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# **Brain and Language**

# Earlier age of second language learning induces more robust speech encoding in the auditory brainstem in adults, independent of amount of language exposure during early childhood

--Manuscript Draft--

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Abstract:	Learning a second language (L2) at a young age is a driving factor of functional neuroplasticity in the auditory brainstem. To date, it remains unclear whether these effects remain stable until adulthood and to what degree the amount of exposure to the L2 in early childhood might affect their outcome. We compared three groups of adult English-French bilinguals in their ability to categorize English vowels in relation to their frequency following responses (FFR) evoked by the same vowels. At the time of testing, cognitive abilities as well as fluency in both languages were matched between the 1) simultaneous bilinguals (SIM, N=18); 2) sequential bilinguals with L1-English (N=14); and 3) sequential bilinguals with L1-French (N=11). Our results show that the L1-English group show sharper category boundaries in identification of the vowels compared to the L1-French group. Furthermore, the same pattern was reflected in the FFRs (i.e., larger FFR responses in L1-English > SIM > L1-French), while again only the difference between the L1-English and the L1-French group was statistically significant; nonetheless, there was a trend towards larger FFR in SIM compared to L1-French. Our data extends previous literature showing that exposure to a language during the first years of life induces functional neuroplasticity in the auditory brainstem that remains stable until at least young adulthood. Furthermore, the findings suggest that amount of exposure (i.e., 100% vs. 50%) to that language does not differentially shape the robustness of the perceptual abilities or the auditory brainstem encoding of phonetic categories of the language.	
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# Earlier age of second language learning induces more robust speech encoding in the auditory brainstem in adults, independent of amount of language exposure during early childhood

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Replies to the reviewers' comments

Reviewer #1: The present study examines the effects of onset exposure to English in the perceptual identification of English vowels and the early sensory encoding of these vowels with respect to vowel pitch (F0) and quality (the first formant; F1). Using the frequency following response (FFR), an auditory evoked potential that reflects phase locking activity from subcortical and cortical networks involved in early auditory processing, the authors report a more robust sensory encoding of vowel pitch and quality in English-French sequential bilinguals with English as their L1 (Eng-L1), relative to French-English sequential bilinguals with French as their L1 (Eng-L2), but not relative to simultaneous English-French bilinguals (Sim). The authors also report a trend in vowel perceptual identification patterns with a sharper perceptual identification boundary in Eng-L1, relative to Eng-L2.

With their results, the authors contribute to a better understanding of the effects of early bilingual exposure in the sensory encoding of speech properties, a topic that has been partially but not sufficiently addressed in the literature. Specifically, the authors extend previous FFR findings on bilingual pitch processing to a vowel contrast that, unlike pitch, is not shared across language competences (English and French). By doing so, they show that the early sensory encoding of speech in bilinguals is also modulated by onset exposure to language-specific structural differences.

I like the study and I think is already in good shape. However, there are few points that I would recommend to address in a another round.

<u>Author's response:</u> We thank the editor and the reviewer for their very helpful comments. We responded to each of the points raised by the reviewers below and believe that the manuscript has improved thanks to the comments of the reviewers.

(1) 2nd paragraph in the discussion section: "Our findings generally supported these hypotheses, showing sharper category boundaries as well". This reads like a strong claim for a marginal statistical significance. I think this claim would sound more convincing if there is a significant correlation between perceptual identification slopes (or perceptual boundary sharpness) and F1 sensory encoding robustness.

Author's response: We thank the reviewer for pointing out that a reader might misunderstand our writing. We believe that the statistics support the claim that there are sharper perceptual category boundaries in the L1-English group vs. the L1-French group, even though the statistical significance is marginal. The fact that we found the same result patterns in the behavioral data and in the neural data (neural response to F0 as well as F1) suggests that these results are robust. Still, we adapted the sentence according to the suggestion of the reviewer and added that the significance of these results was marginal: "Our findings generally supported these hypotheses, showing marginally sharper category boundaries as well as...:". The correlation between the perceptual identification slopes and the F1 encoding was not significant.

