TITLE: Simultaneous learning of two languages from birth positively impacts intrinsic functional connectivity and cognitive control

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Abstract

This study explores the effect of individual differences in the age of acquisition of a second language using resting-state functional magnetic resonance imaging (rs-fMRI) to examine functional connectivity and its relation with cognitive control within bilinguals. We compared simultaneous bilinguals, who learned two languages from birth, to sequential bilinguals, who learned a second language following mastery of their first language. Results show an effect of language experience on the strength of anticorrelation between the default mode network and the task-positive attention network and on cognitive control, with simultaneous bilinguals demonstrating stronger anticorrelations between the two networks, as well as superior cognitive control compared to sequential bilinguals. These findings demonstrate that the timing of language learning may have an impact on cognitive control, with the simultaneous learning of two languages being associated with more optimal brain connectivity for cognitive control compared to sequential language learning.

KEYWORDS: resting-state functional magnetic resonance imaging (rs-fMRI); intrinsic functional connectivity; bilingualism; cognitive control; default mode network (DMN); task-positive attention network

1. Introduction

It has now been well documented that language experience has an impact on the brain and that being bilingual may positively affect cognitive control processes (e.g., Bialystok, Craik, & Luk, 2012; Costa & Sebastian-Galles, 2014), but what aspect of the bilingual experience exerts an influence is still a matter of debate. Bilingual language experience has been related to changes in brain structure and function in terms of white matter integrity (e.g., Luk, Bialystok, Craik, & Grady, 2011; Pliatsikas, Moschopoulou, & Saddy, 2015), cortical thickness (e.g., Klein, Mok, Chen, & Watkins, 2014; Mårtensson et al., 2012), gray matter density (e.g., Berken, Gracco, Chen, & Klein, 2015; Mechelli et al., 2004) and functional activity in various brain regions (e.g., Berken, Gracco, Chen, Watkins, et al., 2015; Kovelman, Baker, & Petitto, 2008). Although the majority of studies have compared bilinguals to monolinguals, some have shown differences within bilingual groups themselves, with factors such as age of second language acquisition (AoA) or language proficiency exerting an influence on brain organization.

In bilinguals, AoA has been found to be related to cortical thickness such that earlier AoA has been associated with thinner cortex in the left inferior frontal gyrus (IFG) and thicker cortex in the right IFG, although in this study language proficiency was not held constant (Klein et al., 2014). In another study, simultaneous and sequential bilinguals, who were matched in terms of second language (L2) proficiency and differed only with respect to their accent in L2, were found to have differences in gray matter density (Berken, Gracco, Chen, & Klein, 2015); however, only limited behavioural measures were considered, making it difficult to interpret the exact role of specific brain regions in relation to bilingual language experience. A recent fMRI investigation in sequential bilinguals has also shown that AoA was related to the degree to which brain regions associated with speech-motor control and orthographic to phonological mapping were activated to a greater extent in L2 compared to L1 while reading (Berken, Gracco, Chen, Watkins, et al., 2015). Others have found that AoA is related to patterns of brain activation during lexical retrieval (Perani et al., 2003) and speech processing (Archila-Suerte, Zevin, & Hernandez, 2015) in an L2. AoA of an L2 has also been related to the laterality of language organization, with a meta-analysis showing that early bilinguals (AoA before 6 years old) show bilateral language organization as compared to late bilinguals who showed left hemisphere dominance for language (Hull & Vaid, 2007). Different EEG patterns during language processing tasks have also been related to AoA (Genesee et al., 1978). Taken together, these findings demonstrate the impact of different language experiences, namely AoA, on brain structure and function as well as language organization – a conclusion also reached by Hull and Vaid (2007).

To date, most studies have focussed primarily on brain structural measurements and task-related brain activity in order to examine the effect of different language experiences on the bilingual brain. More recently, people have looked to resting-state functional magnetic resonance imaging (rs-fMRI) as it can identify task-independent effects of language experience on brain function and connectivity. Resting-state fMRI is a measure of spontaneous low frequency (<0.1 Hz) fluctuations in the blood oxygen level-dependent (BOLD) signal while the brain is not engaged in an external task (i.e., at rest) (Cordes et al., 2001). Using rs-fMRI, functionally connected brain regions have been found to show correlated spontaneous low frequency fluctuations in the BOLD signal over time (e.g., Biswal, Zerrin Yetkin, Haughton, & Hyde, 1995; Hampson, Peterson, Skudlarski, Gatenby, & Gore, 2002; Smith et al., 2009). In terms of bilingualism, greater functional connectivity and a more distributed pattern of connectivity has been observed in bilingual as compared to monolingual older adults (Grady, Luk, Craik, & Bialystok, 2015; Luk et al., 2011). A correlation between rs-fMRI and AoA has also been shown, with greater functional connectivity between the left and right inferior frontal gyri and the inferior parietal lobule being associated with earlier AoA (Berken, Chai, Chen, Gracco, & Klein, 2016), although again no additional behavioural evidence was provided to interpret these findings in terms of cognitive control directly.

