ability. The same study also performs a GMV comparison using the functional map as ROIs, and find left inferior parietal lobule GMV is less in the dyslexics compared to the age-and reading-matched groups. Again, these results further suggest the importance of the posterior reading network to the deficits observed in dyslexia.

While these lines of research suggest a causal role of posterior brain abnormalities in dyslexia there are some important considerations which leave the question open. The studies in pre-readers work well in showing a disruption before learning to read, however, not all of the children in those studies will be dyslexic so they cannot directly address the question of causality (Raschle et al., 2011, 2012). The reading level matched comparisons also fall short due to the lack of whole brain comparisons of their reading-matched groups (Hoeft et al., 2006, 2007). In the case of the GMV portion of the study, the only comparison between the reading-matched groups is performed within the ROI selected from the functional results because the authors are
looking for structure-function relationships. Not performing whole brain analyses for the reading-matched groups, or at the very least interrogating an ROI based on the GMV results, leaves the question unanswered. Further complicating the picture, results from illiterate studies which suggest these same regions change functionally and/or structurally after learning to read, suggesting a possible consequential relationship between function/structure and reading ability (Carreiras et al., 2009; Dehaene et al., 2010).

**Dissertation Studies**

Studies examining the brain basis of dyslexia have been thorough in their attempt to explain the behavioral deficits in dyslexics compared to typical readers. However, several important questions remain and this dissertation will investigate some of these questions in an attempt to further the current knowledge of the field.

In the first study, the reading level match design is used to examine GMV and white matter volume (WMV) in dyslexic children and typical readers. The aim of this study is to examine the causal role of anatomical differences in dyslexia for the first time using a whole brain approach. The age-matched comparison should reveal results similar to those discussed previously in this introduction. The key comparison is between the dyslexic group and the younger, reading level-matched control group. Any regions that are consistent across the two comparisons would suggest a causal role in the behavioral deficits observed in dyslexia. Those regions that appear in the age-matched but not reading level-matched comparison may then reflect a consequence of the altered reading experience.
In the second study, we examined dyslexic children who underwent a reading intervention with behavior and neuroimaging at three time points: before the intervention, following the reading intervention, and following either an active control (math intervention) or developmental control (non-intervention null period). The aims of this study are to: 1) examine the functional changes following a reading intervention in dyslexic children and 2) examine where functional activity prior to the intervention predicts gain in reading skill following the intervention. This is the first study addressing functional changes and predictors of outcome in the same group of participants. It also is the first remediation study using a within subjects design for the control intervention period. This allows for the changes during the reading intervention to be attributed specifically to the reading intervention and not due to more general effects of learning or increased effort on the part of the participants.

The third study examines a subset of the participants from the second study who received high resolution anatomical scans at all three time points of the intervention. Here, the aim is to examine whether there are any GMV changes following the reading intervention. This is the first study in the dyslexia literature to examine GMV changes following an intervention. Also, it is the first training study to date showing GMV changes in children. Knowing where GMV changes occur allows for better interpretation of the functional changes reported here and those published previously. If the GMV changes are in the same areas as the functional changes, this raises the possibility that the functional changes are driven by GMV changes (the increase in blood oxygenation level dependent, BOLD, signal could be simply due to more GMV).
However, if they are not in the same regions, the functional changes could represent something else such as a change in connectivity.

Together, these studies address several holes in the existing dyslexia neuroimaging literature, specifically relating to current topics of interest including: the causal role of brain-based differences between dyslexics and typical readers, the effects of reading intervention on brain function and structure, and brain-based predictors of intervention outcome. Together, the work presented here examines the effect of reading experience on the functional and structural anatomy of the developing human brain.
CHAPTER I: ANATOMICAL COMPARISON OF GRAY AND WHITE MATTER VOLUME BETWEEN DYSLEXIC CHILDREN AND AGE- AND READING LEVEL-MATCHED CONTROLS

Introduction

Developmental dyslexia is characterized by difficulty with word decoding, word recognition, and spelling, which are believed to be related to problems with phonological coding (Peterson and Pennington, 2012). It is estimated to affect between 5% and 13% of school aged children (Katusic et al., 2001) and these deficits may also lead to secondary problems in reading comprehension and vocabulary (Lyon et al., 2003). With the importance of literacy for obtaining success in both the realm of education and the work force, continued progress in understanding the neurological basis of reading and reading disability remains critical for research, as well as for practitioners in regard to early identification and treatment.

Anatomical studies of dyslexia began with the *post mortem* studies of Galaburda and colleagues showing atypical symmetry of the *planum temporale* in both males and females with dyslexia (Galaburda et al., 1985; Humphreys et al., 1990) as compared to the leftward asymmetry noted in the majority of the typical population (Geschwind and Levitsky, 1968). Later the same investigators found cortical anomalies, referred to as ectopias, indicative of cortical development errors *in utero* which were observed primarily in left perisylvian cortex (Galaburda et al., 1985). These findings naturally led to theories that anatomical variations in this region were the cause of the reading deficits observed in dyslexia, and this idea was further substantiated when structural and functional brain imaging studies also revealed aberrations in
left perisylvian cortex in dyslexics compared to their controls. Specifically, the advent of magnetic resonance imaging (MRI) allowed for the use of larger sample sizes and in vivo measurement of brain anatomy. While MRI manual tracing studies of dyslexics show mixed results in regards to planum temporale symmetry (Dalby et al., 1998; Hynd et al., 1990; Larsen et al., 1990; Leonard et al., 1993; Preis et al., 1998; Robichon et al., 2000; Rumsey et al., 1997), they also revealed new observations, such as greater rightward asymmetry of Broca’s area in dyslexia (Robichon et al., 2000), decreased temporal lobe gray matter for dyslexics (Eliez et al., 2000), less total volume and gyrification (Casanova et al., 2004), differences in cerebellar symmetry (Leonard et al., 2001) and right cerebellar volume (Eckert et al., 2003) between dyslexics and controls. Inconsistencies of results across these studies have been attributed to sample size and subject characteristic differences (Eckert, 2004), and some of the asymmetry results may be explained by more general linguistic deficits and not dyslexia itself (Clark and Plante, 1998; Morgan and Hynd, 1998).

More recently, studies have focused on gray matter and white matter differences between dyslexics and controls specifically, as opposed to gross anatomical measurements. In these studies, the method of Voxel-Based Morphometry (VBM) has been used extensively and has revealed reduced gray matter volume (GMV) in dyslexia in bilateral superior temporal gyrus and inferior parietal lobule; differences have also been observed in bilateral cerebellum and frontal regions (Brambati et al., 2004; Brown et al., 2001; Eckert et al., 2005; Hoeft et al., 2007; Kronbichler et al., 2008; Silani et al., 2005; Steinbrink et al., 2008; Vinckenbosch et al., 2005). The consistency of temporoparietal findings has recently been emphasized by two meta-analyses.
of published reports of VBM studies, revealing less GMV in dyslexia in left superior temporal sulcus and right superior temporal gyrus (Linkersdörfer et al., 2012) and bilateral supramarginal gyri (Richlan et al., 2012). The latter meta-analysis also revealed less GMV in left occipito-temporal cortex and bilateral cerebellum. Interestingly, few studies have shown regions where dyslexics have more gray matter than control subjects (Silani et al., 2005; Vinckenbosch et al., 2005; Kronbichler et al., 2008) with no such reports in the meta-analyses (Linkersdörfer et al., 2012; Richlan et al., 2012).

While most reports interpret these findings as consistent with deficits in language, specifically phonological coding subserved by temporoparietal cortex, the issue of causality has presented a problem in the interpretation of these findings. As discussed by Linkerdörfer and colleagues, it is unknown whether these differences between dyslexics and typical readers are the neurobiological cause of the reading problem, or a “secondary, experience-dependent developmental change” (Linkersdörfer et al., 2012).

A causal theory might propose that the anatomical findings by Galaburda and colleagues, which have been attributed to neuronal migration errors prior to birth, are reflected also at the level of gross anatomical differences (i.e. GMV) and that these impede reading acquisition. As such, reports focus on less GMV in dyslexics compared to controls. However, another interpretation is that typical readers have more GMV than the dyslexics simply as a consequence of having learned to read to a more advanced level. Support for this explanation comes from studies on GMV change following the acquisition of reading and other training-induced skills.
Specifically, training studies have been performed in typical subjects learning how to juggle (Draganski et al., 2004; Boyke et al., 2008; Driemeyer et al., 2008), studying for a medical school exam (Draganski et al., 2006) and mirror reading (Ilg et al., 2008), and in patients receiving cognitive behavioral therapy (De Lange et al., 2008) and a reading intervention (Krafnick et al., 2011). All of these studies find increases in GMV with training. Importantly, learning to read changes the brain both functionally and structurally, as shown when ex-illiterates (those who learned to read as adults) are compared to illiterates (Carreiras et al., 2009; Dehaene et al., 2010). This suggests that learning to read mobilizes the GMV growth in areas that are engaged in reading. As pointed out by Carreiras and colleagues, these results speak strongly to the idea that the acquisition of literacy is driving GMV increases and that these fail to occur in dyslexia as a consequence of their reading disability rather than their cause (Carreiras et al., 2009).

Whether GMV differences are the cause of reading failure or the consequence can be tested by the use of a reading level-matched control group (Goswami and Bryant, 1989). The theory behind this approach is that differences between age-matched groups alone cannot answer the question of causality. However, interrogating a group of younger controls matched on reading level and looking for consistent differences across both types of group comparisons provides evidence for causation, as opposed to consequence. The reading level-match design has been critical to behavioral studies on the causality of phonological awareness deficits in dyslexia. Poor performance in phonological awareness in dyslexics relative to a younger, reading level-matched group, demonstrated that even when reading is equal (and the dyslexic group has the benefit of a
higher cognitive age), the dyslexics still show a deficit in phonological awareness, indicating that this weakness is the cause rather than the consequence of their reading problems (Goswami and Bryant, 1989). Using the same approach, one would predict that if the GMV differences were causal to dyslexia, they would also be seen in comparisons of dyslexics and typical readers matched on reading age, even though the dyslexics are more advanced in chronological age.

Hoeft et al. have addressed this important question by examining GMV in dyslexic children contrasted to a reading level-matched control group in addition to the typical chronological age-matched control group (Hoeft et al., 2007). However, in that study only those regions identified to differ between dyslexics and age-matched controls in brain function (during an fMRI study of word processing) were examined for GMV anatomical differences, while all of the GMV studies using VBM in dyslexia discussed above have demonstrated anatomical differences between groups matched on age using a whole brain analysis approach. As such, a reading level-match comparison needs to be carried out using the same methodological approach that has been applied for chronological age-matched comparisons reported in the literature, including similar sample sizes and statistical methods. The same applies to the study of white matter differences in dyslexia, as here too, the reading level-match design is essential to the question of causality.

Such an approach provides an important parallel with behavioral studies, which have recognized that the differences in dyslexia may not necessarily point to an etiology, but rather, poor reading and less opportunity for reading in and of itself can influence children’s cognitive abilities, and therefore present as part of the behavioral profile of reading disability (Goswami and Bryant, 1989).
Here, we directly addressed the causality of GMV differences in dyslexia by first contrasting a group of dyslexic children with a chronological age-matched control group using whole brain, voxel based methods that are consistent with previous reports of investigations into differences in GMV and WMV in dyslexia. We also compared the same group of dyslexics to a younger, reading level-matched control group again using a whole brain approach, followed by an ROI approach directly interrogating the regions identified in the age-matched analysis. We expected to replicate the previously reported differences in brain morphology observed between dyslexics and aged-matched controls in left temporoparietal regions and, importantly, replicate those results when comparing the dyslexics to a younger group of children matched on reading ability. Therefore, these areas would be confirmed as causal to dyslexia, rather than merely representing the status of brain anatomy that is consistent with children’s reading status.

Methods

Subjects and Subject Testing Procedures

Participants were 15 children with dyslexia, largely recruited from a private school that specializes in teaching students with dyslexia (n = 13) as well as a public school (n = 2) and 30 typically reading children, recruited from the general population to serve as controls. Inclusion criteria for the dyslexic children were: (1) having a documented history of dyslexia, as reported by their school; (2) single real word reading standard score of less than 92 (Woodcock-Johnson III Tests of Achievement Letter-Word Identification subtest; Woodcock et al., 2001); (3) Full Scale IQ greater than 80 (Wechsler Abbreviated Scale of Intelligence; Wechsler, 1999);
monolingual English speaker; and (5) no significant medical, neurological or psychiatric illness. For the control children, the inclusion criteria were identical except their single real word reading standard score was greater than 92. Children were all screened prior to entering the study to make sure they had no history of neurodevelopmental disability, congenital or acquired neurological disorder (such as a traumatic brain injury, disease affecting brain function, or known history of birth complications), or a diagnosis of specific language or hearing impairment. They also had no contraindication to MRI scanning such as metallic implants, plates or pins, or claustrophobia.

Half of the typically reading children (n=15) constituted the chronological age-matched control group (Control Group 1; n=15) by which to compare the dyslexic group. The dyslexic group and age-matched controls were also matched on performance IQ (see Table 1). The two groups were contrasted to one another to identify areas where dyslexics had relatively less or relatively more GMV and WMV than the controls. The other half of the typically reading children (n=15) formed the reading level-matched control group (Control Group 2; n=15). This group was therefore equivalent to the dyslexic group in their reading performance (matched using the single real word reading age equivalent), as well as performance IQ (see Table 1). This group was compared to the dyslexic group on GMV and WMV using the same procedures as those employed for the dyslexic versus control comparison using the age-matched design.
The study was approved by the Institutional Review Board of Georgetown University Medical Center. Parent consent and child assent were obtained. Subjects received book vouchers and choices of prizes for their participation.

**Behavioral Testing**

Psycho-educational tests were administered to evaluate single word reading accuracy and reading comprehension. All measures of reading described here provide age-referenced standardized scores with a mean of 100 and a standard deviation of 15. Standard score averages are reported for the chronological age-matched comparisons of dyslexics versus controls in Table 1. However, for the reading level-matched comparisons of dyslexics and controls, reading age level was used to ensure groups were matched for single real word reading. Average reading ages are reported in the portion of the table describing the dyslexic sample and their reading level-matched control group.

Single real word reading entailed untimed, out loud reading of single real words of increasing difficulty and was assessed using the Letter-Word Identification subtest from either the Woodcock-Johnson III Tests of Achievement (Woodcock et al., 2001) or the Woodcock Reading Mastery Tests-Revised (Woodcock, 1987). Single pseudoword reading was measured using the Word Attack subtest from the same testing instruments. This subtest requires subjects to read pronounceable non-words and provides a good measure of phonological decoding. Reading comprehension was assessed via the Passage Comprehension subtest (Woodcock-Johnson III Tests of Achievement; Woodcock et al., 2001) and required subjects to fill in the missing word
of a sentence. Together, these tests address untimed, out loud reading and decoding of single words and comprehension of silently read text.

Handedness was determined in all subjects by using the Edinburgh Handedness Inventory (Oldfield, 1971). Twenty-one subjects were determined to be right handed (laterality quotient > 33), three left handed (laterality quotient < -33: one age-matched control, one reading level-matched control, and one dyslexic) and six participants were not strongly lateralized (two age-matched controls, three reading level-matched controls, and one dyslexic).

Imaging Procedures

Anatomical MRI scans were acquired on a 3.0 Tesla Siemens (Erlangen, Germany) Trio whole-body MRI system. High resolution T1 weighted 3D MPRAGE images were acquired for each subject: repetition time (TR)/echo time (TE) 1600/4.38 ms, 256x256 field of view (FOV), 160mm slab thickness, 256x256x160 matrix (effective resolution is 1.0 mm$^3$), 1 excitation, and a 15° flip angle). All subjects participated in functional MRI studies as part of the protocol. For most subjects, three structural MRIs were acquired, with some subjects receiving fewer or greater, depending on time and subject compliance. In a population that is susceptible to head motion, multiple scan acquisition is one way to increase chances of obtaining an artifact free image given that a single, short head movement during the scan’s acquisition cannot be removed and can degrade image quality significantly. All images were inspected and rated by two research assistants blind to the subjects’ diagnostic group. For each subject, the scan with the least motion artifact was used for analysis. Retrospective analysis of the rater’s average scores
revealed that there were no significant differences in the mean rating score of the dyslexic versus control groups for both the age-matched as well as the reading level-matched comparisons.

Pre-Processing and Analysis

Structural MRI scans were pre-processed using voxel-based morphometry (VBM) in SPM 8 (Wellcome Department of Imaging Neuroscience, London, UK) with the modulation option selected such that resulting analyses represent tissue volume as opposed to density. Separate pre-processing was performed for the age- and reading level-matched groups. The following processing steps were completed: (1) Each subject’s image was manually aligned to the anterior commissure to decrease variability and co-registered to the SPM8 white matter template. (2) Images were segmented into gray matter, white matter and CSF using the New Segment toolbox (Ashburner and Friston, 2005). (3) DARTEL was used to register each structural image to a custom, study specific template derived from the subject’s images. (4) The template file generated by DARTEL was affine registered to closer align and spatially normalize the images to MNI space. (5) The resulting images were smoothed using a 8-mm full width at half maximum (FWHM) Gaussian kernel and an intensity threshold of 0.2 was used to remove voxels of low intensity from the analysis as well as to prevent possible edge effects.

Between group differences for gray and white matter volume (GMV and WMV) contrasting the dyslexics with both the age and reading level-matched groups were generated using two sample t-tests in SPM8. Height thresholds of p<0.01 uncorrected and extent thresholds of p<0.01 corrected were applied. Analyses were conducted using both the typical family wise-
error (FWE) correction as well as a non-stationary cluster correction (Hayasaka et al., 2004). Previous studies have utilized both of these types of cluster level corrections and we have included both here for greater ease of comparison with the existing literature. Peak coordinates as reported by SPM8 were converted from Montreal Neurological Institute (MNI) to Talairach space. Anatomical labels were assigned by using the anatomy toolbox included with SPM8 and verified by two independent investigators using the Talairach Atlas (Talairach and Tournoux, 1988). Total intra-cranial volume (TIV) was calculated by adding WMV, GMV and CSF (after intensity thresholding) for each subject. There were no differences in TIV between the dyslexic and control groups for either the age-matched comparison or the reading level-matched comparison, and specifically no differences were observed in total gray matter or total white matter between groups.

Extraction of GMV and WMV signal from identified clusters was performed using the MarsBaR toolbox (Brett et al., 2002). This process extracts the average signal from the voxels within the region identified for each subject. In order to determine whether the regions showing differences between the controls and the dyslexics matched for age also showed differences when comparing dyslexics and controls, this time matched on reading level, GMV and WMV signal contrasting reading level-matched dyslexics and controls was extracted from those clusters identified in the first (age-matched) analysis. Two sample t-tests were performed on the extracted signal for each ROI and p-values were Bonferroni corrected for multiple comparisons.

**Results**
Gray Matter Volume Whole Brain Comparisons

For the controls>dyslexics chronological age-matched comparison, five clusters were found (height threshold p<0.01 uncorrected, FWE cluster corrected p<0.01; Figure 1; Table 2). Two were located in the left hemisphere: temporal lobe (BA 21), located mostly in middle temporal gyrus, extending into superior temporal gyrus and secondly, the cingulate gyrus (BA 24). Three right hemisphere clusters were identified: precentral gyrus (BA 6), middle frontal gyrus (BA 10) extending into inferior frontal gyrus (BA 46), and thirdly, the superior temporal gyrus (BA 38). This contrast was repeated using a non-stationary cluster level correction as this has been suggested to better account for VBM data (see Methods). When this correction is applied, only the left middle temporal gyrus (BA 21) and right precentral gyrus (BA 6) clusters survived. The reverse contrast (dyslexics>controls) revealed no significant results using either the FWE or non-stationary cluster corrections.