2. The authors report a more robust sensory encoding of F1 in Eng-L1 vs. Engl-L2. This result was obtained for both English vowel exemplars (vowels u and a) and French vowel exemplars (vowel a). I acknowledge the lack of a significant effect for the interaction of group and vowel exemplars in the ANOVA. However, given that Eng-L1 was exposed to English earlier than Eng-L2, I think it would be worth discussing why the sensory encoding enhancement of vowel quality (F1) for this group generalized to vowel exemplars that also exist in French.

<u>Author's response:</u> We thank the reviewer for pointing out this interesting thought. However, the vowel \a\ used in this study does not exist in the Canadian French vowel exemplars. The F2 of a typical Canadian French \a\ sound would be higher (1090 Hz vs. 1185 Hz). In contrast, the vowel \a\ in our study rather represents an English vowel exemplar (see Table 2 for details), similar to the other vowels used in this study.

3. Figures showing the FFRs are quite small and group differences, as well as FFR SNR-quality, are really difficult to visualize. I would recommend increasing the size of the FFRs in both the time and spectral domains.

<u>Author's response:</u> Thank you for pointing it out. We increased both axes and increased the quality of the figure. The group differences are now much more visible for the temporal and spectral domains.

4. Regarding the FFR montage, could you explain in more detail the location of each electrode? (active, ground, and reference).

<u>Author's response:</u> We thank the reviewer for reading carefully. We added the information of the location of the ground electrode, which was missing by mistake. It now reads: "Subcortical responses were recorded at an electrode placed on cervical vertebrae 7 (C7) and referenced against the right mastoid, while the grounds (CMS/DRL) were placed on the left side of the forehead next to each other."

Reviewer #2: This study compares three groups of adult English-French bilinguals on their categorization of English vowels and encoding of those vowels as measured by the frequency-following response (FFR). They found that better categorization and more robust FFR of English vowels was seen in the individuals who acquired English earlier. They also conclude that amount of exposure early in life does not differentially shape these brain and behavior effects. Overall, this is a well-written manuscript.

#### Author's response: Thank you for this helpful review.

The results are clearly stated, but the visual demonstration of the effects can be improved. Specifically, Figure 3 has a lot of text making the time and frequency domain waveforms too small and difficult to interpret. The plots need to be enlarged in the vertical direction.

<u>Author's response:</u> We thank the reviewer for pointing out how to improve Figure 3. Accordingly, we improved the quality of Figure 3. We increased the waveforms in both the time and the frequency domain and removed unnecessary text in the figure. The effects are more clearly visible now.

Additionally, given that the simultaneous bilinguals fall in-between the L1-French and L1-English groups, it's difficult to conclude that early exposure has no effect. It may be that the groups are too small to pick up on the difference. I would either tone down the exposure interpretation or collect data in additional participants to strengthen this claim.

Author's response: We thank the reviewer for this comment. Our data do not provide evidence for a difference between the L1-English and the simultaneous bilingual groups. Furthermore, even with a larger sample size, we would predict a potential difference to be very small given that the difference between the L1-English and the L1-French group is already marginal (but robust). Also, in the limitation section, we discussed potential problems with the small sample size: "Another limitation is the relatively small sample size, especially of the L1-French group. However, it remains a challenge to find bilinguals who fit all of the inclusion and exclusion criteria, even in a bilingual city such as Montréal". Still, in order to better accommodate the reviewer's concerns, we added a sentence to this paragraph stating: "Future research with larger samples as well as novel measures of L2 exposure during early childhood will be

important in the investigation of perceptual and auditory brainstem plasticity as a function of simultaneous bilingual versus monolingual exposure to language sounds."