The current investigation importantly, moves away from a comparison of bilinguals to monolinguals, which, as has been previously noted by others, is a potentially problematic confound given that bilinguals have variable language experience (e.g., Kaushanskaya & Prior, 2014; Luk, 2015; Luk & Bialystok, 2013). We focus more specifically on different groups of bilinguals to determine what aspect of the bilingual language experience might have an effect on brain organization and cognitive control. Using both rs-fMRI and behavioural measures of cognitive control we examine whether the timing and manner in which the L2 is learned has implications for language-experience related differences at the level of the brain and behaviour. We do this by comparing equally high proficiency bilingual individuals who differ in whether they learned both of their languages simultaneously or learned their L2 sequentially, following mastery of their native language. Thus, the question is whether the simultaneous exposure and learning of two languages exerts different influences on brain organization and cognitive control than sequential exposure and use of two languages. In other words, does setting up the language system for two languages from birth have different implications in terms of brain organization and cognitive control than learning an additional language later, using the neural architecture of the already established language?

Previous research has used a variety of tasks to investigate language group differences in cognitive control, including primarily the Stroop task (e.g., Bialystok, Craik, & Luk, 2008; Bialystok, Poarch, Luo, & Craik, 2014; Kousaie & Phillips, 2012, 2017; Kousaie, Sheppard, Lemieux, Monetta, & Taler, 2014), the flanker task (e.g., Abutalebi et al., 2012; Kousaie & Phillips, 2012, 2017), and the Simon task (e.g., Bialystok, 2006; Bialystok, Craik, Klein, & Viswanathan, 2004; Bialystok et al., 2008; Kousaie & Phillips, 2012, 2017; Kousaie et al., 2014). However, findings of language group differences have been variable within and across tasks, and it has been shown that these different tasks show little convergence suggesting that observed effects of language group may be task-specific (see Paap, Johnson, & Sawi, 2015). In the current investigation we chose to use the Simon task because it is a non-verbal cognitive control task that has previously been shown to be sensitive to differences between monolinguals and bilinguals and the inclusion of the three conditions in the Simon task used here allows for a calculation of different measures of cognitive control (described in more detail in the Methods).

In terms of rs-fMRI and cognitive control, research has demonstrated that the resting brain is intrinsically organized in two opposing, or anticorrelated networks, one demonstrating task-related increases in activation (task-positive attention network) and the other showing task-related decreases in activation (task-negative or default mode network; DMN) (Fox et al., 2005). Furthermore, and of particular relevance to the current investigation are studies showing that variations in the degree or strength of anticorrelation between these two networks are related to performance of executive function tasks. Specifically, greater anticorrelations between the DMN and the attention network have been associated with more stable performance on a flanker task (Kelly, Uddin, Biswal, Castellanos, & Milham, 2008), as well as better working memory performance (Hampson, Driesen, Roth, Gore, & Constable, 2010; Keller et al., 2015).

Here we use rs-fMRI to detect language experience-related differences in intrinsic connectivity within the bilingual brain. Specifically, we explore the relationship between the DMN and the task-positive attention network, how this relationship differs as a function of L2 language learning experience, and how this relationship is related to cognitive control. Based on previous research showing that the degree of anticorrelation between the DMN and task-positive network is related to cognitive control (e.g., Kelly et al., 2008) and that AoA impacts brain structure (e.g., Berken, Gracco, Chen, & Klein, 2015; Klein et al., 2014), function (e.g., Archila-Suerte et al., 2015; Berken, Gracco, Chen, Watkins, et al., 2015; Perani et al., 2003), including rs-fMRI (Berken et al., 2016), and organization (Hull & Vaid, 2007), we hypothesized that if AoA has an impact on the development of these brain networks related to cognitive control then: 1) the simultaneous exposure and acquisition of an L2 would be associated with stronger anticorrelation between the two resting state networks as compared to the sequential acquisition of two languages, and 2) this difference in brain connectivity would in turn be associated with differences in cognitive control, with stronger anticorrelations being associated with better cognitive control.

2. Materials and Methods

2.1 Participants

The unique language environment of Montreal provides access to distinct samples of bilinguals who are consistently exposed to both French and English in their daily lives and differ only with respect to when they learned their L2, allowing us to take advantage of homogeneous samples of participants. We tested two groups of French/English bilinguals – one who learned their two languages simultaneously from birth (n=11; mean AoA=0) and the second who learned their L2 after the age of 6 years old, and were matched with the simultaneous group in terms of their L2 proficiency (n=10; mean AoA=7.4). All were highly proficient right-handed English/French bilinguals who use both languages on a daily basis. The groups were matched for chronological age, years of formal education, and general intelligence (Table 1). Participants self-reported good health and did not have knowledge of any languages other than French and English. Exclusion criteria included history of a traumatic brain injury or neurological disorder, any medical conditions or medications known to affect cognitive functioning, or any conditions incompatible with MRI (e.g., metal implants, braces, electronically, magnetically, or mechanically activated devices such as cochlear implants, or claustrophobia). Individuals with musical training were also excluded given the link between musical training and brain organization (Gaser & Schlaug, 2003), as well as the possible interaction between musicianship and bilingualism on cognitive control processes (Schroeder, Marian, Shook, & Bartolotti, 2016).

INSERT TABLE 1 HERE

2.2 Stimuli and Materials

All participants underwent a behavioural testing session in which they completed a test of cognitive control, a language proficiency assessment, and a test of general intelligence.