For the controls>dyslexics reading level-matched comparison, one cluster was identified (height threshold p<0.01 uncorrected, FWE cluster corrected p<0.01 or non-stationary cluster correction; Figure 1; Table 2): right precentral gyrus (BA 6) extending into post-central gyrus. The location of this cluster is very close to one found in the age-match comparison above and is the only cluster that emerged from both the age- and reading level-match comparisons. When the non-stationary cluster correction was applied there were no statistically significant results. The reverse contrast (dyslexics>controls) revealed one cluster in left middle temporal gyrus (BA 21), but nothing survived the non-stationary cluster correction. Hence there were no regions in which dyslexics consistently showed more GMV.
White Matter Volume Whole Brain Comparisons

Parallel analyses were run on the white matter VBM data. For the controls>dyslexics age-matched comparisons, eight clusters were identified (height threshold $p<0.01$ uncorrected, FWE cluster corrected $p<0.01$; Figure 2; Table 3). The four left hemisphere clusters were all located in frontal cortex and included paracentral lobule extending into medial frontal gyrus, white matter underlying the middle frontal, precentral and the superior frontal gyri. Four right hemisphere clusters were identified including middle frontal gyrus, an area below the precentral gyrus, subgyral temporal (below middle/superior temporal cortex) and a region just anterior to the thalamus. When the non-stationary cluster correction was applied here, only the area around the right precentral gyrus remained significant.

The reverse contrast (dyslexics>controls) revealed no significant results for either the FWE or non-stationary cluster corrections.

For the reading level-matched comparison, the controls>dyslexics contrast revealed no significant results (height threshold $p<0.01$ uncorrected, FWE cluster corrected $p<0.01$). The reverse contrast (dyslexics>controls) revealed a single cluster just posterior to the right putamen. However, there were no significant results when applying the non-stationary cluster correction (Figure 2; Table 3). Together there were no regions where WMV differences were consistently observed between dyslexics and both of the control groups.
Gray Matter ROI Comparisons

While there was a region in the analyses of GMV differences that showed consistent differences between the dyslexic group and both non-dyslexic control groups (right precentral gyrus), we wanted to investigate if an ROI approach for the reading level-match comparisons would reveal additional support for the results from the age-matched comparisons obtained at the level of the whole brain. To do so, the regions shown to differ in GMV between the dyslexics and their age-matched controls (listed at the top of Table 2) were applied to the images from the comparison of the dyslexics with the reading level-matched controls and between-group statistics performed within this restricted area.

Of the five total clusters showing significant differences in GMV between dyslexics and controls from the age-matched group, only the right precentral gyrus showed a significant difference between the reading-matched groups (two sample t-tests, p<0.05 Bonferroni corrected for multiple comparisons). As noted above, the right precentral gyrus was significant for the between-group analyses using both types of control groups at the level of the whole brain and so this ROI approach did not yield any additional regions.

White Matter ROI Comparisons

Of the eight clusters showing significant differences in WMV between dyslexics and controls from the age-matched group, none showed significant differences between the reading-matched groups (p<0.05, Bonferroni corrected for multiple comparisons).
Using a non-Bonferroni corrected p value of <0.05 here did not result in any additional significant clusters for either the GMV or WMV comparisons.

**Discussion**

Here we addressed the question of causality of neuroanatomical difference reported in developmental dyslexia. We compared both GMV and WMV in dyslexics with a chronological age-matched control group, as well as a younger, reading level-matched control group. The goal was to establish whether previous findings of less GMV and WMV in dyslexia can be interpreted as the cause of reading impairments, as would be evidenced by observing GMV and WMV differences in dyslexia for comparisons with both typically reading control groups. Surprisingly, while we identified between-group difference in our dyslexics and their age-matched controls, consistent with prior publications on gray and white matter, the analogous analysis with reading level-matched controls failed to demonstrate the same between-group differences. As such, we conclude that differences in brain matter volume are not the cause of reading disability. Instead, growth in brain matter volume likely present a consequence of learning to read (Carreiras et al., 2009) and differences in dyslexia are due to a lack of reading experience.

The comparison between dyslexic children and chronological age-matched controls generally yielded results that are consistent with the existing literature using the voxel-based analysis approach. Relatively less GMV was observed in the left middle temporal gyrus extending into left superior temporal gyrus in the dyslexics, as has been previously reported in studies of children (Hoeft et al., 2007) and adults (Brown et al., 2001; Silani et al., 2005; Vinckenbosch et
al., 2005). The location of the maxima for the middle/superior temporal gyrus cluster is close to the region within the left superior temporal lobe captured by recent meta-analyses of GMV studies in dyslexia (Richlan et al., 2012). The proximity of this area to regions that are known to support phonological processing (Pugh et al., 2000a, 2001; Jobard et al., 2003; Frost et al., 2009) has always been considered logical, given that phonological processing is a principle dysfunction in individuals with dyslexia (for a recent review see Peterson and Pennington 2012). We also found relatively less GMV in the left cingulate gyrus as well as right hemisphere precentral, middle frontal and superior temporal gyri.

Turning to white matter, the comparison between the dyslexics and controls matched on age revealed several bilateral frontal regions as well as clusters located in the right temporal lobe and anterior to the thalamus where dyslexics had less WMV compared to controls. Previous VBM studies have also shown lesser left frontal WMV in dyslexics compared to controls (Eckert et al. 2005; Silani et al. 2005), suggesting the results of the current study are in line with the literature employing similar techniques as those used here (to our knowledge, no meta-analyses have been performed on white matter VBM studies in dyslexia). For both the investigation into gray matter and white matter we did not observe any regions where the dyslexics had more brain matter compared to the age-matched controls.

While the age-matched comparison results confirm previous findings on less gray and white matter in dyslexia, they do not speak to the direction of the relationship between reading and brain anatomy. That is, differences between the two groups could be due to dyslexia per se,
suggesting that anatomical differences are causal to impaired reading; or they could represent a consequence of the reduced reading experience that is concomitant with being a struggling reader. The inclusion of a reading level-matched control group provides a way to help interpret the directionality of the relationship and has been used in behavioral studies to establish causal factors of dyslexia (Goswami and Bryant, 1989).

Neither the whole brain nor ROI analyses of the reading level-matched comparison replicated the brain areas shown to be reduced in dyslexia via the age-matched comparison (except for right precentral gyrus GMV at a less stringent threshold). Most notably, the finding of reduced left middle/superior temporal GMV in dyslexics observed in the age-matched comparison and indicated in a recent meta-analytical study of GMV in dyslexia (Richlan et al., 2012) did not replicate in the reading-level match dyslexia versus control comparison. This indicates that the relative differences in GMV are not a neurobiological cause of dyslexia, but rather reflect an increase in GMV that typically occurs during the normal course of reading acquisition, and one that is hindered in the case of reading disability.

Our findings may seem surprising, but on further reflection are supported by other relevant work. While it is tempting to associate the anatomical anomalies observed with MRI to those microstructural changes measured at postmortem, and therefore assume that both are attributed to anomalies during prenatal cortical development (Galaburda et al., 1985, 2006), it is equally likely that the two observations are not directly linked. That is, while cortical ectopias may (or may not) reside in left perisylvian regions of the cortex of dyslexics, their presence may have no
relationship to the gross anatomical findings observed with MRI when dyslexics are compared to controls. Ectopias may interfere with reading acquisition in dyslexic children, but even here a causal connection has not firmly been established.

On the other hand, studies demonstrating GMV increases following experience-dependent behavioral improvement (Draganski et al. 2004; Boyke et al. 2008; Driemeyer et al. 2008; Draganski et al. 2006; Ilg et al. 2008) speak to the possibility that learning to read is accompanied by increases in GMV. Work in illiterate populations suggests that functional and anatomical changes are associated with reading experience (Castro-Caldas et al. 1998, 1999; Carreiras et al. 2009; Dehaene et al. 2010). Learning to read has been shown to change functional language networks including left hemisphere temporoparietal regions (Dehaene et al., 2010) and, especially relevant to the current study, an anatomical study found greater gray matter in temporoparietal regions (including posterior middle temporal gyrus) in late literates as compared to illiterates (Carreiras et al., 2009). With this in mind, along with the growing literature showing experience-dependent changes in GMV (discussed above) it is highly plausible that the reduced reading experience for children with dyslexia may be the explanation for less GMV in posterior left hemisphere language regions as opposed to this region being the cause of the reading deficits.

It is worth noting that in our comparisons between dyslexics and controls matched for age, we never found greater gray matter volume in the dyslexic group compared to the controls, which is consistent with most prior publications. If anatomical differences were causal to dyslexia rather
than a consequence, there is little reason to believe that they would only manifest as *smaller* volumes of gray and white matter. Interestingly, the majority of the work on training-induced changes in GMV have shown increases in gray matter with skill learning (Draganski et al., 2004, 2006; Boyke et al., 2008; Driemeyer et al., 2008; Ilg et al., 2008; Krafnick et al., 2011) with only one showing decreases (Draganski et al., 2006), again suggesting that it might be the reading experience that is responsible for growing GMV in typical readers to a greater degree than in dyslexics.

Our findings need to be reconciled with evidence that left temporoparietal cortex contains less GMV in pre-readers with family history of dyslexia compared to pre-readers without family history of dyslexia, suggesting they are not the consequence of impoverished reading (Raschle et al., 2011). Functional work from the same group also suggest deficits in this area are due to dyslexia as opposed to a consequence of struggling to read (Raschle et al., 2012). Early developmental studies are critical to solving the question of causality. It may be, however, that such studies need to focus on younger children than those studied to date, because it cannot be ruled out that it is their earlier oral language experience (relevant to phonological processing) that is responsible for these anatomical differences between groups at age 5. Longitudinal studies of dyslexics and controls, tracking changes in GMV well before and over the time that reading acquisition takes place will help to shed light on this matter.

In light of our inability to replicate the finding by Hoeft and colleagues, it is worth pointing out that these investigators took a fundamentally different approach to this question, by focusing
their analyses on regions of interest determined by functional MRI data acquired from their study of reading. The use of functional data to determine the ROIs may have biased the results in so far that areas that exhibit lower fMRI signal may represent areas that have less GMV and for that reason generate a lower BOLD signal. Our approach was different and more similar to previous studies investigating GMV in dyslexia. The first step was to replicate the GMV findings (and WMV findings) that have been reported in the literature for dyslexics and controls matched on age using the same analysis methods (i.e. whole brain, use of VBM, same sample size, etc.), and then to extend this same approach to the comparison of dyslexics and reading level-matched controls. Our statistical threshold is consistent with previous VBM studies in dyslexia: we used both FWE cluster level corrected statistics as well as non-stationary cluster corrected statistics on whole brain data. While the non-stationary correction has been considered by some to be more appropriate when using VBM (see Methods), both corrections have been used in the published studies on dyslexia. As such both are provided here for consistency and the choice of one over the other does not change the interpretation of our findings. Given the consistency of the methodological approach and the sample sizes of the present study with prior publications, along with our ability to replicate published findings in the age-matched comparison, failure to show differences between the dyslexics and controls matched for reading level cannot be attributed to a lack of statistical power.

The only region whose interpretation is somewhat dependent on statistical thresholding, is the right precentral gyrus. A difference between dyslexics and controls for GMV was revealed in the right precentral gyrus for the whole brain analysis of dyslexics versus age-matched and reading-
matched comparisons, but the latter did not survive non-stationary cluster correction. A difference here emerged from the ROI analysis for both comparisons. While we did not find any consistent differences in white matter between the dyslexics and the two relevant control groups, the WMV ROI analysis of right precentral gyrus showed a trend for significance at p<0.1 (uncorrected). Together, these results indicate that the difference in gray matter of the right precentral gyrus (and a hint of underlying white matter) may likely to be causal to dyslexia.

The right precentral gyrus is not a region we would have expected to identify as being critical in dyslexia, especially with so much interest dedicated to left hemisphere language areas. However, of the two GMV VBM studies of dyslexia in children (Eckert et al. 2005; Hoeft et al. 2007), the Hoeft study also showed less GMV bilaterally in large clusters that extend into precentral gyrus in the dyslexic group compared to controls, but this area was not interrogated in the reading level-matched group. While the Hoeft et al. study included girls and boys and the study by Eckert and colleagues only included males and it did not report differences in the right precentral gyrus (Eckert et al. 2005). Also of note, an adult VBM study of primarily females found right supplementary motor area (SMA; BA 6) to have less GMV in the dyslexic group compared to controls, although this was not a whole brain analysis (Menghini et al., 2008). This suggests perhaps that when females are included in the study, the right precentral gyrus plays a more prominent role in the list of anatomical variations caused by dyslexia. Interestingly, this right precentral finding suggests that the motor system or related functions (e.g. motor learning) may be involved in dyslexia. This theory is one of several that is being investigated and has some
support (Nicolson et al., 2001; Menghini et al., 2006, 2008); however see Ramus, (2003). Our results may be further evidence in support of such a theory.

Conclusion

The first part of this study confirmed the previously observed anatomical differences in gray matter and white matter volume in dyslexics compared to age-matched typical readers. Relatively less brain matter volumes were observed for the dyslexics in both hemispheres, including less GMV in left temporal cortex. When the question of cause versus consequence was tested by comparing the dyslexics with a younger reading level-matched group, we found that most of the brain regions identified in the age-matched comparison did not differ in the reading level-match comparison. Only the right precentral gyrus was reduced in gray matter in the dyslexic group relative to both control groups, but left hemisphere temporal regions implicated in reading and dyslexia did not differ in the reading-level match comparison. These results suggest that relatively less brain matter volumes reported in dyslexia are not causal to their reading difficulties. Consistent with earlier observations that learning to read increases GMV in adults, our results argue for anatomical differences in dyslexia being a consequence of their reading impediment.
Figures and Tables

Figure 1: GMV: Age-Matched and Reading Level-Matched Differences

Whole brain renderings for the GMV comparisons of both age and reading level matched groups at a height threshold of p<0.01 uncorrected and a cluster level threshold of p<0.01 FWE corrected. The left side of the figure shows the age matched group differences and the right side of the figure shows reading matched group differences. No significant results were found for the Dyslexics > Controls contrast for the age matched group comparison. See Table 2 for information on all clusters.
Whole brain renderings for the WMV comparisons of both age and reading level matched groups at a height threshold of p<0.01 uncorrected and a cluster level threshold of p<0.01 FWE corrected. The left side of the figure shows the age matched group differences and the right side of the figure shows reading matched group differences. No significant results were found for the Controls > Dyslexics contrast for the reading matched comparison or the Dyslexics > Controls contrast for the age matched group comparison. See Table 3 for information on all clusters.
Table 1: Participant Characteristics and Group Matching

<table>
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<th>CONTROL 2 (reading age-matched)</th>
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<tr>
<td>Performance IQ</td>
<td>Standard</td>
<td>101.3 (12.3)</td>
<td>102.3 (8.2)</td>
<td>109.2 (14.0)</td>
<td>ns</td>
</tr>
<tr>
<td>Score</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Word Identification</td>
<td>Standard</td>
<td>77.4 (7.6)</td>
<td>118.2 (8.3)</td>
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<td>0.0000</td>
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<tr>
<td>Reading Age</td>
<td>7.6 (0.9)</td>
<td></td>
<td>8.1 (0.5)</td>
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<tr>
<td>Passage Comprehension</td>
<td>Standard</td>
<td>78.3 (11.0)</td>
<td>110.3 (10.0)</td>
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<td>0.0000</td>
</tr>
<tr>
<td>Score</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Reading Age</td>
<td>7.3 (1.0)</td>
<td></td>
<td>7.6 (0.6)</td>
<td></td>
<td>ns</td>
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Table 2: Peak Coordinates and Cluster Details of Gray Matter Volume Differences

<table>
<thead>
<tr>
<th>Hemisphere</th>
<th>Cluster Coordinates (X, Y, Z)</th>
<th>Cluster Size</th>
<th>Z Score</th>
<th>Cluster P value</th>
<th>Peak Anatomical Location</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age-Matched Comparison</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controls&gt;Dyslexics</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>L</td>
<td>-45, -15, -12</td>
<td>1289</td>
<td>3.79</td>
<td>4.37x10⁻⁷</td>
<td>Middle Temporal Gyrus, BA 21</td>
</tr>
<tr>
<td>L</td>
<td>-16, -18, 40</td>
<td>1026</td>
<td>3.17</td>
<td>7.79x10⁻⁶</td>
<td>Cingulate Gyrus, BA 24</td>
</tr>
<tr>
<td>R</td>
<td><strong>42, -10, 33</strong></td>
<td>1545</td>
<td><strong>5.33</strong></td>
<td><strong>3.18x10⁻⁸</strong></td>
<td><strong>Precentral Gyrus, BA 6</strong></td>
</tr>
<tr>
<td>R</td>
<td>34, 38, 12</td>
<td>711</td>
<td>4.66</td>
<td>3.49x10⁻⁸</td>
<td>Middle Frontal Gyrus, BA 10</td>
</tr>
<tr>
<td>R</td>
<td>51, 9, -13</td>
<td>518</td>
<td>3.29</td>
<td>0.005</td>
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<td><strong>Dyslexics&gt;Controls</strong></td>
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</tr>
<tr>
<td><strong>No significant results</strong></td>
<td></td>
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</tbody>
</table>

| **Reading Level-Matched Comparison** |                                |              |         |                 |                          |
| Controls>Dyslexics |                                |              |         |                 |                          |
| R          | 42, -13, 32                   | 694          | 3.72    | 0.001           | Precentral Gyrus, BA 6   |
| **Dyslexics>Controls** |                                |              |         |                 |                          |
| L          | **-63, -53, 1**               | 672          | **5.04**| **0.001**       | **Middle Temporal Gyrus, BA 21** |

Entries in bold face indicate clusters that survive both FWE and non-stationary cluster corrections
Table 3: Peak Coordinates and Cluster Details of White Matter Volume Differences

Entries in bold face indicate clusters that survive both FWE and non-stationary cluster corrections

<table>
<thead>
<tr>
<th>Hemisphere</th>
<th>Talairach Coordinates (X, Y, Z)</th>
<th>Cluster Size</th>
<th>Z Score</th>
<th>Cluster P value</th>
<th>Peak Anatomical Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age-Matched Comparison</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controls&gt;Dyslexics</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>L</td>
<td>-4, -24, 52</td>
<td>784</td>
<td>4.53</td>
<td>4.78x10⁻⁹</td>
<td>Paracentral Lobule</td>
</tr>
<tr>
<td>L</td>
<td>-39, 43, 13</td>
<td>1051</td>
<td>4.05</td>
<td>9.94x10⁻⁹</td>
<td>Middle Frontal Gyrus</td>
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<tr>
<td>L</td>
<td>-30, -13, 45</td>
<td>918</td>
<td>3.49</td>
<td>6.53x10⁻⁷</td>
<td>Middle Frontal Gyrus</td>
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<tr>
<td>L</td>
<td>-20, 44, 32</td>
<td>356</td>
<td>3.54</td>
<td>0.007</td>
<td>Superior Frontal Gyrus</td>
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<tr>
<td>R</td>
<td>48, 26, 24</td>
<td>1063</td>
<td>4.47</td>
<td>8.42x10⁻⁵</td>
<td>Middle Frontal Gyrus</td>
</tr>
<tr>
<td>R</td>
<td>42, -8, 36</td>
<td>3465</td>
<td>4.30</td>
<td>3.54x10⁻¹⁹</td>
<td><strong>Precentral Gyrus</strong></td>
</tr>
<tr>
<td>R</td>
<td>40, -42, -5</td>
<td>878</td>
<td>3.52</td>
<td>1.17x10⁻⁶</td>
<td>Subgyral Temporal Lobe</td>
</tr>
<tr>
<td>R</td>
<td>10, -3, 17</td>
<td>1021</td>
<td>3.38</td>
<td>1.51x10⁻⁷</td>
<td>Anterior to Thalamus</td>
</tr>
</tbody>
</table>

**Dyslexics>Controls**

|                 |                                 |              |         |                |                               |
| Reading Level-Matched Comparison |                                 |              |         |                |                               |
| Controls>Dyslexics |                                 |              |         |                |                               |

|                 |                                 |              |         |                |                               |
| **Dyslexics>Controls** |                                 |              |         |                |                               |
| R                | 27, -4,-9                       | 386          | 3.80    | 0.004          | Lateral to Putamen            |

No significant results
CHAPTER II: USING FUNCTIONAL MRI TO CHARACTERIZE AND PREDICT RESPONSE TO INTERVENTION IN DYSLEXIA

Introduction

Developmental dyslexia is a common learning disability, affecting between 5% and 13% of the population (Katusic et al., 2001). It is defined by difficulties with word recognition and word decoding that are incongruent with other cognitive skills, classroom experience and motivation to learn how to read (Lyon et al., 2003; Peterson and Pennington, 2012). The word decoding problems (sounding out of novel words) are believed to be due to problems with understanding how sounds in spoken words are isolated (phonological coding or phonological awareness) and therefore impede mapping onto their corresponding graphemes (Scarborough and Brady, 2002). Word form recognition is also impaired, either independent of or as a consequence of poor phonological coding skills. To address both problems, explicit instructions in phonological coding as well as repetitive training in word recognition are often a key element in interventions designed to address dyslexia.