## Minor Comments

1. Does English age of acquisition across all three groups correlate with FFR values or categorization measures?

<u>Author's response</u>: We thank the reviewer for the suggestion to compute correlations between English-AoA and FFR/categorization in addition to the group differences. However, we refrained from such analyses because the L1-English and the simultaneous group both have an English-AoA of 0 years meaning that there is no variation in this variable across 2/3 of the participants. We believe that this suggests that the results could be unreliable and would not add more information than the analyses of the group differences (even though the correlations between English-AoA and FFR of F0 and categorization reach significance in the expected direction). For the sake of completeness, we attach the results of these analyses here:

1) Significant correlation between AoA-English (x-axis) and FFR to F0 (y-axis): r = -0.42, p = .005



2) No significant correlation between AoA-English (x-axis) and FFR to F1 (y-axis): r = -0.03, p = .83



3) Significant correlation between AoA-English (x-axis) and FFR to F0 (y-axis): r= 0.31, p=.05



2. Page 8, section 2.3, second paragraph, should it be 'RTs shorter than 250 ms and longer than 1200 ms' instead of 'RTs slower than 250 ms...'

Author's response: Thank you. Done.

## Highlights

- Exposure to a language during the first years of life induces lifelong functional neuroplasticity in the auditory brainstem
- Earlier age of second language acquisition induces more robust speech encoding in the auditory brainstem in adults
- Amount of language exposure (100% or 50%) during early childhood does not differentially shape perception in adulthood
- Similarly, no evidence that amount of language exposure changes the robustness of auditory brainstem processing in adults











Table 1

Click here to access/download **Table** Table1.docx Table 2

Click here to access/download **Table** Table2.docx

## Earlier age of second language learning induces more robust speech encoding in the auditory brainstem in adults, independent of amount of language exposure during early childhood

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Total word count: 6627 (including highlights and statement of significance)

## Abstract

Learning a second language (L2) at a young age is a driving factor of functional neuroplasticity in the auditory brainstem. To date, it remains unclear whether these effects remain stable until adulthood and to what degree the amount of exposure to the L2 in early childhood might affect their outcome.

We compared three groups of adult English-French bilinguals in their ability to categorize English vowels in relation to their frequency following responses (FFR) evoked by the same vowels. At the time of testing, cognitive abilities as well as fluency in both languages were matched between the 1) simultaneous bilinguals (SIM, N=18); 2) sequential bilinguals with L1-English (N=14); and 3) sequential bilinguals with L1-French (N=11).

Our results show that the L1-English group show sharper category boundaries in identification of the vowels compared to the L1-French group. Furthermore, the same pattern was reflected in the FFRs (i.e., larger FFR responses in L1-English > SIM > L1-French), while again only the difference between the L1-English and the L1-French group was statistically significant; nonetheless, there was a trend towards larger FFR in SIM compared to L1-French.

Our data extends previous literature showing that exposure to a language during the first years of life induces functional neuroplasticity in the auditory brainstem that remains stable until at least young adulthood. Furthermore, the findings suggest that amount of exposure (i.e., 100% vs. 50%) to that language does not differentially shape the robustness of the perceptual abilities or the auditory brainstem encoding of phonetic categories of the language.

**Words**: 250

Key Words: auditory brainstem responses, sequential bilinguals, simultaneous bilinguals, vowels,

phonetic processing, speech processing, second language learning, age of language acquisition

## Highlights

- Exposure to a language during the first years of life induces lifelong functional neuroplasticity in the auditory brainstem
- Earlier age of second language acquisition induces more robust speech encoding in the auditory brainstem in adults
- Amount of language exposure (100% or 50%) during early childhood does not differentially shape perception in adulthood
- Similarly, no evidence that amount of language exposure changes the robustness of auditory brainstem processing in adults

## **Statement of significance**

Previous studies have indicated that early age of second language (L2) acquisition induces functional neuroplasticity in the auditory brainstem during processing of the L2. This study compared three groups of adult bilinguals who differed in their age of L2 acquisition as well as the amount of exposure to the L2 during early childhood. We demonstrate for the first time that the neuroplastic effect in the brainstem remains stable until young adulthood and that the amount of L2 exposure (i.e., 50 vs. 100%) does not influence behavioral or brainstem plasticity. Our study provides novel insights into low-level auditory plasticity as a function of varying bilingual experience.