2.2.1 Simon Task

An arrows version of a Simon task (Simon & Rudell, 1967) was used to assess cognitive control, following previous work examining differences in cognitive control between monolinguals and bilinguals (e.g., Bialystok et al., 2008). The task comprised six blocks of trials (two blocks each of three conditions, presented in counterbalanced order). Participants completed three conditions (control, reverse, and conflict) that comprised directional arrows on a Dell Precision M2800 laptop with 15.1” screen, Windows 7 64-bit operating system, and intel CORE i7-4610M CPU, using E-Prime v2.0 presentation software (Psychology Software Tools, Pittsburgh, PA, USA). The control condition measured response times when no additional processing was required, and was comprised of centrally presented arrows for which participants were required to indicate in which direction the arrow was pointing, using left (for a leftward pointing arrow) and right (for a rightward pointing arrow) response keys. In the reverse condition participants were presented with the same stimuli as in the control condition, but were required to indicate the opposite direction to the direction that the arrow was pointing in (e.g., left key response for rightward pointing arrow). The conflict condition consisted of congruent and incongruent trials randomly intermixed where participants indicated the direction of the stimulus using the left and right response keys; on congruent trials the directional arrow was presented on the same side of the laptop screen as the correct response (e.g., leftward pointing arrow presented on the left side of the screen), on incongruent trials the arrow was presented on the opposite side of the screen as the correct response (e.g., leftward pointing arrow presented on the right side of the screen). Thus, participants were required to ignore the irrelevant spatial information from the position of the stimulus in order to respond to the direction of the arrow. In total there were 96 trials of each type for a total of 384 trials across the entire task.

The three conditions allowed us to examine different components of cognitive control. Specifically, we calculated measures of the Simon effect (i.e., the increase in response time for incongruent relative to congruent trials within the conflict condition), response inhibition (i.e., the ability to inhibit a habitual response, calculated as the increase in response time for the reverse compared to the control condition), and interference suppression (i.e., the ability to suppress interfering spatial information; calculated as the increase in response time for the conflict compared to the control condition).

2.2.2 Language proficiency assessment

Participants completed letter fluency, category fluency, and sentence repetition tasks in both English and French to ensure that they were highly proficient in both of their languages. Scores for the language assessment can be found in Table 1.

In the fluency tasks participants were asked to produce as many exemplars as they could for each letter or category in one minute. For letter fluency in English participants were asked to do this for the letters *F*, *A*, and *S*. In French the letters included *P*, *F*, and *L*. the number of exemplars generated across all three letters in each language were summed to give a single letter fluency score in each language. For category fluency, the categories were *animals* and *fruit* in English and French, respectively. The number of exemplars produced for each category/language was counted to give a score for category fluency in each language.

For the sentence repetition task, participants completed the Recalling Sentences subtest of the Clinical Evaluation of Language Fundamentals – fourth edition (CELF-4; Semel, Wiig, & Secord, 2003), which is an evaluation originally designed to determine if an individual has a language disorder or delay, but it was used here to assess relative language proficiency in English and French. Participants were read sentences out loud and were asked to repeat them immediately after hearing them. Each sentence received a score out of *3*, with *3* representing no errors and *0* representing four or more errors in the repetition of the sentence. Participants completed the sentence recall in both English and French. It should be noted that we did not use standard scoring procedures given that the CELF-4 is designed to identify language disorders or delays in children aged 5-21 years and the majority of our participants were over 21 years old. Rather, we compared performance across languages in order to obtain a measure of relative proficiency in L2.

2.2.3 Matrix Reasoning

The matrix reasoning subtest of the Wechsler Adult Intelligence Scale fourth edition (Wechsler, 2008) was used as a proxy measure of general intelligence. Participants are presented with an incomplete series or 2 x 2 matrix and were required to select the response that best completed the series or matrix from four alternatives. The maximum score was 26 and the test was discontinued when the participant gave 3 consecutive incorrect responses. Standardized scores are provided in Table 1.

2.3 Procedure

Participants completed two testing sessions (an initial session for the behavioural testing and a second session for fMRI scanning. Ethics approval for this study was obtained from the Research Ethics Board at the Montreal Neurological Institute, McGill University, and participants gave their written consent.

2.4 Imaging procedure and analysis

Data were acquired on a 3T TrioTim Siemens scanner using a 32-channel head coil at the Montreal Neurological Institute. Participants were instructed to fixate on a cross that was presented at the center of the screen and to clear their mind. Resting scan images were obtained in 38 3.5 mm thick transverse slices, covering the entire brain (TR=2260 ms, TE=30 ms, matrix size=64 x 64, FoV=224 mm, flip angle=90°); 132 volumes were obtained in 5:04 minutes. High-resolution T_1-weighted images were obtained from a 3D magnetization prepared rapid acquisition gradient echo sequence (slice thickness=1 mm, TR=2300 ms, TE=2.98 ms, matrix size=256 x 256, FoV=256 mm, flip angle=9°, interleaved excitation) were obtained for each participant and used as an anatomical reference. The resting scan was acquired as part of a larger study for which participants completed several functional tasks in the scanner and underwent diffusion tensor imaging; resting scans were acquired at the beginning of the MRI testing session, immediately following acquisition of the anatomical scan.