The neural correlates of word form recognition (orthographic processing) and phonological coding have been studied in skilled readers. These findings have been aligned with models of reading (Pugh et al., 2001), which for example, hold that there is a "direct" ventral route in which orthographic information is used to access semantic information via addressed phonology, and an "indirect" dorsal route involved in grapheme-phoneme conversion (assembled phonology) in order to access semantic information (Coltheart et al., 2001). Specifically, the ventral route maps onto the mid fusiform gyrus region of the left inferior occipitotemporal cortex (containing the so
called visual word form system) involved in recognition of whole real words (Cohen et al., 2002; Glezer et al., 2009). It has been suggested that visual recognition of words in this region is the consequence of recycling or reusing neurons traditionally involved in other types of object recognition (e.g. faces), under pressure exerted by learning to read (Dehaene and Cohen, 2007; Dehaene et al., 2010). This provides an important contrast to the regions in the dorsal route (left posterior superior temporal gyrus, inferior parietal cortex) that are aligned with spoken language, and as such have a longer evolutionary history (Dehaene and Cohen, 2007). It has been suggested that beginning readers use the dorsal stream to decode newly encountered words and that, through experience, words that are encountered more frequently become processed by the ventral stream in a more automated way (Pugh et al., 2000a, 2001).

Critical questions in neuroscience of reading include: which of these brain areas are preventing successful reading acquisition in dyslexia and importantly, which areas salvage the problem when the reading disability is overcome by intervention. A recent meta-analysis of children with dyslexia exemplifies the converging evidence of functional differences in children with dyslexia: underactivity in left inferior occipitotemporal regions and inferior parietal cortex consistent with the word recognition and word decoding problems observed in dyslexic children (Richlan et al., 2011). However, inferior parietal cortex underactivity is not reliably observed in adults with dyslexia (Richlan et al., 2011). This may be indicative of adjustments or compensatory mechanisms utilized by adult dyslexics, who in turn show underactivity in IFG, an observation that is not readily made in children (Richlan et al., 2011). The IFG has been shown not to become mature until adulthood in typical readers (Turkeltaub et al., 2003), suggesting that
dyslexics fail to make this final progression, and it may play a key role in bringing about normal reading due to its role in phonological coding and retrieval/articulation (Pugh et al., 2000a, 2001).

Interestingly, both adults and children with dyslexia show underactivity in left inferior occipitotemporal cortex (Richlan et al., 2011), suggesting that it is the most recalcitrant component of the neural basis of dyslexia. Further, adults show less gray matter volume in this same region (Linkersdörfer et al., 2012), suggesting that less experience-dependent use results in relatively less brain matter. Left occipitotemporal cortex might be the hardest to mobilize for reading, given that its functional history is not in reading, but object processing (Dehaene and Cohen, 2007; Dehaene et al., 2010).

fMRI has been used to specify differences in brain function in dyslexia (Shaywitz et al., 1998; Pugh et al., 2000b; Hoeft et al., 2007), and in detecting developmentally-induced (Turkeltaub et al., 2003) or intervention-induced (Aylward et al., 2003; Temple et al., 2003; Eden et al., 2004; Shaywitz et al., 2004; Meyler et al., 2008) changes in the neural basis of reading, and to predict reading outcome two years later (Hoeft et al., 2011). This raises the possibility that it could be used to identify specific versus more general training induced changes caused by reading training (as opposed to training on another cognitive skill) and to predict success of reading intervention. Prior studies on reading intervention in dyslexia (Aylward et al., 2003; Temple et al., 2003; Eden et al., 2004; Shaywitz et al., 2004; Meyler et al., 2008) have invoked theories of “normalization” and “compensation” in some of these regions, yet none of these theories have been adequately
tested because control interventions that test the specificity of any change were not employed. As such, which brain regions change specifically as a consequence of reading intervention (as opposed to general effects of learning or due to a placebo effect) and which areas are indicative of the successful intervention are unknown. Uncovering these areas is critical to the understanding of this common reading disability and for characterizing the neural course of successful intervention.

Here we employed functional MRI to systematically address the question of response to intervention and brain-based prognostic information in developmental dyslexia. We test the hypothesis that brain regions that signify the success of response to intervention are the same as those that change following the intervention and that these involve the dorsal stream language areas that subserve phonological coding and which have been in place for much of the evolutionary history of language. On the other hand, we also predict that left extrastriate inferior occipitotemporal cortex shown to be compromised in dyslexia is less likely to be malleable during intervention as its involvement requires being co-opted into word form recognition.

We used a task previously employed to ascertain the neural correlates of typical reading development (Turkeltaub et al., 2003), revealing underactivity in left inferior temporal cortex (Olulade et al., under review), consistent with findings from a recent meta-analysis (Richlan et al., 2011). For the current study, 31 children with dyslexia were scanned prior to and following their participation in a reading intervention. The intervention was delivered by tutors over an eight week period and targeted phonological and orthographic skills which we expected would
drive processes subserved by both the indirect and direct routes of reading. Studies of this nature are highly susceptible to the Hawthorne effect because children (some of whom often feel left out because of their academic struggles) change their behavior simply because they are the focus of a study. Likewise, the risk of a placebo effect is significant because the intervention is provided by motivated tutors who work with small groups of students using reward schema. To ensure that reading gains following the intervention could not be attributed to these types of effects, or be attributed to the normal developmental changes that occur during an eight week period, we subjected all children to an equally long control period during which they either received a math intervention from the same tutors (using similar instructional methods by which the reading intervention was delivered), or no intervention. This design provided an active (placebo) control period as well as a developmental control period by which to compare the reading intervention period and ensure that any gains, as well as their neural correlates, could be fully attributed to the reading intervention.

The reading intervention resulted in measurable gains in reading and in intervention-induced increases in activity in left inferior frontal gyrus. These differences in behavior and in brain function were specific to the reading intervention as demonstrated by a lack of such changes in the same children following the control periods.

Next, we examined what brain regions predicted the success of the reading intervention and discovered pre-intervention bilateral supramarginal gyri activity forecast reading gains produced by the reading intervention. These were specific to the reading intervention, as activity in the
supramarginal gyri did not predict gains in standardized measures of math performance following a math intervention (active control period). Our results show that activity in both supramarginal gyri serve as a specific marker of reading readiness, with greater activity in these regions during a word reading task being predictive of a greater leap in reading gain in response to reading intervention. At the same time, intervention-induced increases in activity occurred within the left frontal lobe, with intervention-induced decreases in multiple right hemisphere regions. Finally, left inferior occipitotemporal cortex which distinguishes dyslexic children from typically reading children is not involved in the remediation process, nor does brain activity in this area predict success of response-to-intervention.

Results

Behavioral Changes Following Reading Intervention

Thirty-one dyslexic children aged 7.4 to 12.6 years underwent an eight week reading intervention. The behavioral profile of all subjects prior to intervention is summarized in Table 1. Children were randomly assigned into one of three groups. All groups received the reading intervention and, during another time period, either an eight week math intervention or an eight week null period (to serve as an active or developmental control respectively). Nineteen subjects received math intervention as well as reading intervention (n=10 received reading intervention followed by math intervention and n=9 received math intervention followed by reading intervention) and twelve subjects received reading intervention followed by the developmental control. There were no significant differences in age and behavioral performance for the measures of IQ, reading and skills related to reading listed in Table 1 across these three different
groups at the outset of the study (one-way ANOVA with factor group), showing that the randomization had been effective (Appendix Table B.1).

One-way repeated measures ANOVAs were conducted on the standard scores of eight behavioral measures of reading and reading-related skills hypothesized to improve as a result of the intervention and obtained at three time points: at the outset of the study, following the reading intervention, and following the control period. Six of the eight measures showed a significant effect of Time Point (see Table 1 for ANOVA results): single real word reading (W-J WID), pseudoword reading (W-J Word Attack), passage comprehension (W-J PC), phonemic awareness (LAC-3), rapid automatized naming (RAN) of letters and numbers, symbol imagery (SI). Two of the measures did not reach significance: rapid automatized naming (RAN) of colors and objects and working memory (Digit Span). Post-hoc t-tests showed a significant increase in standard score following the reading intervention, but not following the control periods for those six measures (Figure 1). This allowed us to conclude that the intervention was successful in not only bringing about gains in the domains that were directly trained during the reading intervention (phonemic awareness and symbol imagery skills), but that these also generalized to measures of single real and pseudoword reading accuracy and even penetrated to gains made in reading comprehension. Importantly, these changes were not dependent on which intervention order the subject was placed in, as shown by repeated measures ANOVAs with between subjects factor group included; only automatized naming of letters and numbers showed a significant effect of group (Appendix Table B.1).
Limiting this analysis to only the children who participated in the math intervention control period offers a more tightly controlled comparison, as it assesses the children’s reading gains following intensive tutoring for skills that support reading directly to any gains made in reading when the content of the tutoring was focused on math instead of reading. This analysis (n=19) resulted in similar findings with the following measures demonstrating significant gains during the reading intervention whereas no gains were made during the math intervention: single real word reading (p=0.002), pseudoword reading (p=3.31x10^{-6}), passage comprehension (p=0.005), and symbol imagery (p=1.47x10^{-5}). Phonemic awareness (p=0.195), rapid naming of letters and numbers (p=0.577), rapid naming of colors and objects (p=0.424), and working memory (p=0.870) did not significantly change in this group. No gains were made in any of these behavioral measures during the math intervention period (though significant gains were made in math skills, see Appendix B and Appendix Figure B.1). These results confirm that the gains in reading and reading comprehension made by the children following the reading intervention are specific to the reading intervention and cannot be explained by more general accounts, such as the positive reinforcements provided by tutors or other placebo or Hawthorne effects.

Finally, not all reading interventions have long-term benefits. In this case however, the group maintained their gains, with no significant difference revealed by comparisons of standard scores of reading between their post-reading-intervention score and a one year follow-up score (12.4 months average) assessed in 26 of the original 31 participants (paired t-tests for standard scores revealed no significant changes (p>0.05) for any of the eight behavioral measures listed above.
Physiological Changes following Reading Intervention

During acquisition of fMRI data, subjects performed an implicit reading task that has been previously employed to study reading in children and adults (Price et al., 1996; Turkeltaub et al., 2003, 2004). Subjects viewed real words and false font stimuli and responded with a button press in one hand if the word or false font string contained an ascender (e.g. the l in the word alarm) and with a button press in the other hand if the word or false font string did not contain an ascender (e.g. grasp). Task performance did not differ across visits during the study (See Appendix B).

To establish which brain regions change in their level of activity during this task after the reading intervention we conducted a whole brain analysis on all of our dyslexic subjects (n=31; i.e. including the group receiving reading intervention followed by null period) using paired t-tests at a height threshold of p < 0.01 (uncorrected) and at least 50 contiguous voxels, consistent with the thresholds used in previous intervention studies of dyslexic children (Temple et al., 2003; Meyler et al., 2008). This analysis revealed increased activity in left inferior frontal gyrus (BA 45) and midline cerebellum (Talairach coordinates: 0, -38, -20). Decreases in activity following reading intervention were found in the left hemisphere in the anterior cingulate gyrus (BA 24), precentral gyrus (BA 4) and postcentral gyrus (BA 3), and in the right hemisphere in the inferior parietal lobule (BA 40), posterior cingulate gyrus (BA 23 and 29), precentral gyrus...
(BA 6) and precuneus (BA 7). To ensure the changes reported here were specific to the reading intervention we applied an exclusive mask of the changes that occurred during the control period (math and developmental null periods combined) at p<0.01 height and at least 50 contiguous voxels. This analysis allowed us to repeat the whole brain analysis while masking out any areas that changed during the control period of the intervention. This analysis produced nearly identical results, with the main difference being the increased activity in the midline cerebellum was no longer present. Additionally, the decreased activity observed in right inferior parietal lobule (BA 40) and posterior cingulate gyrus (BA 29) saw reductions in cluster size from 277 to 226 and 112 to 103 respectively due to slight overlap with changes during the control period. Table 2A and Figure 2A present the results of the analysis including the exclusive mask.

Next, for a more closely matched comparison, whole brain maps for the changes during reading intervention were generated for just the 19 subjects who received both reading and math interventions. It is important to note that the math intervention resulted in measurable gains in standardized math (but not reading) measures, thereby not only providing an active control for the same duration as the reading intervention, but also eliciting significant behavioral changes, making the comparison all the more meaningful. The comparisons again derived from the reading task, showed very similar results as for the entire sample (also at p < 0.01 uncorrected height threshold and at least 50 contiguous voxels). Three left frontal clusters in inferior frontal (BA 45/47) and middle frontal (BA 9) gyri were more active following the reading intervention. Decreases following the reading intervention were observed in one left hemisphere cluster in precentral gyrus (BA 4), and five right hemisphere clusters located in anterior cingulate gyrus
(BA 24), precentral gyrus (BA 6), parahippocampal gyrus (BA 28), medial frontal gyrus (BA 6) and caudate (extending into anterior cingulate BA 24). As with the analysis in the whole group we ran the whole brain analysis again, using an exclusive mask for the regions that changed during the math intervention. The results were again almost identical with the main difference being the decrease in right hippocampal gyrus no longer being present. Additionally, the cluster showing a decrease in right caudate/anterior cingulate gyrus split into two distinct clusters with peaks in the caudate and anterior cingulate gyrus (BA 24) respectively, and the right precentral gyrus cluster was reduced from 518 to 504 voxels. The increases in left frontal gyri were identical to the analysis without the mask. Table 2B and Figure 2B show the results from the analysis including the exclusive mask. Appendix Figure B.2 shows functional changes following the math intervention and Appendix Table B.2 provides cluster details.

**Brain Activity that Predicts Magnitude of Reading Gain following Intervention**

Whole brain simple regression analyses were run separately for standard score changes following reading intervention for W-J WID (single real word reading) and W-J WA (single pseudoword reading). At a height threshold of p<0.001 uncorrected with an extent threshold FWE corrected at p<0.05, we identified a relationship with single word reading in two clusters: left hemisphere supramarginal gyrus/inferior parietal lobule (BA 40), extending into angular gyrus (BA 39) and superior temporal gyrus (BA 22) (peak signal Talairach coordinates: x = -46, y = -45, z = 24), and right inferior parietal lobule (BA 40), including supramarginal gyrus (BA 39) (peak signal Talairach coordinates: x = 46, y = -28, z = 25) in (Figure 3, Table 3). The same analysis for pseudoword reading standard score did not yield significant results. Signal within
these clusters was not significantly correlated with change in calculation standard score following math intervention in the 19 subjects who received the math intervention, suggesting that activity for words false font here is specific to prediction of reading related skills. These results were not driven by areas showing a decreased response to false font vs. fixation (See Appendix B). Also, gray matter volume could explain differences observed in fMRI signal; however, a whole brain analysis of GMV predictive of change in reading standard score identified right precentral gyrus and not supramarginal/angular gyrus (see Appendix B).

**Discussion**

In this study we examined 31 dyslexic children who underwent a reading intervention aimed at mobilizing both the direct and indirect routes of reading. Gains were observed at the group level for direct measures of reading (single word reading, pseudoword reading, and passage comprehension) as well as skills related to successful reading (phonemic awareness, rapid naming, and symbol imagery). Accompanying these behavioral improvements, increased activity in left inferior and middle frontal gyri was observed following the reading intervention, along with decreased activity in several right hemisphere regions. Through the use of a within subject control design (either the active control math intervention or developmental control null period) we showed that these gains were specific to the reading intervention period and not due to developmental or general training effects. As hypothesized, the increased activity was observed in a left frontal region involved in phonological coding (Pugh et al., 2000a, 2001) as opposed to inferior occipitotemporal cortex which is likely less malleable to change over this period of time. Of all the regions that changed during the reading intervention, only left inferior
frontal gyrus showed a difference between dyslexics and a group of controls matched on age, performance IQ, and sex when we performed a region of interest analysis extracting signal from those clusters (See Appendix B). Thus, this increase likely represents a normalization of brain activity as a result of the instruction received during the reading intervention as opposed to a compensatory strategy.

Treatment approaches targeting phonological awareness have been shown to result in measurable reading gains in dyslexic students (for review see Alexander and Slinger-Constant, 2004). However, the response to treatment is variable amongst individuals, and while some studies have tried to measure behaviors that predict reading scores (Hatcher and Hulme, 1999; Torgesen et al., 2001), the magnitude of the reading gain following an intervention (Torgesen et al., 1999), or long term outcome in dyslexic adolescents (Hoeft et al., 2011), results have been weak and inconsistent. In the same vein, we did not find behavioral predictors of gains in the current study (See Appendix B). What we did find was that bilateral supramarginal/angular gyrus activity prior to the reading intervention was correlated with gains in single word reading standard score immediately following the intervention. Functional neuroimaging has been used in several domains to predict patient outcome in a variety of clinical conditions including: major depressive disorder (Canli et al., 2005; Siegle et al., 2006), anxiety disorders (McClure et al., 2007; Whalen et al., 2008), stroke recovery (Dong et al., 2006; Saur et al., 2010), temporal lobectomies (Janszky et al., 2005; Binder et al., 2008, 2010), and dyslexia outside the context of a specific intervention (Hoeft et al., 2011). While some might think that findings such as the ones described here should lead to diagnostic and predictive scans for children prior to a diagnosis, it
is not currently feasible. In addition to the expensive nature of neuroimaging and the likelihood that predictors would be specific to the intervention being employed, there would need to be large numbers of subjects used to create a model that was successful in predicting individual subjects’ likelihood of success. What this data does provide is insight into the role of inferior parietal cortex in developmental dyslexia.