## 1. Introduction

Previous research on bilinguals has suggested that the enriched linguistic experience, as well as the increased demands of controlling multiple languages, alters high-level processes such as executive control and attentional control (for a review see Bialystok et al., 2012), as well as low-level perceptual encoding of sound in the brainstem and the cortex (for a review see Hayakawa & Marian, 2019). For example, studies have indicated that individuals who were exposed to two languages from early childhood show more robust and stable brainstem responses, as measured by the frequency following response (FFR), to simple speech sounds compared to monolinguals (Krizman et al., 2012, 2014; Skoe et al., 2017). Typically, the observed FFRs are expressed in stronger neural encoding of the fundamental frequency (F0) of the speech sound and lower inter-trial variability (Krizman et al., 2012, 2014). Both parameters have been linked to language skills such that larger FFR amplitudes have been associated with better or faster speech-in-noise perception (Kraus et al., 2000; Yellamsetty & Bidelman, 2019) and better higher-order language abilities (Hornickel & Kraus, 2013). Thus, enhanced processing in the auditory brainstem may provide a platform for higher-order auditory processes in the cortex. This suggests that there is long-term learning-induced neuroplasticity in bilinguals that may build up over many years of bilingual experience. In sum, there is substantial evidence for functional neuroplasticity in the auditory brainstem that shapes the automatic encoding of speech sounds in bilinguals.

However, despite the growing evidence of the functional consequences of bilingual exposure, many questions about language development in a multilingual world remain unanswered, especially if they go beyond a bilingual versus monolingual comparison. It has been argued that research should emphasize bilingual variability in order to address the diverse language experience among bi- and multilinguals (Baum & Titone, 2014). In the present study, we adopted a different approach and compared different groups of bilinguals on whether the effects of the amount of L2 exposure in early childhood influences auditory processing in the auditory brainstem. In a study of auditory brainstem speech encoding in bilinguals who were exposed to two languages from birth, and sequential bilinguals, who learned their second language at around 4 years of age, Krizman and colleagues (2015) found larger F0-related FFR amplitudes to the synthesized syllables /ba/ and /ga/ and lower inter-trial variability to /ba/ in the simultaneous compared to the sequential group. Furthermore, the years of bilingual experience correlated positively with the magnitude of the two FFR parameters (Krizman et al., 2015). Thus, the observed neural enhancements in the auditory brainstem increased with longer bilingual experience, or, in this case, with lower age of acquisition (AoA) of the L2. Overall, these findings confirm that L2-AoA is one important factor contributing not only to individual differences in the bilingual experience, but also to variability in brainstem neuroplasticity as a function of the enriched exposure to sounds.

5

Nonetheless, it remains a puzzling question how the auditory brainstem becomes more sensitive to speech sounds with greater bilingual experience, while it goes hand in hand with getting less exposure to the sounds of each language system (i.e., the "native" language L1 and the L2) compared to monolinguals (assuming that bilingual parents speak a similar amount of time with their children as monolingual parents (Costa & Sebastián-Gallés, 2014)). Previous research has mainly used languageneutral speech stimuli (Krizman et al., 2012, 2014, 2015; Skoe et al., 2017) that do not permit disentanglement of the effects of bilingualism on the subcortical neural processing of a distinct language such as the L1 or the L2 specifically. Thus, in order to investigate the degree to which the amount of exposure to one language --in the present study English-- induces neuroplastic effects in the auditory brainstem, we compared FFRs to representative English sounds across three groups of bilinguals differing in their amount of English-exposure during early childhood. The three bilingual groups were: 1) simultaneous bilinguals who were presumably exposed to English approximately 50% of the time during early childhood; 2) sequential bilinguals with L1-English and L2-French, being exposed only to their L2 (French) for the first time between 2 and 6 years of age (L1-English, N=14) and 3) sequential bilinguals with L1-French and L2-English, who were exposed to their L2 (English) for the first time between 2 and 6 years of age (L1-French, N=11).