Resting-state fMRI data were preprocessed using SPM8 (Wellcome Department of Imaging Neuroscience, London, UK), using standard preprocessing steps. Images were realigned and unwarped, slice time corrected, normalized in MNI space and smoothed with a 6 mm full width at half maximum (FWHM) Gaussian kernel. Artifact/outlier scans were excluded from analysis. Outlier scans were identified using ART (Artifact Detection

Tools) and were defined as images in which average intensity deviated more than 3 SDs from the mean intensity in the session, or composite head movement exceeded 1.5.mm from the previous image. The functional connectivity analysis was performed using the custom software CONN (Chai, Castanon, Ongur, & Whitfield-Gabrieli, 2012; Whitfield-Gabrieli & Nieto-Castanon, 2012).

Seed-to-voxel correlations were performed by estimating the temporal correlation between the blood oxygen level-dependent (BOLD) signal in our a priori ROI (seed) and the BOLD signal in all other voxels of the brain. We performed the resting-state connectivity analysis from a single seed region in the ventromedial prefrontal cortex (vmPFC; -3 +39 -2) defined as a 6 mm sphere (Fair et al., 2009); the vmPFC seed was chosen given that it is an anchor region in the DMN (Raichle et al., 2001). This seed-based approach was used in order to identify the brain regions in which the BOLD signal correlated positive and negatively (i.e., anticorrelation) with the vmPFC across the entire group of participants (i.e., the two participant groups were pooled for the overall functional connectivity analysis). First-level correlation maps were produced by extracting the residual BOLD time course from the seed region and computing Pearson's correlation coefficients between the time course in the seed and that in all other voxels in the brain across the entire sample of participants irrespective of Language Group membership. In order to allow for second level GLM analyses, the correlation coefficients were converted to normally distributed z scores using the Fisher's transformation. All reported clusters survived an FWE-corrected threshold of $p<.01$, with voxel-level significance uncorrected of $p<.001$, two-sided.

3. Results

3.1 Language proficiency assessment and Matrix Reasoning

Independent samples t-tests revealed no group difference for any of the measures of language proficiency and general intelligence (all *p*s > .08; Table 1).

3.2 Simon Task

The three conditions of the Simon task were compared across the two groups of participants in a oneway analysis of variance (ANOVA) with the factor Language Group. In addition, the three conditions were used to calculate three different measures of cognitive control (i.e., Simon interference, response inhibition, and interference suppression) that were also analyzed in a oneway ANOVA with the factor Language Group.

Simultaneous bilinguals showed smaller interference suppression scores (calculated by subtracting response times for the control condition from response times for the conflict condition) than the sequential bilinguals (*F*(1,19)=4.49, *MSE*=1022.9, *p*<.05), indicative of better interference suppression. The two language groups did not differ in terms of raw response times for any of the three conditions (all *p*'s > .19) or other measures of cognitive control (all *p*'s > .61); Figure 1 depicts the behavioural data.

INSERT FIGURE 1 HERE

3.3 Functional connectivity analysis

Several classic regions in the DMN (see Buckner, Andrews-Hanna, & Schacter, 2008) showed BOLD fluctuations that correlated positively with the ventromedial prefrontal cortex seed, including the precuneus, posterior cingulate cortex, bilateral angular gyrus, and bilateral temporal regions (seen in red in Figure 2). In addition, there were several regions that showed anticorrelated BOLD signal activity with the vmPFC, including bilateral inferior frontal gyri, dorsal lateral prefrontal cortex (dlPFC) and superior parietal lobule, and left inferior parietal lobule. These regions have previously been shown to be part of the task-positive attention network (Fox et al., 2005) and are involved in attention control (Raz, 2004) (seen in blue in Figure 2); peak coordinates are provided in Table 2.

INSERT FIGURE 2 HERE

INSERT TABLE 2 HERE

In a second step, we examined whether the strength of anticorrelation with the vmPFC differed for the two language groups. Given that we were interested in the relation between cognitive control and the degree of anticorrelation between the DMN and the attention network, we focused on two regions in the frontal-parietal network from the DMN anticorrelated regions (see Table 2) that have been shown to be related to cognitive control: the dlPFC, and inferior parietal lobule (IPL; e.g., Cole & Schneider, 2007; MacDonald III, Cohen, Stenger, & Carter, 2000; Sambataro et al., 2013; Singh-Curry & Husain, 2009). A oneway ANOVA revealed stronger anticorrelations with the vmPFC in simultaneous than sequential bilinguals in the right dlPFC ($F(1,19)=10.44$, $MSE=0.01$, $p<.01$) and the left dlPFC ($F(1,19)=7.69$, $MSE=0.01$, $p=.01$). There were no group differences in the anticorrelation between the vmPFC and the left IPL ($p>.5$). Figure 3, panels A and B depict these regions and functional brain connectivity as a function of Language Group.