The role of supramarginal/angular gyrus in predicting reading gains fits with previous neuroimaging literature of typical and dyslexic readers. Left temporoparietal cortex represents the indirect route in the dual route model for reading and is thought to subserve phoneme-grapheme mapping (Pugh et al., 2000a, 2001; Coltheart et al., 2001; Jobard et al., 2003). It has also been previously shown to correlate with phonological awareness skills in children for both print-speech and pseudoword-consonant string contrasts (Frost et al., 2009). Right temporoparietal cortex has been previously implicated in dyslexia in terms of being more active than in controls and/or correlated with reading measures in dyslexics more strongly than in controls (Shaywitz et al., 1998; Rumsey et al., 1999; Pugh et al., 2000b). Both the left and right temporoparietal results from this study map closely to those identified in the meta-analyses of control and dyslexic subjects. Maisog et al. (2008) found hypoactivation in dyslexics compared to controls located at -46, -44, 26 and 48, -34, -26 (x, y, z MNI coordinates) while our current findings were located at -46, -48, 24 and 46, -30, 26 (x, y, z MNI coordinates). It is hypothesized that left temporoparietal cortex becomes active during reading early in the learning process in typical readers and remains involved during reading through adulthood (Turkeltaub et al., 2003; Frost et al., 2009). The ability of this region to map the sounds of words on to the orthographic
representations of words may represent a brain that is ready to make gains in reading. Post-mortem work has found that several patients with dyslexia exhibit several neuronal ectopias and dysplasias (disordered neuronal architecture) in left perisylvian regions, but less typically in right perisylvian and left occipitotemporal cortex (Galaburda et al., 1985). These variations in neuronal architecture may underlie the dysfunction of temporoparietal cortex observed in dyslexia and help explain why treatment effects are not seen in the direct route of reading in this study.

Taken together, the results of the study show that evolutionarily older regions traditionally involved in spoken language (Dehaene and Cohen, 2007; Dehaene et al., 2010) (and represent the indirect route in reading, Pugh et al., 2000a, 2001; Coltheart et al., 2001) are crucial to the relationship between dyslexia and remediation, though not in the same way. The ability to engage bilateral inferior parietal cortex when reading seems to represent a readiness to benefit from the intervention used in this study, while inferior frontal regions show an increase in activity following the intervention much as this same region shows an increase over development (Turkeltaub et al., 2003). It is possible that the inability to properly engage areas involved in phonological processing prevents occipitotemporal areas from properly developing or showing changes following reading intervention.

Methods

Subjects
Thirty-one dyslexic children (14 female, 17 male) were recruited from a private school specializing in students with dyslexia and other reading disorders. School records were used to identify children with Woodcock-Johnson III Letter-Word Identification (W-J WID; Woodcock et al., 2001) scores 93 and below. The average age of the children was 9.6 years (range: 7.4-12.6 years). Wechsler Abbreviated Scale of Intelligence (WASI; Wechsler, 1999) scores were obtained prior to the intervention and in order to be included the children needed to score at least 80 on verbal (VIQ), performance (PIQ) and full scale (FSIQ) IQ. Average scores and standard deviations for these subjects’ reading skill and IQ prior to the intervention may be found in Table 1. Children were randomly assigned to one of three groups. One group (n = 10) received reading intervention followed by math intervention, a second group (n = 9) received math intervention followed by reading intervention and the third group (n =12) received reading intervention followed by a null period (no instruction). See Appendix Figure B.3 for a visual of the study design. The math intervention serves as an active control, while the null period serves as a developmental control. Groups did not differ prior to intervention on age, W-J WID, IQ, or any of the behavioral measures described below (See Results section). Although this group showed deficits in reading as measured by W-J WID, IQ scores range from low normal to above average. All children were in good health and reported free of other developmental disabilities, neurological disorders or any disease affecting brain function. Other exclusion criteria included diagnosed language or psychiatric disorders, hearing disorders, or contraindications to MRI scanning such as metallic implants or severe claustrophobia.

Behavioral Tests
All subjects received a battery of reading and reading related behavioral tests at all three stages of the intervention (prior to intervention, after reading intervention, after math intervention or null period). Researchers acquiring the behavioral data were blind to the child’s status of intervention at the time of data collection. The Woodcock-Johnson Word Identification (W-J WID; single real word reading; Woodcock et al., 2001), Woodcock-Johnson Word Attack (W-J WA; single pseudo-word reading; Woodcock et al., 2001) and Woodcock-Johnson Passage Comprehension (W-J PC; Woodcock et al., 2001) tests were used to directly measure reading ability. The additional tests: Lindamood Auditory Conceptualization Test (LAC-3; phonemic awareness; Lindamood and Lindamood, 1971), Rapid Automatized Naming (RAN L&N and C&O; naming fluency for letters/numbers and colors/objects; Denckla and Rudel, 1976a, 1976b), and Digit Span (working memory; Wechsler, 1999) were used because they measure skills supporting reading acquisition and/or are indicative of future reading ability. Symbol Imagery (Bell, 1997b) measures visual memory for letters and orthographic patterns and was included because of the strong use of mental imagery in the intervention strategy. For the math intervention: Woodcock-Johnson Calculation (Woodcock et al., 2001) measures mathematic computational ability, Woodcock-Johnson Math Fluency (Woodcock et al., 2001) is a timed test of mathematical computation ability, and Woodcock-Johnson Applied Problems (Woodcock et al., 2001) are mathematical word problems. All test scores reported for behavioral and regression analyses are standard scores (Mean = 100, SD = 15).

Reading Intervention
All 31 children underwent the reading intervention Seeing Stars (Bell, 1997a). This intervention uses a multisensory approach to help integrate internal visual and phonological representations of letters and letter strings. The imagery portion increases in difficulty starting with single letter imagery and increasing through two and three syllable words. A tactile/motor portion involves finger tracing of visualized letters and a language production portion involves verbalization of letter and syllable sounds out loud as they are finger traced in the air. The use of imagery/visualization in this reading intervention (Bell, 1997a) is based on several studies involving the use of imagery in reading, including: self-report of imagery during reading (Long et al., 1989), imagery in semantic retrieval (Kosslyn, 1976) and use of imagery directly related to improved processing and comprehension (Linden and Wittrock, 1981; Sadoski, 1983). The control math intervention used here, On Cloud Nine (Bell and Tuley, 1997), also utilizes a multisensory approach focusing on imagery, tracing and verbalization, similar to the reading intervention. The difference is the focus on numbers and number lines instead of letters and syllables, thus serving as a strong control for the reading intervention used. Both reading and math interventions (eight weeks each) were administered at the subjects’ school by employees of Lindamood-Bell Learning Processes, specifically trained to administer these programs.

Imaging Procedures and Analysis

Blocks of real words and false font stimuli were separated by blocks of fixation on a central cross. For activation conditions, subjects performed an implicit reading paradigm in which they responded with a button press in their right hand if the word or false font string contained a “tall” letter or character (e.g. ‘alarm’ contains ‘1’) and a button press in their left hand if the word or
false font string did not contain a “tall” letter or character (e.g. sauce has no tall letters). Each subject received two scans at each time point (before intervention, after reading intervention and after math intervention/null period). Blocks of real words and false fonts contained ten trials each, lasting 42s, and fixation blocks lasted 18s each between activation blocks with additional fixation at the beginning and end of the sequence resulting in a total scan time of four minutes, twenty-seven seconds. fMRI data was acquired using an echo planar imaging (EPI) sequence using a 3 Tesla Siemens Trio whole-body MRI system (TE = 30ms, TR = 3s, 64 x 64 matrix, 50 descending interleaved slices, 2.8mm slice thickness, 3mm x 3mm x 2.8 mm cubic voxels, flip angle 90°). Three high resolution, 3D T1-weighted MPRAGE images were obtained at T1 (prior to intervention) on the same Siemens Trio whole-body MRI system to aid in anatomical localization and for VBM analysis.

Pre-processing for functional analysis began by segmenting the subjects’ MPRAGE images and normalizing to a standard template brain (Montreal Neurological Institute, MNI). For all functional runs the first five scans were removed, and the remaining scans were corrected for head motion by realigning to the mean image, co-registered to the subjects’ MPRAGE, normalized using the same parameters for the MPRAGE image and finally smoothed using a 6mm x 6mm x 5.8 mm Gaussian kernel. For each subject’s 1st level analysis, both runs were included and contrasts were generated for real words vs. false font conditions, real word vs. fixation conditions and false font vs. fixation conditions. Motion parameters and global mean signal were included as regressors of no interest to account for subject movement and global signal change during each run.
Functional changes after intervention were examined using paired t-tests for word-false font and word-fixation contrasts. Each subjects’ session immediately preceding and following reading intervention was used (1st and 2nd visit or 2nd and 3rd visit depending on subjects group). A height threshold of $p < 0.01$ uncorrected with at least 50 contiguous voxels for the extent threshold was applied. To control for changes due to increased instruction, functional changes after math intervention were also examined at the same threshold. Masks for each contrast map were created (post-pre and pre-post for reading and math/null intervention periods). Using the combine ROI feature, masks were created and used during the analysis of changes during the reading intervention to eliminate any areas that also changed during the math intervention (or math intervention/null period) as described in the Results section of the text.

To investigate where functional response to real words-false font prior to the intervention is predictive of gain in reading standard scores following reading intervention, a regression analysis was run in SPM8 (Statistical Parametric Mapping, Wellcome Trust Centre for Neuroimaging, London) at an uncorrected height threshold of $p < 0.001$ and an extent threshold of $p < 0.05$ family wise error (FWE) corrected. From clusters identified in these analyses, percent signal change for real words and false font stimuli compared to fixation were extracted using the MarsBaR toolbox (Brett et al., 2002). This process extracts the mean percent signal change within the cluster for each subject. Pearson’s correlation coefficients for real words and false font percent signal change with change in reading standard score were generated in order to
determine how the regression for the words-false font contrast was driven. Significant correlations survived Bonferroni correction where indicated in Appendix B.

For the VBM analysis described in Appendix B, the high resolution MPRAGE scans obtained using the 3 Tesla Siemens Trio whole-body MRI system prior to the intervention were used (TE = 4.38ms, TR = 1600ms, TI = 640ms, FOV = 256mm, 160 slices, slice resolution 1mm, voxel size 1mm³). Each subject had three MPRAGE images acquired, and a blind image rating system using two raters picked the best MPRAGE for each subject in order to select the image with the lowest possible motion artifact, a problem that often occurs in this age group. VBM analysis was carried out in SPM8 using the DARTEL tools. Images were co-registered to the same space, segmented into gray matter, white matter and cerebrospinal fluid, group specific templates were created, images were normalized to MNI space and modulated in order to make volume inferences. These processed images were then thresholded at 0.2 such to remove voxels of low gray matter intensity from analysis. Whole brain regression analyses in SPM8 were run using the VBM gray matter images and change in standard scores after reading intervention for each subject (height threshold p < 0.001 uncorrected, extent threshold p < 0.05 non-stationary toolbox correction).
Significant changes were found for six of eight measures during the reading intervention, but not during the math intervention/null period: real single word reading, pseudoword reading, passage comprehension, phonemic awareness, symbol imagery, and rapid naming of letters and numbers. Standard score changes were not dependent upon intervention order. (A) Direct measures of reading skill: W-J WID (single word reading), W-J WA (pseudo-word reading), W-J PC
(passage comprehension. (B) Phonological processing: LAC-3 (phonemic awareness), symbol imagery (visual imagery). (C) Reading Related Skills: rapid naming of letters and numbers, rapid naming of colors and objects, digit span (working memory). P-values refer to difference in standard score before and after intervention period.
Figure 2: Brain Regions Showing Change Following Reading Intervention for Implicit Reading Task

Paired t-test whole brain maps of word > false font contrasts at p<0.01 and at least 50 contiguous voxels. An exclusive mask of changes during the math intervention/developmental null period was applied to ensure changes were specific to the reading intervention period. Increases are shown in red and decreases are shown in blue. (A) Changes following reading intervention for all
31 subjects in the study. (B) Changes following the reading intervention for the 19 subjects who underwent both reading and math interventions. See Table 2 for list of foci.
Figure 3: Functional Activity Predictive of Change in Single Word Standard Score Following Reading Intervention.
(A) Simple regression analysis of word\textgreater>false font activity prior to intervention vs. change in single word reading standard score (WJ-WID) following the reading intervention period. (B) Scatterplot of word\textgreater>false font activity vs. change in single word reading standard score from clusters in (A). Blue = Right hemisphere cluster, Red = Left hemisphere cluster.
Table 1: Behavioral Profile at Outset of Study and Repeated Measures ANOVA Examining Effects of Reading Intervention and Control Periods (n = 31)

<table>
<thead>
<tr>
<th></th>
<th>Mean (s.d.)</th>
<th>F-Statistic</th>
<th>P-value</th>
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<tr>
<td>Age (years)</td>
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<td>Wechsler Abbreviated Scale of Intelligence (WASI) *</td>
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</tr>
<tr>
<td>Verbal IQ</td>
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<tr>
<td>Performance IQ</td>
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</tr>
<tr>
<td>Full IQ</td>
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<td></td>
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</tr>
<tr>
<td>Measures of Reading</td>
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<tr>
<td>Single Word Reading</td>
<td>77.4 (8.0)</td>
<td>17.451</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Pseudoword Reading</td>
<td>91.8 (6.4)</td>
<td>10.816</td>
<td>&lt;0.001</td>
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<tr>
<td>Passage Comprehension</td>
<td>78.4 (13.9)</td>
<td>10.345</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Measures of Skills that support Reading</td>
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<tr>
<td>Phonemic Awareness</td>
<td>98.4 (8.4)</td>
<td>3.435</td>
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<td>Rapid Naming (Letters &amp; Numbers)</td>
<td>78.6 (12.4)</td>
<td>5.724</td>
<td>0.005</td>
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<tr>
<td>Rapid Naming (Colors &amp; Objects)</td>
<td>84.5 (12.4)</td>
<td>0.288</td>
<td>0.751</td>
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<tr>
<td>Digit Span</td>
<td>93.7 (11.8)</td>
<td>0.130</td>
<td>0.879</td>
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<tr>
<td>Symbol Imagery</td>
<td>80.2 (10.0)</td>
<td>32.219</td>
<td>&lt;0.001</td>
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* = WASI scores only available for 30 of 31 subjects
Table 2: Peak Coordinates for Functional Changes Following Reading Intervention

<table>
<thead>
<tr>
<th>Taliban Peak Coordinate</th>
<th>Cluster Size</th>
<th>T-statistic</th>
<th>Z-score</th>
<th>Anatomical Location</th>
</tr>
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<tbody>
<tr>
<td><strong>A: All Subjects</strong></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Word&gt;False Font Increase Following Reading Intervention</td>
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</tr>
<tr>
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<td>4.21</td>
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<td>x = -38, y = -11, z = 54</td>
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<td>3.51</td>
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<td>x = 26, y = -62, z = 40</td>
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<td>3.62</td>
<td>3.27</td>
<td>Precuneus BA 7</td>
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<td><strong>B: Subjects Receiving Both Interventions</strong></td>
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<tr>
<td>Word&gt;False Font Increase Following Reading Intervention</td>
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<tr>
<td>x = -50, y = 13, z = 18</td>
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<td>x = -46, y = 17, z = -1</td>
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<td>3.32</td>
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<td>x = 4, y = 11, z = 33</td>
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<td>x = -38, y = -11, z = 54</td>
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<td>70</td>
<td>3.37</td>
<td>2.93</td>
<td>Medial Frontal Gyrus BA 6</td>
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Table 3: Coordinates for Regression Analyses of Single Word Reading Standard Score Changes

<table>
<thead>
<tr>
<th>Talairach Peak Coordinate</th>
<th>Cluster Size</th>
<th>T-statistic</th>
<th>Z-score</th>
<th>Anatomical Location</th>
</tr>
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<tbody>
<tr>
<td>Change in Single Word Reading SS</td>
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<td></td>
</tr>
<tr>
<td>x = -46, y = -45, z = 24</td>
<td>205</td>
<td>5.76</td>
<td>4.66</td>
<td>Inferior Parietal Lobule/Supramarginal Gyrus, BA 40</td>
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<tr>
<td>x = 46, y = -28, z = 25</td>
<td>126</td>
<td>5.80</td>
<td>4.69</td>
<td>Inferior Parietal Lobule/Supramarginal Gyrus, BA 40</td>
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</table>
CHAPTER III: GRAY MATTER VOLUME CHANGES FOLLOWING READING INTERVENTION IN DYSLEXIC CHILDREN

Introduction

Developmental dyslexia is a neurobiologically-based learning disability in which individuals have difficulty with word decoding, word recognition and spelling and these in turn may negatively impact other reading abilities such as reading comprehension and vocabulary growth (Lyon et al., 2003). These deficits exist even though the individual has the intelligence, educational opportunity and motivation to learn to read (Lyon et al., 2003; Eckert, 2004; Vellutino et al., 2004). Dyslexia is more commonly observed in males than females and estimated to affect between 5.3% and 11.8% of school aged children (Katusic et al., 2001). Given this high incidence of dyslexia and the critical role of reading in the acquisition of knowledge and successful academic outcome, improving reading abilities in these children is an important priority for educators, policy makers and scientists. Over the past decade there has been increased interest amongst neuroscientists to quantify and characterize changes in brain structure, usually gray matter volume (GMV) following controlled learning experiences. These efforts, especially those focusing on the relationship between changes in brain structure and academic achievement in a formal learning environment (Draganski et al., 2006), have important implications for better understanding learning and skill acquisition in the classroom, especially in those children who encounter challenges in their efforts to acquire literacy. To date, no attempts have been made to measure changes in the brain’s gray matter in children with dyslexia following a formal, structured learning experience. Here we address this gap and make the

connection between behavioral intervention for reading disabilities and measures of brain morphometry, to inquire about the nature of GMV changes following intensive tutoring of children with dyslexia. The results, in conjunction with current understanding of brain-behavioral relationships, will help inform both educators and researchers in an effort to better understand the neural basis for successful reading intervention and potentially to develop programs to best help children who have trouble reading.

There exists now a significant corpus of work characterizing the neuroanatomical profile of dyslexia (for a review see Eckert, 2004). This research includes post mortem studies (Galaburda et al., 1985) and in vivo magnetic resonance imaging (MRI) research comparing dyslexic with non-dyslexic populations. The initial MRI research involved manual tracing of a variety of brain regions implicated in language and reading, however more recent research has quantified the neuroanatomical differences in dyslexic children and adults by using a technique known as voxel-based morphometry (VBM) (Ashburner and Friston, 2000). Using this automated method, a variety of brain structures have been shown to have smaller gray matter volume (GMV) in dyslexics as compared to controls. VBM studies comparing adult dyslexic to age matched control groups have shown less left temporal GMV (Brown et al., 2001; Vinckenbosch et al., 2005) and less bilateral temporal GMV for the dyslexic groups (Brambati et al., 2004; Steinbrink et al., 2008). Brambati et al. (2004) found less bilateral GMV for dyslexics in the cerebellar nuclei and Brown et al. (2001) also found less left inferior frontal and right cerebellar GMV in the dyslexics. The only two studies of children with dyslexia employing VBM have shown less GMV in bilateral inferior parietal lobule and temporal gyri and left inferior frontal gyrus (Hoeft
et al., 2007) and less bilateral lingual gyrus GMV compared to controls as well as left supramarginal gyrus and left posterior cerebellar lobe (Eckert et al., 2005). These regions are consistent with those implicated in studies using other structural analysis methods as described in Eckert (2004).