As a final consideration of the relationship between intrinsic functional connectivity and cognitive control, we examined whether the degree of anticorrelation between the DMN and the regions of the attention network that showed language group differences in connectivity correlated with behavioural Simon task performance. Indeed, the degree of anticorrelation between the two networks was positively correlated with our measure of interference suppression, indicating that smaller interference suppression scores (i.e., better cognitive control) were associated with greater anticorrelation between the two networks. The Pearson Correlation was significant for both the right dlPFC (r=.45, p=.02) and the left dlPFC (r=.43, p=.03) across all participants. Additionally, we examined the correlations within each group of bilinguals separately and found that there was no significant correlation between interference suppression and functional connectivity between the two networks in the simultaneous group (p's>.30); however, within the sequential group the correlation was significant for the right dlPFC (r=.73, p=.01), and approached significance for the left dlPFC (r=.38, p=.14); these data are represented in Figure 3, panel C.

INSERT FIGURE 3 HERE
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4. Discussion

We examined the effect of bilingual language experience on brain organization and cognitive control using rs-fMRI to look at patterns of intrinsic functional connectivity in simultaneous and sequential bilinguals who were matched on proficiency and language usage. By examining the anticorrelation between the DMN and the task-positive attention network, and their relation with performance on a Simon task that was performed outside of the scanner, we show differences in the effect of bilingualism on cognition across the two groups, with an advantage for simultaneous bilinguals who learned their L2 from birth in terms of both intrinsic resting state networks and behaviour. Specifically, simultaneous bilinguals showed greater anticorrelation between the DMN and the task-positive attention network and better interference suppression than sequential bilinguals.

We show that it is not simply being bilingual that affects cognitive control, but that language experiences have the ability to impact these processes differentially, with simultaneous bilinguals showing smaller interference suppression effects than sequential bilinguals, indicating better cognitive control. This effect was specific to interference suppression and did not emerge in any of the other measures of cognitive control that we examined, or in the individual conditions included in the Simon task. Thus, the bilinguals tested here showed differences in their ability to supress interfering information, but not in their ability to inhibit a prepotent response. This may not be surprising if one considers the processes involved in managing two competing languages, whereby bilinguals must supress interference from one, non-target language while engaging in the target language. According to the adaptive control hypothesis, the constant management of two competing languages requires general cognitive control mechanisms, and different language experiences/contexts can result in adaptation in different control processes (Green & Abutalebi, 2013). Our findings suggest that in our sample of highly proficient bilinguals who use both of their languages on a regular basis, L2 AoA may have a specific impact on the adaptation of the control processes involved in interference suppression.

Interestingly, we did not find a group difference on the Simon effect, which was the increase in RT for incongruent trials compared to congruent trials within the same task block. The Simon effect may be thought of as being similar to interference suppression, which was the increase in response time for the conflict block as compared to the control block. The main difference between these two measures is that for congruent trials the direction of the arrow and the spatial location are congruent, which should result in facilitation, therefore the Simon effect is a measure of the difference between facilitation on congruent trials and interference on incongruent trials. On the other hand, interference suppression is a measure of participants' ability to suppress interfering information, regardless of whether it is facilitatory or interfering. The specificity of the language group effect to interference suppression suggests that the simultaneous bilinguals were better able to supress the interfering spatial information overall, and that the group difference was not related to differences in facilitation effects. Given that bilinguals are required to supress interference from the non-target language while engaging in the target language, this finding suggests that even within highly proficient bilinguals who use both of their languages on a regular basis such as those examined in the present investigation, AoA seems to influence the adaptation of the control processes implicated in interference suppression. These findings are also consistent with previous research demonstrating AoA effects on executive function (Yow & Li, 2015), with early AoA being associated with smaller interference effects on a Stroop task, but not related to the other aspects of executive function that were measured in that study, including response inhibition, set shifting, and information updating and monitoring.

We also observed a significant relationship between the strength of the anticorrelation between the right dlPFC and vmPFC and interference suppression within the sequential bilingual group, with greater anticorrelation between the DMN and the task-positive attention network being related to better cognitive control. Taken together, our findings suggest that simultaneous learning is associated with superior cognitive control, but sequential bilinguals who show stronger anticorrelation also show better cognitive control performance. The significant association in the sequential bilinguals, but not in the simultaneous bilinguals, suggests that simultaneous bilinguals as a group show strong anticorrelations between the two resting state networks and robust interference suppression, whereas the strength of the association between brain connectivity and the efficiency of interference suppression are significantly related in sequential bilinguals whose cognitive control is less optimal. The specificity of this association between brain connectivity and behavioural performance in the sequential group may suggest that in sequential bilinguals, who learned their L2 following the mastery of their L1, there exists a more direct relationship between brain connectivity and cognitive control, or interference suppression. It is possible that the later learning of an L2 results in a greater association between cognitive control processes to manage the two languages and brain connectivity. It has been shown that the anticorrelation between the DMN and the task positive network increases with development from childhood to adolescence and adulthood (Chai, Ofen, Gabrieli, & Whitfield-Gabrieli, 2014), therefore it is possible that the later learning of an L2, and associated use of cognitive control processes to manage two languages, develop in association with brain connectivity. On the other hand, in simultaneous bilinguals, who learn their two languages from birth in a naturalistic environment, the cognitive control processes required to manage the two languages are engaged from birth, and brain connectivity follows a different developmental trajectory, possibly resulting in greater independence between cognitive control and resting state network connectivity. Direct comparisons with a monolingual control group or other groups varying in L2 acquisition could help clarify this possibility.