In parallel, functional brain imaging technologies (functional magnetic resonance imaging: fMRI; positron emission tomography: PET) have been used to investigate reading and language processing in the dyslexic brain. From these Pugh et al. (2001) has proposed a model describing the neural circuitry for reading in normal and disabled readers. The model proposes that three left hemisphere regions are relied upon for typical reading: an inferior frontal region involved in phonological output, a temporo-parietal region involved in rule-based orthographic to phonological processing as well as semantic analysis, and an occipitotemporal region involved in single word identification. These areas are commonly found to be less activated in individuals with dyslexia during paradigms of reading or reading-related skills. Specifically, temporoparietal and occipitotemporal regions consistently show hypoactivation for children and adults with dyslexia compared to normal readers in phonologically demanding (real and pseudoword reading) tasks; the inferior frontal cortex is sometimes hyperactive in dyslexics compared to controls on similar tasks (Shaywitz and Shaywitz, 2008). A recent activation likelihood estimate (ALE) meta-analysis of predominantly adult studies of functional brain imaging in dyslexics compared to controls found left hemisphere temporal and parietal areas were most likely to be less active in dyslexics than controls, although support for inferior frontal hyperactivation was not found (Maisog et al., 2008).
In a study of dyslexic children, different results were found for reading matched vs. age matched controls. When compared to both control groups the posterior network hypoactivation was found for dyslexics, however the hyperactivation in the frontal network was only found when compared to age matched controls, suggesting that the hypoactivation represents a functional deficit of dyslexia, while the hyperactivation is more representative of reading ability (Hoeft et al., 2007). Together these studies in children and adults point to a left hemisphere network that is impacted by an individual's reading disability. Notably these brain regions overlap with those that have demonstrated anatomical differences, as described above.

Most recently these functional brain imaging methodologies have been used to investigate whether the differences observed between dyslexic and normal readers change when the investigators intervene and improve reading ability in dyslexic individuals. Intervention studies in dyslexic children have shown changes in behavioral measures (i.e. increased performance in reading) and physiological changes measured using fMRI (Aylward et al., 2003; Temple et al., 2003; Shaywitz et al., 2004). While different types of interventions were given in these studies, similar patterns of increased activity were observed in bilateral frontal and temporoparietal regions. An intervention study in adult dyslexics showed increases in activation in bilateral temporal and parietal areas as well as the right inferior frontal gyrus (Eden et al., 2004).

While these studies speak to physiological changes in brain function following intensive training regimens focused on reading, it is not yet known if there are parallel changes in cortical
anatomy. Several longitudinal studies using VBM analysis have shown changes in subjects’ GMV after training. Draganski et al. (2004) followed a group of adults who were scanned before and after learning to juggle, and after not juggling for 3 months. An increase in GMV in area V5/MT (known to be integral to visual motion processing) was observed following the training, yet after the third scan, following a period of no training, there appeared to be a reversal of this pattern in the form of GMV decrease (although it was not significant over the time observed). Other longitudinal VBM studies have examined GMV change after a variety of tasks including more juggling tasks (Boyke et al., 2008; Driemeyer et al., 2008; Scholz et al., 2009), medical students studying for an exam (Draganski et al., 2006), mirror reading (Ilg et al., 2008), as well as repetitive transcranial magnetic stimulation (rTMS) on the left superior temporal gyrus (May et al., 2007), cognitive behavioral therapy (CBT) in a chronic fatigue syndrome population (De Lange et al., 2008) and pharmacological (quetiapine) treatment of a schizophrenic population (Stip et al., 2009).

Taken together, this literature has provides insight into the plasticity of the adult brain during learning. Increases in gray matter density seen early on (i.e. within one week after onset of training) (Driemeyer et al., 2008), suggest changes in spine/synapse density or cell body increases rather than neuronal or glial genesis. Longer term increases in hippocampal gray matter (Draganski et al., 2006) are more likely to reflect this slower process of neurogenesis. Anatomical changes after training have been observed in adults ranging from their early 20’s (Draganski et al., 2004) to early 60’s (Boyke et al., 2008), but has yet to be studied in a pediatric population.
To this point, changes in GMV after reading intervention have not been shown in children or adults with dyslexia. However, the above studies of training-induced changes in GMV and the fact that brain anatomy varies as a function of reading status (as shown for dyslexic versus non-dyslexic comparisons as well as in studies of illiterates; Castro-Caldas et al., 1998), suggest the possibility that such changes in the cortex might be measurable.

The current study was designed to investigate whether children with dyslexia who receive a reading intervention over an eight week period show changes in GMV. A longitudinal VBM analysis comparing GMV before the intervention, after the intervention and after an equal time period of non-intervention was performed to examine if any changes in gray matter could be observed as a result of the training. This three time point design follows the original Draganski et al. (2004) juggling studies. Based on the anatomical differences known to distinguish dyslexics from non-dyslexics (Eckert, 2004), the physiological changes previously reported following successful reading interventions (Aylward et al., 2003; Eden et al., 2004; Shaywitz et al., 2004) and the nature of the intervention used in the current study (visual imagery of words, multisensory integration and development of the sound representation of words) areas for which GMV changes were predicted included left hemisphere ventral visual, parietal and frontal cortices.

Methods

Subjects
The eleven dyslexic children (8 male, 3 female) whose data was submitted to this analysis were recruited as part of a larger study from a private school specializing in students with dyslexia. The school records were used to identify students with Woodcock-Johnson III Letter-Word Identification (W-J WID; Woodcock et al., 2001) scores less than 92. Average age of the eleven subjects was 9.1 years (Range 7 yrs 5 months-11 yrs 11 months). IQ scores were obtained prior to the intervention using the Wechsler Abbreviated Scale of Intelligence (WASI; Wechsler, 1999) which measures verbal (VIQ), performance (PIQ) and full scale (FSIQ) IQ. To be included in the study subjects had to score greater than 80 on these measures. Table 1 presents average scores and standard deviations for this group. Average IQ scores for this group all fell within the normal range (85-115), whilst reading of real words on the Letter-Word Identification fell well below the normal range. All subjects were free of any developmental disabilities, congenital or acquired neurological disorders and any injury or disease affecting brain function. Other exclusion criteria included diagnosed language or psychiatric disorders, hearing disorders, diagnosis of any major medical condition and any metallic implants, severe claustrophobia or any other contraindications to MRI scanning.

**Behavioral Tests**

A battery of behavioral tests were administered prior to and after the intervention as well as after the period of no intervention. Researchers acquiring the behavioral data were blind to the child's status of intervention. Woodcock-Johnson Word Identification (W-J WID; single real word reading; Woodcock et al., 2001), Woodcock-Johnson Word Attack (W-J WA ; single pseudo-word reading; Woodcock et al., 2001), and Woodcock-Johnson Passage Comprehension
(W-J PC; Woodcock et al., 2001) were used as direct measures of reading ability. The Lindamood Auditory Conceptualization Test (LAC-3; Lindamood and Lindamood, 1971), which measures phonemic awareness, Rapid Automatized Naming (RAN L&N; Denckla and Rudel, 1976a, 1976b), which measures naming fluency for letters and numbers, and Digit Span (Wechsler, 1999), which measures working memory, all measure skills that support reading acquisition and were therefore of interest to this study (Wagner and Torgesen, 1987). Symbol Imagery (Bell, 1997a) which measures visual memory for letters was used because of the strong imagery component of the reading intervention. All test scores are reported as standard scores (Mean = 100, SD = 15) except Symbol Imagery where the raw score is reported. The measures of single word reading, pseudoword reading and passage comprehension used multiple test forms (in an alternating A/B schedule). While the other tests did not include multiple forms, these are highly reliable tests and should reflect actual changes in score as opposed to test/retest effects. Figure 1 presents the behavioral data acquired over the three time points.

**Reading Intervention**

The reading intervention, Seeing Stars (Bell, 1997a), focuses heavily on imaging/visualization starting with single letters and increasing in difficulty to imaging one syllable and up to two and three syllable words. In addition to visualization, the intervention also has a tactile/motor aspect in which the students finger trace letters, as well as a language production aspect in which they say the letter or sound name out loud while they are tracing the letter in the air. Thus, the intervention utilizes multiple sensory modalities in order to help improve internal visual and phonological representations. The use of imagery as a focus of the
intervention is based on several studies relating the use of imagery in reading including a self-report study of imagery during reading (Long et al., 1989), use of imagery in semantic retrieval (Kosslyn, 1976), and more direct measures of relating imagery during reading to improved processing and comprehension (Linden and Wittrock, 1981; Sadoski, 1983). The intervention was administered at the subjects’ school by employees of the Lindamood-Bell Learning Processes who were specifically trained to administer the program. Subjects underwent eight weeks of this intervention followed by an eight week period of no intervention to serve as a control period. All behavioral testing prior to and following the intervention was conducted by research assistants who were members of the research team.

Imaging Procedures

Anatomical scans were obtained at the following three time points of the study: before the reading intervention (T1), after the reading intervention (T2) and after the period of no intervention (T3). At each of these three time points, three 3D T1-weighted MPRAGE images were obtained using a 3 Tesla Siemens Trio whole-body MRI system (TE= 4.38 ms, TR= 1600 ms, TI= 640 ms, FOV= 256mm, 160 slices, slice resolution 1mm, voxel size 1mm³). A blind image rating system using two raters was used to select the highest quality image from this set of three scans for each subject at each time point. This allowed for the selection of the image with the least amount of motion artifact, a problem that frequently occurs in this age group. An analysis of variance (ANOVA) was performed on the rating scores of the scans used for the three time points to ensure that there were no differences in rating scores across the three time points. As a precaution for avoiding head motion artifacts, children underwent training in a mock
scanner prior to the acquisition of MRI data to help acclimate them to the MRI environment (i.e. confined space and noise). Additionally, to ensure fluctuations in the quality of the image was not a contributing factor in the results of this study, noise outside of the brain was measured in two spherical ROIs and shown to be stable across the three time points.

Analysis

To evaluate the efficacy of the reading intervention on standardized measures of reading and skills known to support reading acquisition, a one-way repeated measures ANOVA was conducted on all of the behavioral measures for the three time points, followed by post-hoc t-tests.

For the analysis of the MRI data, those images selected for analysis (one per subject at each time point) were processed according to the optimized VBM protocol (VBM2 toolbox pipeline) described by Good et al. (2001) in SPM2 (Statistical Parametric Mapping, Wellcome Department of Cognitive Neurology, London). This analysis created a GMV template that is specific to this group using the first scan for each subject. Images were then segmented into gray matter, white matter and cerebral spinal fluid. The gray matter images were spatially normalized using the first time point as the source image for each subject. Although the VBM2 toolbox does not automatically modulate images, images were modulated here in order to make volume inferences from the results. The segmented/normalized/modulated images were smoothed using a default setting of 8mm FWHM. An absolute intensity threshold of 0.2 was used to remove voxels of low gray matter intensity from the analysis.
Statistical analysis for the VBM data was performed using the VBM2 toolbox. In order to determine clusters that significantly changed at any point during the study a one-way within-subjects ANOVA was performed at a height threshold of p<0.001 uncorrected, with an extent threshold of p<0.05. This height threshold has been used in previous VBM studies of dyslexia (Eckert et al., 2005) as well as other longitudinal VBM studies (Boyke et al., 2008; Driemeyer et al., 2008; Ilg et al., 2008). The cluster extent threshold here utilized the non-stationarity correction toolbox for SPM that allows for cluster level statistics on VBM data. Paired t-tests were then computed using the statistical analysis through the VBM2 toolbox to examine the direction of the effects (T2>T1, T3>T2 and T3>T1).

Average GMV signal (in arbitrary units) from these clusters was extracted using the MarsBaR toolbox (Brett et al., 2002). Average percent change in GMV for the clusters shown to increase significantly during the intervention was determined.

In addition to our main question about GMV changes brought about by reading intervention, we also wanted to explore whether the amount of reading improvement correlated with the amount of GMV change. To address this, a correlation matrix of GMV increases between T1 and T2 in regions identified by the above VBM analysis and behavioral test score changes between T1 and T2 was generated to obtain Pearson’s correlation coefficients. The behavioral tests included: single real word reading (W-J WID), pseudoword reading (W-J Word Attack), reading
comprehension (W-J PC), phonemic awareness (LAC-3), rapid naming of letters and numbers (RAN) and Symbol Imagery (SI).

Results

Behavioral Results

One-way repeated measures ANOVAs showed significant within-subjects effects over the three time points for all behavioral measures with the exception of working memory (Digit Span). Specifically, there were significant increases in the scores for single real word reading (W-J WID) $F(2,20)=10.77$, $p=0.001$; pseudoword reading (W-J Word Attack) $F(2,20)=6.321$, $p=0.007$; reading comprehension (W-J PC), $F(2,20)=5.420$, $p=0.013$; phonemic awareness (LAC-3) $F(2,20)=5.150$, $p=0.016$; rapid naming (RAN), $F(2,20)=7.655$, $p=0.003$; and Symbol Imagery (SI), $F(2,20)=30.723$, $p<0.001$. Working memory as measured by the Digit Span tests did not show significant changes, $F(2,20)=0.444$, $p=0.648$.

Post hoc t-tests were run on all behavioral measures (except Digit Span) to compare scores between T1 and T2, T1 and T3 as well as between T2 and T3. For the comparisons of scores between T1 and T2, single real word reading (W-J WID), phonemic awareness (LAC-3), and Symbol Imagery (SI) were each significant at $p<0.001$. Rapid Naming of letters and numbers (RAN) was significant at $p<0.01$. Pseudoword reading (W-J Word Attack) and reading comprehension (W-J PC) were both significant at $p<0.05$. Each of these measures was still significant when comparing T3 with T1 except for phonemic awareness (LAC-3). However, there were no significant changes in performance when comparing the scores between T2 and
T3. A graphic representation of these behavioral score changes over the three Time Points are shown in Figure 1.

**Anatomical Results: ANOVA**

The F test employed in the VBM2 toolbox identified seven regions with significant changes in GMV during the course of the study $F(2,20)$. In the left hemisphere, the anterior fusiform gyrus extending into the hippocampus (BA 20; $x=-36, y=-11, z=-24; F=55.58$), the superior frontal gyrus (BA 10; $x=-11, y=58, z=-12; F=22.92$) and the precuneus (BA 7; $x=-17, y=-60, z=31; F=18.32$) were identified. In the right hemisphere, the hippocampus ($x=32, y=-12, z=-16; F=20.54$), the anterior cerebellum ($x=8, y=-45, z=-10; F=16.18$), the precuneus (BA 7; $x=4, y=-60, z=30; F=15.95$) and the caudate ($x=9, y=16, z=9; F=14.96$) were significant. Details for these clusters can be found in Table 2.

**Anatomical Results: Paired t-tests**

Post hoc t-tests performed using the VBM2 toolbox showed that each of the clusters identified by the ANOVA represented a significant increase in GMV over the course of the study. Specifically, the clusters which increased significantly between T1 and T2 (during the reading intervention) were: the left anterior fusiform gyrus extending into the hippocampus (BA 20; $x=-36, y=-11, z=-24$), left precuneus (BA 7; $x=-17, y=-60, z=31$) right hippocampus ($x=31, y=-14, z=-15$), and right anterior cerebellum ($x=7, y=-46, z=-11$). Clusters from the ANOVA that demonstrated a significant increase between T1 and T3 were: the left anterior fusiform gyrus (BA 20; $x=-36, y=-11, z=-24$), right hippocampus ($x=32, y=-12, z=-16$), left precuneus (BA
7; x= -17, y= -60, z= 31), right caudate (x= 9, y= 16, z= 9) and right anterior cerebellum (x= 8, y= -45, z= -10). Details for these clusters can be found in Table 2. The only regions shown to increase significantly during the null period (between T2 and T3) were the left superior frontal gyrus and right precuneus.

**Anatomical Results: % GMV Change**

For those clusters that were identified as showing an increase in GMV during the intervention (between T1 and T2), the percent change in GMV signal across the three time points was determined using the GMV data extracted using the MarsBaR toolbox (Brett et al., 2002). For these four clusters the average percent GMV signal increases were: 3.40% in the left anterior fusiform, 3.15% in the right hippocampus, 3.51% in the left precuneus and 2.55% in the right cerebellum. Figure 2 pictures these four clusters and graphs the increase in GMV percentage over the three time points.

**Anatomical-Behavioral Correlations**

Correlation analysis between the change in behavioral scores and the GMV increase in the four clusters identified by the VBM analysis revealed two significant correlations. The amount of change in phonemic awareness (LAC-3) correlated positively with GMV change in the left precuneus (R=0.688, p<0.05) and the changes in pseudoword reading (Word Attack) correlated positively with GMV change in the right cerebellum (R= 0.748, p<0.01) (see Figure 3 for the scatter plots of these correlations). However, these correlations do not survive correction for multiple comparisons and were not significant comparing T1 to T3.
Discussion

This study followed eleven children with dyslexia who underwent reading intervention and examined if there were increases in GMV along with any intervention-induced gains in reading performance. From an educational standpoint, the intervention was successful, as it resulted in behavioral gains for measures of reading ability as well as for skills that are associated with good reading acquisition. These gains may be due to the multi-sensory approach used in the intervention (phonological training and visual imagery), but future studies will need to determine which components of programs such as the one employed here are critical in driving these increases in reading performance. Following the experimental design used in previous training-induced plasticity studies examining changes in GMV (Draganski et al., 2004, 2006) MRI scans were obtained before and after the intervention as well as after a null period where no intervention was given. We predicted increases in GMV in left hemisphere ventral visual, parietal and frontal cortices as these are brain areas that (1) subserve the skills targeted by the intervention program, (2) are known to be involved in the process of reading, (3) have been shown to be under-activated in dyslexic readers, and (4) have been shown to increase in activity following a successful reading intervention. The students showed significant gains in reading (as well as reading-related measures) following the intervention and GMV increases specific to the intervention period (T2 compared to T1) were observed in four areas: left anterior fusiform (extending into the hippocampus), left precuneus, right cerebellum and right hippocampus. Importantly, all of the behavioral gains (except for phonemic awareness) and the changes in these four regions were still significant when comparing T3 with T1, but were not significant
when comparing T3 with T2, demonstrating that both behavioral and GMV changes observed on
the second scan were associated with the period of intervention. The maintenance of these gains
through the null period is encouraging in terms of the in classroom benefit for these children.
The left fusiform and precuneus findings support our original predictions based on the nature of
the intervention and what is known about the neural signature for reading and reading disability,
while the increases in GMV in bilateral hippocampus and the right cerebellum may suggest that
a more general learning network was engaged during the intervention period. Importantly, the
GMV increases reported here provide evidence that the learning associated with the intervention
has structural brain correlates. Understanding how specific changes in behavior relate to specific
structural changes in the brain after intensive intervention may be useful in understanding which
regions of the brain are targeted by specific interventions and, if the focus of the intervention is
further tailored with this knowledge, it might eventually provide a better understanding of how
children successfully learn in the general classroom and in special education settings.

**Increased GMV in Left Anterior Fusiform/Hippocampus and Right Hippocampus**

The peak of the cluster in the left anterior fusiform gyrus falls within the predicted changes
for the ventral visual pathway. However, a significant portion of this cluster extended into the
left hippocampus. This along with the cluster identified in the right hippocampus suggest a
bilateral increase in hippocampal GMV. These changes may reflect general learning that is
occurring during the intervention period as has been demonstrated in the right hippocampus for
students preparing for a medical exam (Draganski et al., 2006). Each of these regions will be
discussed in turn.
The cluster with the peak in the left anterior fusiform is part of the ventral visual stream. However, it is more anterior than regions known to be involved in the processing of single words (the so called “visual word form area”; Cohen et al., 2002) and closer to regions that have been implicated in object naming/processing rather than word processing. Renvall et al. (2003) found this region to be more active for naming objects in a complex scene compared to naming colored circles. In a study looking at both naming and viewing of words and objects, multiple regions of the anterior fusiform were identified (Moore and Price, 1999). One peak in BA 20/37 was found to be more active for words and objects (over meaningless stimuli) irrespective of the task, and another peak (slightly anterior and superior in BA 20) was found to be more active specifically during object naming but not word naming (Moore and Price, 1999). The students in the current study were required to make connections between a letter or groups of letters and the sound they make. A possible interpretation is that students with dyslexia are relying on anterior located regions traditionally associated with object processing in order to compensate for regions in the posterior ventral stream that are not supporting word processing in ways that is typical for normal readers. Additionally, Anderson et al. (2000) showed a region in the anterior fusiform that was more active during encoding than retrieval during a word pair association task. Hence another possibility is that the intervention placed increased demand on this region in encoding the connections between letters/groups of letters and sounds, resulting in an increase in GMV. Future studies using functional imaging may be able to address both of these possibilities more directly.
Turning to the hippocampus, it is notable that a GMV study of learning in medical students found hippocampal gray matter to increase over all time points of the study (Draganski et al., 2006). The authors suggest that this could be due to the fact that neurogenesis occurs in the hippocampus, while it does not in other areas where GMV increases have been observed. However, in the current study both the left and right hippocampi showed a significant increase only during the intervention period while increases during the null period did not meet statistical significance. Hence we cannot reinforce the interpretation offered by Draganski and colleagues, although we do not rule out the possibility that future studies could show a more robust increase during the control period.

Increase in GMV in the Left Precuneus

An increase in GMV in the left precuneus is consistent with predictions made for the study, based on the fact that the intervention has a strong emphasis on visual imagery. The precuneus has been implicated in various functions including visuo-spatial imagery and memory retrieval (for a review see Cavanna and Trimble, 2006). The left precuneus has been implicated in visual imagery of letters, specifically the visuo-spatial aspects of the imagery (Raij, 1999). Thompson et al. (2009) found bilateral precuneus activation during a spatial location task compared to a spatial-transformation task. In their study subjects viewed an arrangement of letter stimuli and later were primed with a letter and trisected circle; subjects had to decide which third of the trisected circle would be facing the middle of the screen if it was in the position of the primed letter (Thompson et al., 2009). In addition to these studies in typical populations linking letter imagery to the left precuneus, there is also evidence of differences the precuneus in dyslexic
readers. A meta-analysis by Maisog et al. (2008) found the left precuneus to be less likely to be active in dyslexic adults compared to controls. In another study, the right precuneus was found to have less GMV in dyslexic adults compared to controls (Menghini et al., 2008).

Further, the amount of GMV increase in the left precuneus in our study showed a positive correlation with score change for the phonemic awareness test (LAC-3). As this is one of the skills targeted by the intervention (in addition to visual imagery), it is encouraging to find a direct relationship between the amount of GMV increase and the amount of improvement in this skill. However, a correlation between visual imagery, another integral part of the intervention, and precuneus GMV increases was not found and as mentioned in the results section, the correlations reported for this study did not survive a correction for multiple comparisons, suggesting these results are somewhat tentative.

Increase in GMV in the Right Cerebellum

The right anterior cerebellum also contained a cluster of increased GMV after the reading intervention. While we did not predict a change here, the findings are notable given the theoretical model linking the cerebellum and dyslexia (Stein and Walsh, 1997; Nicolson et al., 2001; Fawcett and Nicolson, 2007; Laycock et al., 2008), specifically in ways that account for the sensorimotor problems that have been reported in individuals with reading disability. For example, using PET, Nicolson et al. (1999) showed that dyslexic adults had lower right cerebellum activation compared to controls while learning a motor sequence and also when later performing that learned motor sequence. The cerebellum has also been included in a number of
reports investigating GMV differences in dyslexic subjects using VBM (Brown et al., 2001; Brambati et al., 2004; Eckert et al., 2005) and other methods used to evaluate anatomical aberrations (reviewed by Eckert, 2004). For example, the right anterior lobe of the cerebellum has been shown to have less overall volume in dyslexic children compared to controls (Eckert et al., 2003). Further, a magnetic resonance spectroscopy (MRS) study showed male dyslexic adults to have biochemical asymmetry in the cerebellum that was suggestive of differences in the cellular density of dyslexics compared to controls (Rae et al., 1998). In a later study by these investigators using anatomical measures, dyslexic adults were found to have abnormally symmetric cerebellar gray matter compared to controls; controls had less left hemisphere gray matter than the dyslexic group (Rae et al., 2002).

It is notable that the amount of increase in GMV observed in the right anterior cerebellum following the intervention showed a positive correlation with the change in score on the pseudoword reading (Word Attack) measure. There is previous evidence for a relationship between phonological decoding skills (e.g. pseudoword reading) and the anterior cerebellum. Subjects characterized as phonological dyslexics (pseudoword decoding scores <90) were shown to have a leftward asymmetry in the anterior cerebellum (Leonard et al., 2001). It is possible that an increase in the right anterior cerebellum reflects a shift to a less asymmetrical anterior cerebellum. However, the subjects in our study were not as weak in their pseudoword reading abilities as the subjects reported by Leonard et al. (2001) (our subjects’ weakness was most prominent for real word reading) and asymmetry of the anterior cerebellum prior to the intervention was not investigated. Also, as previously mentioned, the evidence for a relationship
between GMV increases in the right cerebellum and pseudoword reading advancement is tentative (the correlation did not survive a correction for multiple comparisons).

**Learning and Structural Plasticity**

While our study design followed that employed by Draganski et al. (2004) who measured GMV prior to and following a training period (during which subjects learned to juggle) and again following by a period where no practice occurred, there is an important difference in our study, in that reading interventions should provide a lasting improvement (and it did) and subjects do not cease to read. In other words, the skill learned by the participants in the juggling studies (Draganski et al., 2004; Boyke et al., 2008; Driemeyer et al., 2008) was entirely novel to the subjects and the importance of maintaining long-term improvements was not of the same value as reading gains are to a dyslexic student. However, in this regard our study bears some resemblance to another study by Draganski and colleagues (Draganski et al., 2006) in which they followed medical students while they studied for an exam before the semester break. The type of learning the medical students did for their exam and the learning the children did in this study represent skills that are useful and more likely to be used regularly than those used in the juggling studies.

The pattern of GMV change (except for the hippocampus) for both the jugglers and the medical students showed an initial increase during the learning phase followed by a small but non-significant decrease during the null period (Draganski et al., 2004, 2006; Boyke et al., 2008; Driemeyer et al., 2008). This trend suggests that practice may be necessary to maintain the
structural changes achieved while learning. The GMV change in the current study shows significant increases during the intervention and non-significant increases in the period after the intervention. This pattern is consistent with the behavioral data, where the scores showed significant improvement during the intervention, followed by non-significant changes in the eight weeks afterward. This is an important finding for educational purposes as it suggests these children are maintaining their behavioral gains without the intervention, but it raises an interesting question as to what cortical mechanisms support these sustained gains. May and Gaser (2006) offer a thorough review of the morphometry and plasticity literature including the possible neuronal correlates of the GMV changes. Importantly, while it is possible to speculate on the nature of these GMV changes after various interventions, it is not possible to determine from the current study or the previous longitudinal VBM literature whether these changes are due to learning or practice effects. Additional experimental groups including those varying in length of interventions and control groups including but not limited to those matched for cognitive effort and baseline behavior would be necessary in order to make more definitive conclusions.

Another important distinction from previous longitudinal VBM studies (Draganski et al., 2004, 2006; May et al., 2007; Boyke et al., 2008; de Lange et al., 2008; Driemeyer et al., 2008; Ilg et al., 2008; Scholz et al., 2009) is that the subjects here are children. The participants in the previous studies range from young adults through elderly subjects, but no subjects below the age of 20 have been studied as of yet. One might predict that changes in GMV in pediatric populations may be especially pronounced, since GMV is already undergoing dramatic changes
as part of typical development (Sowell et al., 1999a, 1999b). Even though GMV has not yet been investigated in the context of intervention in children, white matter integrity has been studied. Keller and Just (2009) showed that increases in phonological decoding ability correlated with increased fractional anisotropy (FA) in the left anterior centrum semiovale. While these changes do not correspond with the GMV changes reported in this study, this is not unexpected as both the tissue type analyzed and interventions used are different. Specifically, the duration, intensity and approach of the intervention may modulate which brain regions are impacted. Future studies examining a variety of anatomical measures and addressing different types of interventions will be able to assess the more integral relationships between anatomical changes and reading intervention. Further, because gray matter undergoes significant changes during development from childhood through adulthood (Sowell et al., 1999a, 1999b) these studies will also need to include a wider age range.

Limitations

There are a few important considerations to take into account while interpreting the results of this study. The group was made up of eleven dyslexic children, and while this is similar to group sizes used in previous studies examining GMV changes following training (Draganski et al., 2004) and studies comparing dyslexic subjects to controls (Brambati et al., 2004; Vinckenbosch et al., 2005; Steinbrink et al., 2008), it should be noted that the sample size is small. It is also important to appreciate that we did not have a dyslexic control group that did not receive the intervention to compare with the dyslexic sample receiving the intervention. Instead, the null period following the reading intervention was used as a within subject developmental control
period, which is typical in studies in the field of education, where it is difficult to withhold intervention from students who have significantly fallen behind on their academic skills. Further research into the nature of these changes and their relation to reading skills will help translate what is learned in the research environment to helping children directly in the classroom.

Conclusions

This study showed gains in reading skills and increased GMV in dyslexic children after an eight week reading intervention. GMV increases were observed in the left hemisphere in anterior fusiform/hippocampus and precuneus. The left anterior fusiform region is commonly engaged in tasks involving object processing and object naming and may suggest that the dyslexic students are relying on this region to help improve their processing of words. The left precuneus has been implicated in visual imagery and specifically in tasks involving imagery of individual letters. Right hemisphere GMV changes following the intervention were found in the cerebellum and hippocampus. There is a theoretical framework implicating the cerebellum in dyslexia and this study adds a novel contribution to this theory. Finally, the GMV increases in the left hippocampus (extending from the cluster reported for the anterior fusiform gyrus) and right hippocampus may reflect more general learning that is occurring during the intervention. The increases in GMV were restricted to the intervention period and were not observed after the intervention ended, suggesting that these increases in GMV are related to the intervention. This is the first longitudinal VBM analysis in children and demonstrates that changes in brain structure are brought about by intervention. These findings provide encouragement that learning can result in both lasting behavioral and structural changes in children who struggle in learning
to read. Further investigation will improve understanding not only for how the brain responds to learning, but in how these findings may be translated into refining interventions and improve the learning experience.
Figures and Tables

Figure 1: Behavioral Score Changes Over the Course of Reading Intervention

Test scores for each of the three time periods: before intervention (T1), after intervention (T2) and after the period of no intervention (T3). Solid lines represent direct measures of reading ability and dotted lines represent skills predictive of reading ability (as discussed in text). Symbol Imagery (in white) corresponds with the y-axis on the right because unlike the other measures the scores are raw and not standardized. Significant increases in score are noted (* p<0.05 ** p<0.01 ***p<0.001) as tested by post hoc t-tests run after a one way repeated measures ANOVA. No significant changes were seen between time points 2 and 3.
Figure 2: Gray Matter Volume Increases over the Course of Reading Intervention
Top) Percent change in GMV signal for the four clusters identified in the VBM2 toolbox pipeline. Bottom) Statistical parametric maps showing the four clusters. Ovals around clusters correspond to the color scheme in the top of the figure. A=left fusiform/hippocampus B=right hippocampus C=left precuneus D=right cerebellum. Scales represent the F score. Top scale corresponds to the left fusiform/hippocampal cluster, bottom scale corresponds with the right hippocampus, left precuneus and right cerebellum.
Scatter plot of GMV change in arbitrary units vs. score change. Blue diamonds represent GMV change in the left precuneus cluster vs. score change on the phonological awareness test (LAC-3). The Pearson’s correlation coefficient for this association is $R=0.688$ ($R^2=0.4732$) and was significant at $p<0.05$. Red squares represent GMV change in the right cerebellum cluster vs. score change on the pseudo word reading (Word Attack) test. The Pearson’s correlation coefficient for this association is $R=0.7484$ ($R^2=0.5601$) and was significant at $p<0.01$. 
Table 1: Behavioral Profile

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### Table 2: Coordinates of VBM Results

**Significant Clusters Identified by One Way ANOVA (within subjects)**

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**Significant Clusters Identified by T2-T1 Contrast**

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**Significant Clusters Identified by T3-T1 Contrast**

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DISCUSSION AND CONCLUSIONS

Overview

Since the time of Samuel Orton there has been interest in a brain-based account to explain why individuals with dyslexia struggle to attain reading in the absence of other obvious deficits (Orton, 1925, 1937). With Galaburda’s post mortem studies (Galaburda and Kemper, 1979; Galaburda et al., 1985; Humphreys et al., 1990), the focus turned to neuroscientific theories of dyslexia. The advent of human imaging technologies has led to studies investigating the functional and anatomical differences between dyslexics and typical readers in both adults and children (Richlan et al., 2011, 2012; Linkersdörfer et al., 2012). These differences have tended to be located within areas that are included in models of word reading, such as temporoparietal cortex (superior temporal gyrus/inferior parietal lobule), occipitotemporal cortex (inferior temporal/fusiform gyrus), and inferior frontal gyrus (Pugh et al., 2000a, 2001), helping to solidify the hypothesis of a language based disorder. However, the question of whether these functional and anatomical differences represent a cause of dyslexia or a consequence of their reduced reading ability remains unanswered. This is often described as an open question in many of the published reports on anatomical variance in dyslexia. Recent work from another field, the study of illiterate adults, has highlighted changes in brain anatomy that follow the acquisition of reading (Carreiras et al., 2009). This places further pressure on the need to disambiguate the reading experience that may in part be responsible of the anatomical differences attributed to dyslexia from those driven by the disorder per se. This dissertation tackles this issue through the use of a reading-level match study to examine gray matter volume differences in dyslexia.
Another approach to understanding the underlying cause of dyslexia is the coupling of remediation of the reading deficit with brain imaging data. Examination of structural changes following reading intervention have been limited to diffusion tensor imaging, yet are critical for measures of gray matter volume, as there is a significant body of work on gray matter differences in dyslexia. Whether those regions identified to be smaller in dyslexia also change following intervention, can complete the picture, with the prediction that cortical hypotrophy might be addressed through successful reading remediation.

The acquisition of anatomical data to address the brain basis of successful reading intervention can be significantly enhanced through adding information about brain function using functional MRI measures. There are now several publications in children and adults showing brain activation changes following remediation. However, how specific these changes are to reading as opposed to learning in the more general sense is unknown. This dissertation addresses this issue by using a within subject design that includes an active control intervention so that changes specific to the reading intervention can be disambiguated from more general changes due to learning and being tutored.

Chapter 1 addressed the question of cause vs. consequence of gray matter volume (GMV) and white matter volume (WMV) differences between dyslexics and controls using a reading level-matched design. Reduced temporal GMV in the age-matched comparison was not replicated in the reading level-matched design, while reduced right precentral gyrus GMV was consistent
across both comparisons. We conclude from this study that the left hemisphere differences observed in gray matter volume represent more than etiological differences in dyslexia; one the possibility is that they are in part the consequence of less reading experience. Chapter 2 used a reading intervention to interrogate functional brain changes following the intervention, including a within subjects control math intervention in order to help specify changes specifically attributed to the reading intervention. Increased activation was found following the reading intervention in left inferior frontal gyrus along with reduced activation in several sensorimotor regions including bilateral precentral gyrus. In addition, we were able to find in the fMRI data activity that was predictive of the success of the reading intervention suggesting that a specific level of brain use can signal a “readiness” of the brain to respond well to the intervention. Specifically, bilateral supramarginal/angular gyrus activation was correlated with increase in single word reading scores following the intervention, demonstrating that while the actual intervention-induced changes occur in left frontal cortex, the integrity of posterior brain areas during reading are predictive of the response to intervention. Chapter 3 investigated GMV changes during this same reading intervention in a subset of the participants from Chapter 2, allowing an additional level of interpretation for the brain basis of dyslexia as well as for understanding structure function relationships within the reading network. GMV increases following the intervention were found, surprisingly, not within regions associated with reading, but rather with regions known to play a role in learning, such as bilateral hippocampus and right cerebellum.
This series of results (obtained from the same sample of dyslexic students) converge to show that there are differences in brain anatomy in dyslexia but that some differences likely follow simply because dyslexics read less. At the same time, other anatomical aberrations likely predate reading acquisition and prevent use of areas that are normally engaged in reading, even following intensive reading intervention. In the case of intervention, more general learning mechanisms are called into play, as reflected by anatomical growth in the gray matter in hippocampus and cerebellum. Finally, functional changes are observed in frontal left hemisphere cortex and the response to intervention can be foreshadowed by task-related signal change during reading in bilateral parietal cortex.

The following sections will address implications of the results pertaining to what is known about the reading network in dyslexia in temporoparietal cortex, inferior frontal gyrus, and occipitotemporal cortex, as well regions within the sensorimotor system not typically associated with reading, including the cerebellum and precentral gyrus. The work will also be addressed in terms of how it compares to what has been learned from illiterate studies and what this means for how the dyslexic brain is remediated, and future directions for new work in the field will also be discussed.

The Reading Network in Dyslexia

Temporoparietal Cortex

The role of temporoparietal cortex and dyslexia has been of interest since the early post mortem studies of Galaburda and colleagues (Galaburda and Kemper, 1979; Galaburda et al.,
Together with the behavioral work demonstrating that dyslexics struggle with the linguistic aspects of reading, especially phonological processing (Lyon et al., 2003) these findings converged to form a biologically driven model of language-based reading deficits in dyslexia (Démonet et al., 1992; Pugh et al., 1996, 2000a, 2001; Rumsey et al., 1997b; Jobard et al., 2003). Functional studies using a variety of tasks have implicated reduced activation in dyslexics across several temporoparietal locations including middle and superior temporal gyri, and inferior parietal lobule (Paulesu et al., 1996, 2001; Shaywitz et al., 1998, 2002; Brunswick et al., 1999; Rumsey et al., 1999; Pugh et al., 2000b; Grünling et al., 2004; Hoeft et al., 2006, 2007). Differences in the specific location of these reductions in activity vary likely due to task differences and reporting of peak intensities, but recent meta-analyses have localized differences in temporoparietal cortex across studies to left middle temporal gyrus (Richlan et al., 2011; Linkersdörfer et al., 2012), left superior temporal gyrus (Maisog et al., 2008; Richlan et al., 2011; Linkersdörfer et al., 2012), left inferior parietal lobule (Maisog et al., 2008; Richlan et al., 2011; Linkersdörfer et al., 2012), right superior temporal gyrus (Maisog et al., 2008; Linkersdörfer et al., 2012), and right inferior parietal lobule (Richlan et al., 2011).