In the current investigation we compared simultaneous bilinguals to bilinguals who learned their L2 in a sequential manner after the age of 6 years and find group differences in both behaviour and rs-fMRI. However, the size of our sample and the range of AoA do not allow us to disentangle a linear relationship between AoA, brain connectivity, and cognitive control. In terms of structural brain measures, previous research has shown that AoA is related to cortical thickness such that earlier AoA is associated with thinner cortex in the left inferior frontal gyrus (IFG) and thicker cortex in the right IFG, although in this study language proficiency was not held constant (Klein et al., 2014). Thus, a further examination of the nature of the impact of AoA on brain development is a question to be addressed in future research.

An additional question that is not addressed by our data is whether L2 learning later in life (e.g., in adulthood) has positive consequences for cognitive function. That is, research has suggested that bilingualism can serve as a source of cognitive and/or neural reserve as individuals age (see Perani & Abutalebi, 2015), which may buffer against age-related cognitive change (e.g., Bialystok et al., 2014) and the onset of cognitive impairment (e.g., Craik, Bialystok, & Freedman, 2010), but it remains unknown whether these effects are contingent on AoA. Our data suggest cognitive benefits for simultaneous bilinguals compared to sequential bilinguals; but it would be of interest to assess whether positive effects of bilingualism could emerge with many years of experience using two languages in older adults who may have learned their L2 later.

Our study differs from previous research, which traditionally investigates cognitive control in bilinguals by comparing monolinguals and bilinguals. Although we did find differences between the two groups of bilinguals who varied in terms of L2 AoA, it is unclear whether this is a manifestation of the same language group differences observed when comparing monolinguals to bilinguals, or whether it is a difference that is specifically related to AoA. The inclusion of a monolingual control group in future studies would help to determine if the differences observed within groups of bilinguals who vary in AOA reflect the same or different influences of bilingualism on the brain as the differences observed between monolinguals and bilinguals.

In sum, our results suggest that there may be optimal time windows during which learning has implications for functional changes in the brain; in this particular case the learning of two languages from birth is optimal for brain organization in terms of at least one component of cognitive control. Learning two languages simultaneously from birth, as compared to learning an L2 after a native language, appears to have positive implications for brain organization in terms of intrinsic functional connectivity and cognitive control (i.e., interference suppression). We show that the relationship between the task-positive attention network and the DMN is associated with cognitive control such that greater anticorrelation between these two networks is associated with better interference suppression. Thus, this finding supports previous research demonstrating the association between executive function and the strength of the anticorrelated relationship between the attention network and the DMN (Hampson et al., 2010; Keller et al., 2015; Kelly et al., 2008). In addition, these findings highlight the utility of examining functional brain networks as an alternative to functional brain activity in isolated regions on their own. Using this method we show that the simultaneous learning of two languages is in fact what might be driving language experience related differences in cognitive control, at both the level of the brain and behaviour. These findings add to the growing body of literature demonstrating the importance of more subtle aspects of the bilingual language experience for the neural and behavioural consequences of bilingualism.

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Table 1. *Participant characteristics for simultaneous and sequential bilinguals.*

	Simultaneous † (n=11; 4 males)	Sequential (n=10; 4 males)	*p*
	Mean *(SD)*	Mean *(SD)*	
Age	23.4 *(2.9)*	25.5 *(4.1)*	.18
Education	15.5 *(1.8)*	15.5 *(2.1)*	.96
Age of L2 acquisition	0.0 *(0)*	7.4 *(1.9)*	<.01**
L1 letter fluency	40.7 *(13.9)*	38.8 *(14.4)*	.76
L2 letter fluency	33.6 *(8.8)*	25.5 *(10.6)*	.08
L1 category fluency	24.3 *(7.1)*	24.2 *(8.1)*	.98
L2 category fluency	16.9 *(3.5)*	13.6 *(4.5)*	.08
L1 sentence repetition	59.6 *(6.2)*	63.1 *(7.9)*	.30
L2 sentence repetition	49.2 *(10.5)*	39.3 *(14.6)*	.11
Matrix reasoning	12.3 *(2.6)*	12.0 *(3.1)*	.83

† Simultaneous bilinguals have two native languages, but were asked to identify an L1

L1 =native language; L2=second language

Table 2. *Peak MNI coordinates for regions showing positively and negatively correlated BOLD activity with the vmPFC seed (k=cluster size, number of voxels)*

Brain Area	x, y, z	*t*	*k*
Positive correlation			
Precuneus, cingulate, paracingulate	-40, +40, +0	38.5	28173
Right angular gyrus	+56, -60, +38	10.49	1893
Left angular gyrus	-54, -70, +38	10.07	1758
Right temporal cortex	+58, +0, -26	9.19	1553
Right posterior cerebellum	+46, -58, -40	8.18	181
Left temporal cortex	-60, -6, -26	8.03	1274
Left cerebellum	-42, -74, -38	7.57	270
Bilateral cerebellum	-10, -53, -40	7.35	290
Negative/anticorrelation			
Left dlPFC	-48, +46, +16	11.41	612
Right dlPFC	+48, +56, +14	8.77	410
Left inferior frontal gyrus	-44, +6, +14	7.73	635
Left cerebellum	-24, -72, -54	7.49	348
Left middle temporal gyrus	-56, -60, +6	6.77	169
Left superior parietal lobule	-32, -56, +58	6.71	383
Left inferior parietal lobule	-46, -32, +28	6.59	365
Right superior parietal lobule	+32, -54, +56	6.17	307
Right lateral occipital cortex	+30, -70, +22	6.07	425
Right inferior frontal gyrus	+50, +4, +4	5.66	583