While there seems to be a definite involvement of these regions in dyslexia, the question of their specific role is still in question. Studies in pre-readers report anatomical and functional differences between children with a family history of dyslexia compared to children without a family history of dyslexia (Raschle et al., 2011, 2012). The authors argue that these results speak to the directionally of these anomalies, being casual rather than consequential, and cite genetically driven disruptions in this region as a likely cause (Galaburda et al., 2006). What
happens to these regions when dyslexia is remediated has been examined with functional MRI (but not structural MRI). These studies have shown increasing activation in temporoparietal regions, however often with foci that are located inferior or posterior (Aylward et al., 2003; Temple et al., 2003) to the locations suggested to be involved in reading in non-dyslexic populations (see meta-analyses by (Maisog et al., 2008; Richlan et al., 2011). The inability to engage brain areas used by typical readers, even once dyslexics improve their reading abilities, suggests an insurmountable problem in these regions, that may be offset by recruitment of other regions, potentially representing compensatory mechanisms (Pugh et al., 2000a; Shaywitz et al., 2003). In our studies, we found no functional changes or anatomical changes here following the reading intervention. Instead, we found activity in supramarginal/angular gyrus was predictive of the amount of gain in single word reading skill following the intervention. In other words, brain activity here is beneficial for good reading intervention outcome, but it is not the site of functional or anatomical recovery.

This last result is somewhat surprising, given that one might expect increased brain activity and brain volume in an area that is so commonly found to be associated with reading. Another unexpected aspect of our work is that the anatomical differences observed in this area cannot be entirely attributed to dyslexia per se. Posterior middle temporal gyrus/superior temporal sulcus GMV was reduced in dyslexics compared to their age-matched, but not reading level-matched controls. This suggests that GMV in this region is at least in part due to reading level. Together these two results fail in an important framework for testing causality in dyslexia – the reading level match design and changes following reading intervention. However, in the context of brain
imaging and immediate brain changes following intervention, this approach might be less useful, especially if the anatomical profile in typical readers has been cultivated over years of skilled reading (as opposed to a relatively brief intervention).

From our data, along with previous work by Galaburda and colleagues and the remediation studies described above, one can formulate the process that might be representative of temporoparietal cortex functioning in dyslexia. The ectopias in perisylvian areas as reported in the post mortem studies reflect an anomaly in cortical migration during development (Galaburda and Kemper, 1979; Galaburda et al., 1985; Humphreys et al., 1990). It is possible these ectopias disrupt brain tissue in such a way that reading and phonological processing cannot be properly implemented in these temporoparietal areas. For the same reasons, phonologically based reading intervention also cannot engage temporoparietal cortex and no structural or functional changes ensue that are measured in our study. At the same time, because these areas are not participating in the reading process, they do not grow in terms of gray matter volume. As with other skills, typical readers most likely show some use-dependent growth of GMV here, much as been shown in other situations of learning (e.g. medical students studying for the exams). By comparison, the dyslexics, who do not read as well and as often, do not show such use-induced growth and this manifests as between group differences in GMV. This is why GMV differences are observed compared to age-matched controls (but not reading level-matched), but also why the reading intervention does not result in GMV increase here. Thus, the disrupted integrity of temporoparietal cortex that occurs in development may underlie the functional and structural differences observed in dyslexia as well as limit the ability to remediate this region with training.
Inferior Frontal Gyrus

The inferior frontal gyrus has also been of interest in the dyslexia literature, although the results and their interpretation have been much more variable than that of temporoparietal cortex. Left inferior frontal gyrus is involved in reading and also plays a role in phonological processing, in particular in articulatory coding (Pugh et al., 2000a, 2001). Dyslexia studies often find differences in function and structure in left inferior frontal gyrus, though results have been found in both directions (greater and lesser) for activation in this region, and this is reflected in the variable results in meta-analyses of both activation and GMV differences (Eckert, 2004; Maisog et al., 2008; Richlan et al., 2011, 2012; Linkersdörfer et al., 2012). Greater frontal activity (inferior frontal and premotor areas) has been interpreted as “compensation” for underactivity in posterior brain regions, including parietal and inferior temporal regions (Pugh et al., 2000a, 2001), however as noted above, some studies indicate less activity in the IFG in dyslexia (Paulesu et al., 1996; Rumsey et al., 1997a). A recent meta-analysis shows underactivation here in adults, but not in children (children show overactivation in a more posterior portion of inferior frontal gyrus and precentral gyrus; Richlan et al., 2011). The recent meta-analyses of GMV VBM studies, reports no significant differences in the volume of IFG (Linkersdörfer et al., 2012; Richlan et al., 2012).

Chapter 1 did not show GMV differences in left inferior frontal gyrus for either comparison, though the age-matched WMV analysis showed some underlying WMV reductions in the dyslexic group. Also, GMV did not change with the reading intervention in Chapter 3; however,
the results from Chapter 2 show an increase in activation in left inferior frontal gyrus (including some middle frontal gyrus). Previous remediation studies in children have also shown increases in this area (Aylward et al., 2003; Temple et al., 2003; Shaywitz et al., 2004). While there is still argument over whether activation of this region is hyper or hypoactive in dyslexics compared to controls, a developmental study of reading using the same task and acquisition parameters found that inferior frontal gyrus becomes more active as reading ability increases suggesting that increased activation here may represent a normalization of activity following the intervention (Turkeltaub et al., 2003).

Though Galaburda’s findings also show ectopias within inferior frontal gyrus, the findings tend to be concentrated to perisylvian areas within this region, often leaving large portions of inferior frontal gyrus relatively unaffected (Galaburda et al., 1985). This along with the variability of its involvement in the literature, and the ability of reading interventions to increase activation in this region suggests that inferior frontal gyrus may be less disrupted in developmental dyslexia, and some of the burden of phonological processing typically attributed to temporoparietal regions may be picked up by inferior frontal gyrus. This would explain why we did not see GMV differences in dyslexics compared to controls, but were able to find increased activation following the reading intervention.

Though we did not see GMV increase in inferior frontal gyrus following the intervention in Chapter 3, there are possible explanations for this. First, the thresholds used for significance differed across Chapters 2 and 3, reflecting differences used in structural and functional studies
in the literature. Also, Chapter 3 included a smaller sample size than Chapter 2 which could affect the results. Finally, it is not necessary for there to be underlying GMV changes in order to observe a change in activation. Activation changes could represent local retuning of neuronal populations as opposed to increasing dendritic volume (which could be picked up using VBM). Also, GMV changes may not be realized until after a more prolonged use of this region. Future studies will be needed to disentangle this relationship.

Occipitotemporal Cortex

The third region of the “traditional” reading network, occipitotemporal cortex, is home to the visual word form area which is believed to be involved in the recognition of whole real words (Pugh et al., 2000a, 2001; Cohen et al., 2002). Meta-analyses have shown consistently less activation in dyslexics compared to controls here, interestingly the only region to be consistently underactivated in both studies of adults and children (Maisog et al., 2008; Richlan et al., 2011; Linkersdörfer et al., 2012). Recent VBM meta-analyses are split as to the consistency of GMV reductions here (Linkersdörfer et al., 2012; Richlan et al., 2012). While this region is not typically associated with phonological skills, it is involved in processing commonly encountered words, and Dyslexia is often associated with secondary deficits including a reduced sight word vocabulary (Lyon et al., 2003).

Somewhat unexpectedly, the data from this dissertation does not provide any information on the role of occipitotemporal areas in dyslexia. Previous reading interventions in children have shown increased activation in occipitotemporal regions (Aylward et al., 2003; Temple et al.,
though location of the changes is variable and task differences across studies could be the reason for this. In Chapter 2, we did not report on changes in the left occipitotemporal cortex in the dyslexics following the intervention. Since the intervention involved visual imagery, this outcome was somewhat unexpected. At the same time, the reliability of previous reports of change in this region following intervention is somewhat weakened by the fact that some of these studies did not make use of a control group (e.g. Shaywitz et al., 2004) and can therefore not be assured that changes in occipitotemporal cortex are actually driven by their intervention. We also did not find GMV differences in occipitotemporal cortex in Chapter 1, and this is not unexpected given the lack of consistency in the meta-analyses as discussed above. While the results here do not discount the involvement of occipitotemporal regions in dyslexia, they do not support the anatomical differences observed here between dyslexics and typical readers or changes here in brain activity following the remediation of reading. It is possible that the differences observed here in the literature reflect that this region is susceptible to improper development as a side effect of the problems with reading acquisition.

Precentral Gyrus

Turning to brain regions not traditionally thought of as “reading areas”, a more current model of reading (Dehaene, 2009) comprises several areas outside those discussed by Pugh and colleagues (Pugh et al., 2000a, 2001) including sensorimotor regions such as premotor cortex, early visual cortex in the occipital lobe, and more posterior parietal regions involved in attention. This more distributed model of reading in the brain is also more accommodating to other theories
on dyslexia and expand beyond the more commonly considered core phonological deficit hypothesis by including theories that focus on more general sensorimotor deficits. The framework of one such theory rests upon implicit learning deficits in dyslexia (as measured using the serial reaction time task), with corresponding GMV and activation reductions compared to typical readers in premotor cortex (Vicari et al., 2003; Menghini et al., 2006, 2008). The authors relate this observation to work suggesting involvement of implicit learning as well as visuo-spatial learning and articulatory commands in learning to read.

In the context of this work it is interesting that the only consistent result of less GMV in dyslexia across both the age- and reading level-matched comparisons reported in Chapter 1 was in the right precentral gyrus, suggesting this region, outside the classical language network, may be causal to dyslexia. Bilateral precentral gyrus has been shown to be active when reading words compared to rest in dyslexics and controls (Brunswick et al., 1999) as well as in a developmental study of normal readers (Turkeltaub et al., 2003). Right precentral gyrus specifically has been implicated in the integration of oculomotor and somatomotor information (Iacoboni et al., 1997) which could provide a link to reading in the context of sensorimotor theories in dyslexia. Nevertheless, these findings are somewhat unexpected based on the literature on brain activity underlying reading. It is also of note that the work by Galaburda and colleagues did not reveal ectopias in this region (Galaburda et al., 1985; Humphreys et al., 1990).

However, the importance of this region was echoed in Chapter 2, where we report decreased activation bilaterally in pre- and postcentral gyri following the reading intervention. It is
impossible to relate this finding to the previous publications on the functional changes that follow reading intervention in dyslexia, as these do not report on relative decreases following the intervention. It is possible these regions were being relied on prior to the intervention as a compensatory strategy and following the intervention the activation shifts to left inferior frontal gyrus. On the surface the GMV differences from Chapter 1 and functional decreases following reading intervention from Chapter 2 seem contradictory. However, just because right precentral GMV is less in dyslexia does not mean it should show increased activation with a reading intervention. The children with dyslexia could still have been activating bilateral precentral and postcentral gyri prior to the intervention in a compensatory manner and the reading intervention trains other brain regions (inferior/middle frontal gyrus), leading to a shift in strategy during the task.

Cerebellum

Another non-language based theory of dyslexia centers specifically on another sensorimotor region, the cerebellum. The cerebellar deficit hypothesis posits that dyslexics have problems with automaticity and skill learning and support for this theory comes from behavioral studies using a wide range of skills thought to rely on cerebellar integrity (e.g. balance and dual task motor performance; Nicolson et al., 2001). In addition there is evidence also in cerebellar differences compared to controls in work involving functional and structural brain imaging (Nicolson et al., 1999; Laycock et al., 2008). While the cerebellum is not traditionally included in models of reading, there is much evidence for its involvement in aspects of language such as articulation and word retrieval (For Review see Price, 2012) and bilateral cerebellum was found
to be reduced in gray matter volume dyslexia in one of the recent VBM meta-analyses (Linkersdörfer et al., 2012).

In our own work we found GMV increases within the right cerebellum following the reading intervention (Chapter 3). Further, the GMV increases here positively correlated with increase in pseudoword reading skill. This provides some support for the involvement of the cerebellum in dyslexia and because the GMV change correlates with pseudoword reading, it speaks to the involvement of the cerebellum in phonological decoding. It also, along with the results pertaining to precentral gyrus, suggests an importance of sensorimotor systems in dyslexia that warrants more attention to how these areas contribute to reading problems.

**Lessons from Illiterate Studies**

The interpretation of the results from this dissertation also benefit from an examination of recent work examining ex-illiterates (individuals who did not attend school, but learned to read as adults). Studies of reading acquisition in adults have the benefit of examining the effect of learning to read on brain structure and function at a time when the brain is more stable, whereas studies of reading in children occur in the context of other significant brain changes driven by development. Learning to read as an adult was associated with both functional (Dehaene et al., 2010) and anatomical changes (Carreiras et al., 2009). Greater activation was found with literacy throughout the language network, including the visual word form area, posterior and anterior superior temporal gyrus and inferior frontal gyrus (Dehaene et al., 2010). Greater GMV
was found in ex-illiterates in several posterior regions including left supramarginal gyrus, bilateral angular, superior temporal, middle temporal and dorsal occipital gyri.

The fact that temporoparietal changes were found in adult learners of reading but not in our study of reading interventions in dyslexic children may at first be disappointing, but can be explained. One possibility is that the ex-illiterates have had a longer period to learn to read maturely, whereas our intervention was over an 8 week period. Another possibility is that this again speaks to a disruption in these posterior brain regions in dyslexia such that they cannot properly be engaged and therefore will not show changes with the intervention. The GMV changes observed in Chapter 3 in fact seem to happen in structures more involved in more general aspects of learning and memory (bilateral hippocampus, cerebellum) and imagery (precuneus). These could represent compensatory changes in the absence of being able to engage the more typical brain regions involved in literacy. It is also interesting that that the GMV study of ex-illiterates does not find anterior structures increasing in gray matter with the acquisition of literacy. This is consistent with our observation of no changes in inferior frontal gyrus in GMV in Chapter 3, even though we did find an increase in activation in this area in the dyslexics following intervention (Chapter 2).

Considerations and Future Directions

While the results presented here contribute to the knowledge of the brain basis of dyslexia, they also, like most science does, lead to new questions. Each of the three studies described above were performed with pediatric participants. Pediatric studies are still relatively rare and in
the field as a whole, results obtained in children and adults are often lumped together for making conclusions about the findings. However, there is reason to think the brains of adults and children with dyslexia are not necessarily the same. In one of the recent meta-analyses of functional studies, the authors examine the findings for children and adults separately (Richlan et al., 2011). Interestingly, while there are small areas of overlap, most of the maps of children and adults differ in their location of findings in temporoparietal, inferior temporal and inferior frontal areas. This is unsurprising, as adults have had a greater opportunity to develop strategies and compensate for their reading problems. Some investigators have looked at the long term outcome of adolescents (Hoeft et al., 2011) and adults (Shaywitz et al., 2003) with dyslexia and have found right hemisphere areas, in particular right inferior frontal gyrus to be involved in this compensation.

Unlike studies utilizing functional MRI, anatomical studies examining GMV differences have been performed primarily in adults with dyslexia, and as such there has been no equivalent split of adult and pediatric data for a meta-analysis. To date only two VBM studies besides the data provided here have been done in children (Eckert et al., 2005; Hoeft et al., 2007), and there are numerous reasons to expect that anatomical differences between dyslexic children and controls would differ in nature than those obtained in adult participants. Longitudinal studies looking at reading development and the brain (both anatomically and functionally) in both typical and dyslexic children are needed to allow for a full understanding how both mature and which areas change with reading over time.
Another important point to consider is how to remediate those individuals who do not respond to intervention. While studying dyslexia aids our understanding of typical reading, the primary goal is to help those who struggle to read. In the studies presented here, and previous imaging intervention studies in children, while the average change in reading scores improve following instruction, not all the participants show significant gains (Aylward et al., 2003; Temple et al., 2003; Shaywitz et al., 2004; Meyler et al., 2008). The correlation between bilateral supramarginal/angular gyrus activation and reading gains presented here may suggest that the ability to succeed in a given intervention depends on the ability to engage certain brain regions, thereby serving as a marker of readiness. This is important new information, especially given that the behavioral data does not reveal anything to suggest performance prior to the intervention is indicative of response to intervention. Most reading interventions given in the published literature are focused on training of phonological skills. While this is effective for most, it is unknown whether different types of training would result in reading gains in those individuals who do not benefit from phonologically based remediation. Future investigation needs to address this question.

Lastly, while not directly investigated within the material presented here, there is reason to believe females and males with dyslexia may be different. The incidence of dyslexia is higher in males than females (Rutter et al., 2004) suggesting something inherently different between the sexes with regards to dyslexia. Previous behavioral work has also suggested differences between the sexes, but this issue is rarely if ever addressed in the neuroimaging literature (The Orton Dyslexia Society, 1981; Lambe, 1999). If sex is considered, it is usually only to assure the
groups being compared are matched (including the data presented here). Considering the differences in localization of findings across dyslexia neuroimaging studies and the difference in the ratio of males to females across studies, it is of interest to the field to investigate. Albeit with much too small a sample to truly split by sex, a cursory look at GMV and WMV differences in the age- and reading level-matched groups for males and females separately show the differences may not be consistent across sex (See Appendix A.3-A.6). However, another study from our laboratory (Evans et al., In Press) found different results when comparing men and women as well as boys and girls with dyslexia to sex- and age-matched controls separately. Future studies employing structural and functional imaging need to investigate dyslexia in both sexes and test whether sex is related to any differences in behavior, brain structure, brain function and response to intervention.

Since the time of Samuel Orton’s pioneering work (Orton, 1925, 1937) the field has made great strides in determining the relationship between the brain and the reading deficits in dyslexia, in large part due to increased awareness of the problem and access to non-invasive brain imaging technology. However, many questions remain unanswered and there is still much work to be done to further our understanding of what specific brain abnormalities may cause the reading deficits observed in those who struggle to read and how we can best remediate those deficits.

Conclusions
The studies presented here provide some insights into some of the pressing issues on the neural basis of dyslexia. Specifically, the results presented in this dissertation speak to the complexity and multiplicity of regions involved in reading, including brain areas that are not traditionally considered to be involved in language, but rather sensorimotor functions, such as precentral gyrus and the cerebellum. Less gray matter volume in the left temporoparietal cortex appears to be in part due to a lack of reading experience as well as possibly harboring anomalies that are also the cause of the reading deficit, so much so that successful reading intervention does not produce any changes in this region either in terms of brain function or brain anatomy. At the same time successful reading intervention is predicated by activation in the inferior parietal cortex. In contrast, left inferior frontal gyrus does not show GMV differences compared to controls, but does show an increase in activation following the intervention. This suggests that inferior frontal gyrus is more intact in dyslexia, and is receptive to the phonologically based intervention performed here. Sensorimotor regions including right precentral gyrus and cerebellum also seem to play an important role and the results speak to alternative theories of dyslexia that focus on sensorimotor deficits. Together, the results of this dissertation argue for disruption in both language and sensorimotor networks, and further research will need to address how these two systems contribute uniquely or together to produce the reading deficits observed in dyslexia.
APPENDIX A

Figure A.1: ROI Analysis of GMV from Age-Matched Clusters

Extracted GMV data from MarsBaR generated ROIs.

Average % GMV within clusters identified in the age-matched comparisons for the age-matched groups (top) and reading level-matched groups (bottom). As with the whole brain analysis, the only region of the five that shows a difference in GMV for both the age- and reading level-matched comparisons is the right precentral gyrus.

* = p<0.05 Bonferroni corrected for multiple comparisons
Extracted WMV data from MarsBaR generated ROIs.

Average % WMV within clusters identified in the age-matched comparisons for the age-matched groups (top) and reading level-matched groups (bottom). As with the whole brain analysis, none of the areas identified in the age-matched comparison showed significant differences in the reading level-matched comparison.

* = p<0.05 Bonferroni corrected for multiple comparisons

# = p = 0.05 Bonferroni corrected for multiple comparisons
Figure A.3: GMV ROI Analysis of Age-Matched Groups Split by Sex

Extracted GMV data from MarsBaR generated ROIs.

For the same clusters interrogated in Figure A.1, GMV signal was extracted for each subject and each analysis was split by sex. For the dyslexic group there were 9 females and 6 males. The age-matched control group had 6 females and 9 males. Results look different in the two groups, however sample size is very low and the results are to be considered very preliminary.

* = p<0.05 Bonferroni corrected for multiple comparisons

# = p<0.05 uncorrected for multiple comparisons
Figure A.4: GMV ROI Analysis of Reading Level-Matched Groups Split by Sex

Extracted GMV data from MarsBaR generated ROIs.

For the same clusters interrogated in Figure A.1, GMV signal was extracted for each subject and each analysis was split by sex. For the dyslexic group there were 9 females and 6 males. The reading level-matched control group had 7 females and 8 males. Results look different in the two groups, however sample size is very low and the results are to be considered very preliminary.

* = p<0.05 Bonferroni corrected for multiple comparisons

# = p<0.05 uncorrected for multiple comparisons
Figure A.5: WMV ROI Analysis of Age-Matched Groups Split by Sex

Extracted WMV data from MarsBaR generated ROIs.

For the same clusters interrogated in Figure A.2, WMV signal was extracted for each subject and each analysis was split by sex. For the dyslexic group there were 9 females and 6 males. The age-matched control group had 6 females and 9 males. Results look different in the two groups, however sample size is very low and the results are to be considered very preliminary.

* = p<0.05 Bonferroni corrected for multiple comparisons

# = p<0.05 uncorrected for multiple comparisons
Figure A.6: WMV ROI Analysis of Reading Level-Matched Groups Split by Sex

Extracted WMV data from MarsBaR generated ROIs.

For the same clusters interrogated in Figure A.1, WMV signal was extracted for each subject and each analysis was split by sex. For the dyslexic group there were 9 females and 6 males. The reading level-matched control group had 7 females and 8 males. Results look different in the two groups, however sample size is very low and the results are to be considered very preliminary.

* = \(p<0.05\) Bonferroni corrected for multiple comparisons

# = \(p<0.05\) uncorrected for multiple comparisons
APPENDIX B

In Scanner Behavior

Our findings of change in fMRI signal following the reading intervention cannot be attributed to differences in task performance during the scan. Overall accuracy across the three visits for all subjects (n=31) was 89.15% (s.d=9.01%) and average reaction time across the three visits for all subjects was 1026.15ms (s.d.=176.28ms). One-way repeated measures ANOVAs showed no difference in overall accuracy (F=0.33, p=0.715) and reaction time (F=0.36, p=0.696) across the three visits. Examining real words and false font stimuli separately, average accuracy for real words was 89.57% (s.d.=8.43%) and average reaction time was 1013.45ms (s.d.=161.82ms), while average accuracy for false font was 88.76% (s.d.=9.58%) and average reaction time was 1038.84ms (s.d.=189.66ms). There was no change in the difference between words and false font stimuli, as determined by one way repeated measures ANOVAs, for accuracy (F=2.28, p=0.108) or reaction time (F=0.363, p=0.697) across the three visits.

Changes in Math Skills Following Math Intervention

For the 19 subjects who underwent the math intervention, significant gains in math skills were observed during the math intervention. Prior to the start, average standard score for these subjects was 96 (s.d. 12.5) on Calculation, 105 (s.d. 14.3) on Math Fluency, and 86 (s.d. 14.6) on Applied Problems. Subjects showed significant gains in each of these measures during the math intervention (paired t-tests for pre-math intervention vs. post-math intervention): Calculation standard scores rose an average of 15.4 (s.d. 9.8) (t = 6.871, p = 1.9 x 10-6); Math Fluency standard scores rose an average of 9.2 (s.d. 8.4) (t = 4.774, p = 1.5 x 10-4), and Applied
Problems standard scores rose an average of 2.6 (s.d. 4.7) ($t = 2.377, p = 0.0288$). During the reading intervention, no significant gains were observed in any of the math measures. Calculation standard score ($t = 0.650, p = 0.524$) and Applied Problems standard score ($t = 0.527, p = 0.605$) showed no significant change. Math Fluency showed a significant decrease in standard score on average 5.7 (s.d. 8.1) ($t = 3.047, p = 0.007$).

**Behaviors that Predict Magnitude of Reading Gain following Intervention**

In order to examine behavioral predictors of reading gains on measures of single real and pseudoword reading following intervention (WJ-WID, single real word reading; WJ-WA, single pseudo-word reading), two stepwise regressions were conducted. The first regression examined if skill prior to intervention contributed to single word reading score gain using the standard scores for each of the behavioral measures (described above in the behavioral changes section) and age as predictors. None of these measures contributed significant variance to the change in single word reading score following the intervention. When the regression was run using change in pseudo-word reading following intervention as the dependent variable, there were also no significant results.

**Brain Activity within Prediction Clusters for Words and False Font**

The results could be driven by either an increase in percent signal change for words vs. fixation or a decrease in percent signal change for false font vs. fixation. In order to determine which, mean percent signal change for words and false font for these two clusters was extracted using the MarsBaR toolbox (Brett et al., 2002). Percent signal change for words-fixation showed
a positive correlation with change in single word reading standard score (WJ-WID) in both the left (R = 0.525, R² = 0.276, p = 0.002) and right (R = 0.500, R² = 0.250, p = 0.004) hemisphere clusters (Figure B.4). Percent signal change for false font-fixation showed a negative correlation with change in single word reading standard score (WJ-WID) in both the left (R = -0.507, R² = 0.256, p = 0.004) and right (R = -0.647, R² = 0.419, p < 0.001) hemisphere clusters (Figure B.4). These correlations survive Bonferroni correction for multiple comparisons. The average percent signal change for words vs. false font in these regions prior to intervention was not significantly correlated with change in standard scores following the math intervention/null period.

Further, to make sure the results were not biased toward regions showing a decrease to false font stimuli, whole brain analyses were repeated using an exclusive mask of fixation-false font (to identify areas where false font decreases compared to fixation) at a height threshold of p < 0.05 uncorrected and the results were no different than the original analyses.

**GMV Correlated with Change in Single Word Reading Standard Score**

Gray matter volume and fMRI signal are related in that less gray matter volume could give rise to a lower fMRI signal and vice versa. Our finding of fMRI signal in bilateral SMG as a predictor of reading gains could therefore also be attributed to more GMV in this brain region. In order to determine if gray matter is predictive of gains in reading score following reading intervention, a morphometric analysis was run similarly to the functional analysis above. Using voxel based morphometry (VBM) in SPM8, simple regressions of GMV maps from all subjects were run separately for standard score changes following reading intervention of two direct
measures of reading (WJ-WID, single real word reading; WJ-WA, single pseudo-word reading). The whole brain analysis (height threshold p < 0.001 uncorrected, non-stationary cluster corrected p < 0.05) for single word reading (WJ-WID) revealed one significant cluster in right medial frontal gyrus (Talairach coordinates: x = 12, y = -12, z = 59; BA 6). However, a similar analysis revealed no relationships between GMV and gains in pseudoword reading (WJ-WA) and there were no negative correlations for GMV vs. change in standard scores for reading following the reading intervention. While GMV in right medial frontal gyrus was predictive of single real word reading, GMV cannot account for the relationship between fMRI signal in the supramarginal gyrus clusters identified in the functional analysis and degree of reading outcome. Finally, this observation in right medial frontal gyrus was specific to the reading intervention as it did not surface in a similar analysis of GMV prior to the math intervention and ensuing changes in calculation standard score.

Comparing Functional Changes and Predictors to Controls

Lastly, we wanted to see whether the areas identified as changing following reading intervention or predicting reading gains following the intervention were areas that also differed between dyslexics and controls prior to the intervention. For this purpose we utilized a subset of our dyslexic children (n = 15) with a group of typically reading children (n = 15) who were matched on age (dyslexics: 9.87 years, controls: 9.87 years, p = 0.997), performance IQ (dyslexics: 108.2, controls 114.2, p=0.13) and sex (dyslexics: 8 males, controls 8 males, p = 1.00).
We extracted the percent signal change for words > false font stimuli from the clusters identified in the aforementioned analyses. This was 11 clusters: left and right supramarginal gyrus/inferior parietal lobule (areas predictive of reading gains following intervention), left inferior frontal gyrus (area showing increased word > false font activation following intervention from the analysis of all 31 dyslexics), left anterior cingulate, precentral and postcentral gyri (areas showing decreased word > false font activation following intervention from the analysis of all 31 dyslexics), and right posterior cingulate, precuneus, inferior parietal lobule and precentral gyrus (areas showing decreased word > false font activation following intervention from the analysis of all 31 dyslexics). For these 11 clusters only the left inferior frontal gyrus showed a significant difference between groups (p=0.01, not Bonferroni corrected for multiple comparisons).
Table B.1: Behavioral Profile and Intervention Changes by Group

<table>
<thead>
<tr>
<th>Prior to Intervention</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
<th>F(2,30)</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>10.0 (1.6)</td>
<td>9.0 (1.3)</td>
<td>9.9 (1.5)</td>
<td>1.414</td>
<td>0.260</td>
</tr>
<tr>
<td>VIQ</td>
<td>111.9 (11.6)</td>
<td>110.7 (3.4)</td>
<td>107.5 (11.4)</td>
<td>0.538</td>
<td>0.590</td>
</tr>
<tr>
<td>PIQ</td>
<td>100.7 (10.8)</td>
<td>101.1 (10.9)</td>
<td>104.8 (8.9)</td>
<td>0.403</td>
<td>0.672</td>
</tr>
<tr>
<td>FSIQ</td>
<td>107.2 (11.6)</td>
<td>106.8 (6.2)</td>
<td>106.8 (7.6)</td>
<td>0.007</td>
<td>0.993</td>
</tr>
<tr>
<td>Word ID</td>
<td>80.3 (7.9)</td>
<td>76.2 (8.6)</td>
<td>75.8 (7.3)</td>
<td>0.982</td>
<td>0.387</td>
</tr>
<tr>
<td>Word Attack</td>
<td>93 (5.1)</td>
<td>92.7 (7.5)</td>
<td>89.2 (6.0)</td>
<td>1.018</td>
<td>0.374</td>
</tr>
<tr>
<td>Passage Comprehension</td>
<td>84.4 (9.9)</td>
<td>74.4 (15.4)</td>
<td>77 (14.7)</td>
<td>1.528</td>
<td>0.235</td>
</tr>
<tr>
<td>Phonemic Awareness (LAC-3)</td>
<td>102.4 (9.9)</td>
<td>96.8 (6.4)</td>
<td>96.1 (8.4)</td>
<td>1.783</td>
<td>0.187</td>
</tr>
<tr>
<td>Rapid Naming Letters/Numbers</td>
<td>84.4 (11.6)</td>
<td>74.3 (13.7)</td>
<td>77.8 (9.7)</td>
<td>1.974</td>
<td>0.158</td>
</tr>
<tr>
<td>Rapid Naming Colors/Objects</td>
<td>90.7 (13.5)</td>
<td>83.3 (11.7)</td>
<td>79.2 (10.1)</td>
<td>2.313</td>
<td>0.118</td>
</tr>
<tr>
<td>Digit Span</td>
<td>99.0 (12.9)</td>
<td>90.0 (9.3)</td>
<td>92.3 (12.8)</td>
<td>1.693</td>
<td>0.202</td>
</tr>
<tr>
<td>Symbol Imagery</td>
<td>84.4 (9.8)</td>
<td>80.8 (8.5)</td>
<td>74.7 (11.8)</td>
<td>2.502</td>
<td>0.100</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Change After Reading Intervention</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
<th>F(2,30)</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Word ID</td>
<td>5.9 (6.7)</td>
<td>9.3 (6.3)</td>
<td>4.7 (6.1)</td>
<td>1.493</td>
<td>0.242</td>
</tr>
<tr>
<td>Word Attack</td>
<td>7.0 (3.9)</td>
<td>5.1 (6.8)</td>
<td>5.8 (4.7)</td>
<td>0.346</td>
<td>0.710</td>
</tr>
<tr>
<td>Passage Comprehension</td>
<td>5.4 (7.5)</td>
<td>10.1 (10.8)</td>
<td>3.4 (4.6)</td>
<td>1.790</td>
<td>0.185</td>
</tr>
<tr>
<td>Phonemic Awareness (LAC-3)</td>
<td>3.2 (8.6)</td>
<td>7.6 (5.6)</td>
<td>2.7 (11.1)</td>
<td>1.121</td>
<td>0.340</td>
</tr>
<tr>
<td>Rapid Naming Letters/Numbers</td>
<td>4.5 (8.7)</td>
<td>8.3 (7.1)</td>
<td>-2.5 (7.5)</td>
<td>4.971</td>
<td>0.014*</td>
</tr>
<tr>
<td>Rapid Naming Colors/Objects</td>
<td>0.9 (12.2)</td>
<td>4.1 (9.1)</td>
<td>-5.0 (6.5)</td>
<td>2.318</td>
<td>0.117</td>
</tr>
<tr>
<td>Digit Span</td>
<td>-5.5 (15.9)</td>
<td>4.6 (8.9)</td>
<td>5.0 (6.5)</td>
<td>2.635</td>
<td>0.089</td>
</tr>
<tr>
<td>Symbol Imagery</td>
<td>14.2 (7.5)</td>
<td>10.5 (10.2)</td>
<td>13.6 (9.8)</td>
<td>0.502</td>
<td>0.611</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Change After Math Intervention/Null Period</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
<th>F(2,30)</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Word ID</td>
<td>1.9 (7.1)</td>
<td>-2.6 (6.3)</td>
<td>0.4 (3.5)</td>
<td>1.639</td>
<td>0.212</td>
</tr>
<tr>
<td>Word Attack</td>
<td>0.4 (5.5)</td>
<td>2.1 (7.3)</td>
<td>-2.8 (4.7)</td>
<td>1.659</td>
<td>0.209</td>
</tr>
<tr>
<td>Passage Comprehension</td>
<td>0.1 (8.9)</td>
<td>-1.3 (6.7)</td>
<td>2.1 (11.0)</td>
<td>0.374</td>
<td>0.691</td>
</tr>
<tr>
<td>Phonemic Awareness</td>
<td>-0.9 (11.4)</td>
<td>-4.1 (10.0)</td>
<td>-1.4 (5.9)</td>
<td>0.354</td>
<td>0.705</td>
</tr>
<tr>
<td>Rapid Naming Letters/Numbers</td>
<td>-1.7 (4.8)</td>
<td>-2.3 (6.6)</td>
<td>1.6 (6.9)</td>
<td>1.075</td>
<td>0.355</td>
</tr>
<tr>
<td>Rapid Naming Colors/Objects</td>
<td>-0.4 (8.4)</td>
<td>-4.2 (6.3)</td>
<td>2.8 (9.6)</td>
<td>1.958</td>
<td>0.160</td>
</tr>
<tr>
<td>Digit Span</td>
<td>2.0 (13.0)</td>
<td>-0.4 (13.0)</td>
<td>-5.0 (10.3)</td>
<td>0.787</td>
<td>0.465</td>
</tr>
<tr>
<td>Symbol Imagery</td>
<td>-3.3 (11.1)</td>
<td>-2.8 (9.6)</td>
<td>3.9 (7.1)</td>
<td>1.716</td>
<td>0.198</td>
</tr>
</tbody>
</table>

Behavioral profile and intervention behavioral changes separated by intervention group. Group 1 received reading intervention followed by math intervention (n=10), Group 2 received reading intervention followed by null period (n=12) and Group 3 received math intervention followed by reading intervention (n=9). Behavioral data for Group 2 has been previously published (Krafnick et al., 2011).
Table B.2: Peak Coordinates for Functional Changes Following Math Intervention

<table>
<thead>
<tr>
<th>Talairach Peak Coordinate</th>
<th>Cluster Size</th>
<th>T-statistic</th>
<th>Z-score</th>
<th>Anatomical Location</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Word &gt; False Font Increase After Math Intervention</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>x = 40, y = -6, z = -6</td>
<td>1015</td>
<td>5.80</td>
<td>4.30</td>
<td>Sub-Gyral Middle Temporal BA 21</td>
</tr>
<tr>
<td>x = 16, y = -18, z = 25</td>
<td>479</td>
<td>5.20</td>
<td>4.01</td>
<td>Caudate</td>
</tr>
<tr>
<td>x = -20, y = -20, z = -11</td>
<td>374</td>
<td>4.91</td>
<td>3.86</td>
<td>Parahippocampal Gyrus BA 35</td>
</tr>
<tr>
<td>x = -32, y = -58, z = 7</td>
<td>58</td>
<td>4.33</td>
<td>3.54</td>
<td>Sub-Lobar BA 21</td>
</tr>
<tr>
<td>x = 28, y = 17, z = -6</td>
<td>67</td>
<td>3.99</td>
<td>3.33</td>
<td>Insula BA 13</td>
</tr>
<tr>
<td>x = 65, y = -5, z = 17</td>
<td>94</td>
<td>3.88</td>
<td>3.27</td>
<td>Postcentral Gyrus BA 43</td>
</tr>
<tr>
<td>x = -26, y = -44, z = 57</td>
<td>57</td>
<td>3.73</td>
<td>3.17</td>
<td>Superior Parietal Lobule BA 40</td>
</tr>
<tr>
<td>x = 51, y = -13, z = 21</td>
<td>152</td>
<td>3.59</td>
<td>3.08</td>
<td>Postcentral Gyrus BA 43</td>
</tr>
<tr>
<td><strong>Word &gt; False Font Decrease After Math Intervention</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>x = 8, y = -34, z = -18</td>
<td>174</td>
<td>5.06</td>
<td>3.94</td>
<td>Right Anterior Cerebellum/Pons</td>
</tr>
<tr>
<td>x = 12, y = -55, z = -14</td>
<td>63</td>
<td>3.50</td>
<td>3.02</td>
<td>Right Anterior Cerebellum</td>
</tr>
</tbody>
</table>
Figure B.1: Changes in Math Standard Score Following Intervention

- **Reading Intervention**
- **Math Intervention**

* p < 0.05
** p < 0.01
*** p < 0.001
Figure B.2: Brain Regions Showing Change Following Math Intervention for Implicit Reading Task

Paired t-test whole brain maps of word > false font contrasts at $p<0.01$ and at least 50 contiguous voxels. (A) From Figure 2B, the changes following reading intervention for the 19 subjects who
underwent both reading and math interventions. Red indicates increased activation following the intervention and blue indicates decreased activation. (B) Changes following the math intervention for the same 19 subjects who received both reading and math interventions. All visible areas denote increased activation following the intervention. See Table S2 for list of foci. (C) Non-statistical overlay of (A) and (B) showing slight overlap (seen in teal) in the right hemisphere where the right frontoparietal region showing a decrease during the reading intervention overlaps with a right frontoparietal increase during the math intervention.
Figure B.3: Intervention Study Design

31 dyslexic children were randomly assigned to one of three intervention timelines. All groups received the reading intervention, and either a math intervention (active control) or null period (developmental control). Each visit consisted of acquisition of behavioral and imaging data, with 8 week intervention periods between visits 1 and 2 and visits 2 and 3.
Figure B.4: Correlation plots of percent signal change for words-fixation and false font-fixation from clusters identified in whole brain regression analyses.

(Top) Percent signal change (psc) for words-fixation vs. change in single word reading standard score following intervention showed a positive correlation for both left (red squares) and right (blue diamonds) hemisphere clusters. (Bottom) PSC for false font-fixation vs. change in single word reading standard score following intervention showed a negative correlation for false font-fixation vs. change for both left and right hemisphere clusters.
REFERENCES


Stip, E., Mancini-Marie, A., Letourneau, G., Fahim, C., Mensour, B., Crivello, F., and Dollfus, S. (2009). Increased grey matter densities in schizophrenia patients with negative symptoms after


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