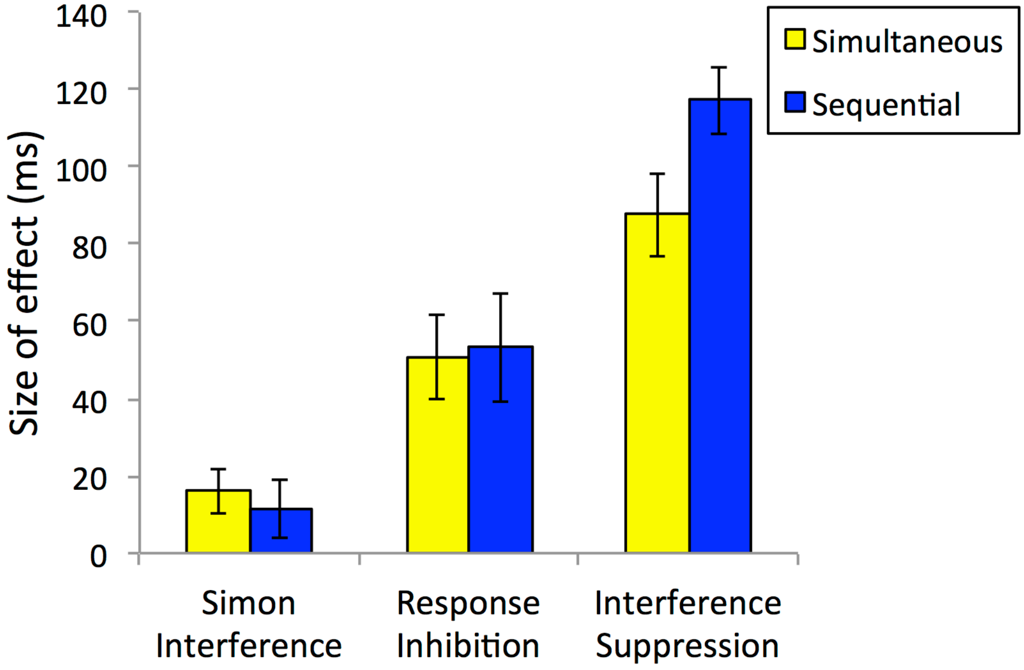

Figure 1. Behavioural data depicting Simon Interference, Response Inhibition, and Interference Suppression as a function of Language Group (error bars represent standard error of the mean).

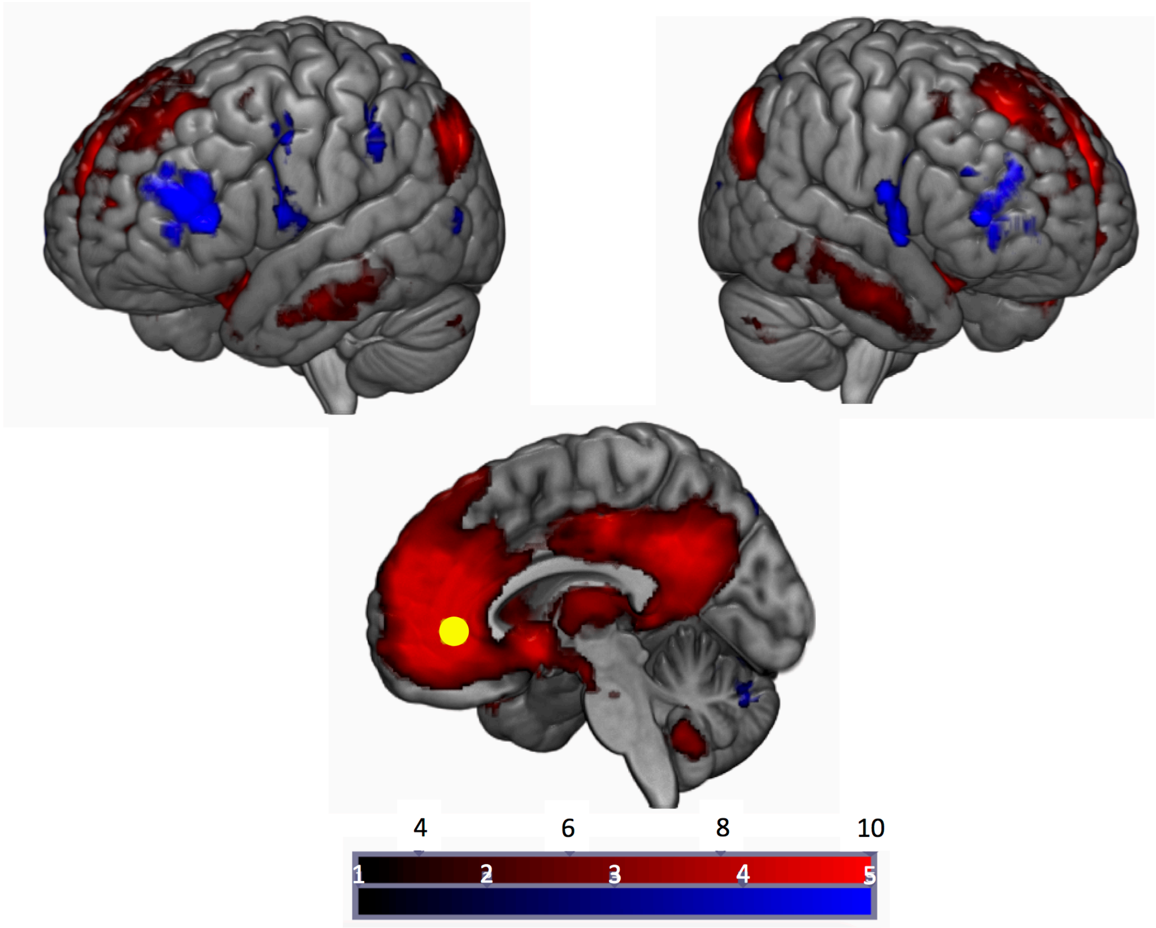

Figure 2. Brain regions showing positive (red) and negative (blue) correlation with the vmPFC seed (yellow).

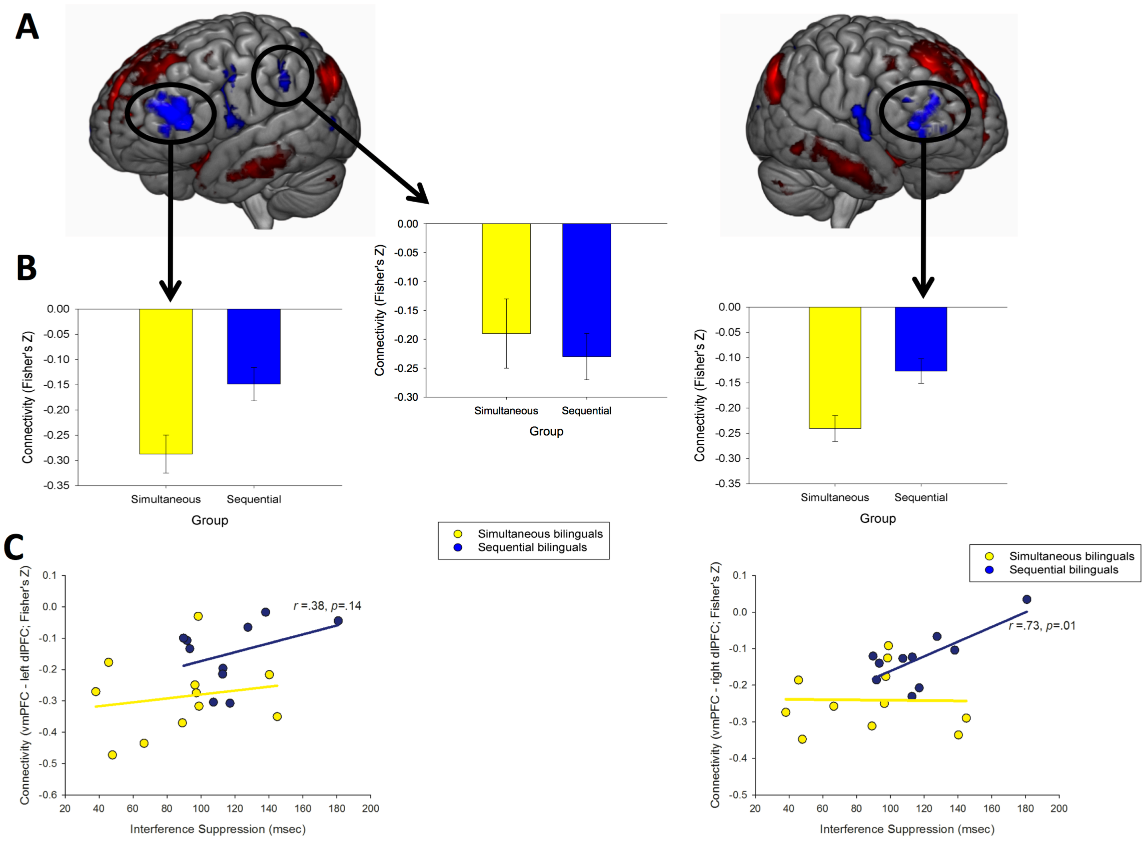

Figure 3. Panel A shows the dlPFC in each hemisphere for which the anticorrelation with the vmPFC seed differed across the language groups, and the left IPL for which the groups did not differ in terms of the anticorrelation with the dlPFC. Panel B shows the connectivity between the vmPFC and dlPFC and between the vmPFC and left IPL as a function of Language Group (error bars represent standard error of the mean). Panel C shows the relationship between interference suppression and the strength of anticorrelation between the vmPFC and the dlPFC for each group of participants (these correlations were not significant in the simultaneous bilinguals (*p*'s>.30)).