In addition to the passive recording of the FFR responses to typical English vowels, participants also performed a vowel identification task using the same speech stimuli used in the FFR task to determine the perceived sharpness of categorical boundaries (Bidelman et al., 2014). Speech sounds such as vowels or syllables, which have been used in FFR recordings (Bidelman et al., 2014; Krizman et al., 2012, 2014, 2015; Skoe et al., 2017), are typically perceived categorically in a categorization task, meaning that they are perceived as belonging to a distinct phonetic category (Bidelman et al., 2014). Such categorical perception has been shown to be influenced by language experience with sharper functions associated with more language exposure (P. Kuhl et al., 1992), suggesting that it is prone to learning-induced plasticity similar to the auditory brainstem responses.

Thus, we investigated to what degree AoA, as well as the amount of language experience during early childhood, shape neuroplastic changes in the auditory brainstem. If AoA is a driving factor of long-term neural plasticity in the auditory brainstem, it can be expected that earlier English-AoA leads to more robust neural encoding of the F0 and the first formant (F1) (i.e., larger neural responses) as well as sharper category boundaries for English speech sounds. Thus, we expected to find a response pattern showing more robust responses in simultaneous bilinguals and L1-English sequential bilinguals compared to L1-French sequential bilinguals. Furthermore, if the amount of exposure to English during the first years of life is also reflected in long-term subcortical auditory plasticity, we also expected to find more robust neural responses and sharper category boundaries for English sounds

in those with more exposure to English during early childhood (i.e., L1-English sequential bilinguals > simultaneous bilinguals > L1-French sequential bilinguals). Thus, based on the results we find, we can, for the first time, disentangle the effects of AoA and quantitative exposure to a language to assess the degree to which each of them shapes the functioning of the auditory brainstem in adulthood.

## 2. Materials and Methods

## 2.1. Participants

For this study, Canadian English – French bilinguals with an age ranging from 18 to 36 years were recruited, including 18 simultaneous bilinguals (SIM), 14 early bilinguals with English as their L1 (L1-English), and 11 early bilinguals with Canadian French as their L1 (L1-French). Simultaneous bilinguals were defined as having learned English and French from birth. Early bilinguals had an age of L2 acquisition (AoA) between 2 and 6 years. Participants had no functional knowledge of a third language. As shown in Table 1, the three groups did not differ in chronological age, gender, or cognitive abilities (i.e., verbal working memory and nonverbal inhibition). Furthermore, the groups did not differ in fluency of English and French at the time of testing, which was based on an average score resulting from a phonemic (English letters: F, A, and S; French letters: P, F, and L) and a semantic (English: animals; French: fruits) fluency task. However, sequential bilinguals (but not simultaneous bilinguals) performed worse in a sentence repetition task when administered in their L2 relative to their L1, suggesting that, at the time of testing, participants were more proficient in their L1 than in their L2, while simultaneous bilinguals had equal proficiency in both languages (see Table 1).

----- insert Table 1 about here -----

All participants were healthy, right-handed young adults with normal hearing as assessed by pure-tone average (PTA) thresholds at 500, 1000, 2000, and 4000 Hz (< 20 dB HL). Professional musicians were excluded from participating in this study. Participants gave written informed consent and were given monetary compensation for their participation.

## 2.2. Stimulus material

For this study, an English vowel continuum was used, which has been described in previous studies (Bidelman et al., 2014; Bidelman & Alain, 2015). The steady-state stimuli were synthesized along a continuum from / $\sigma$ / to /a/, varying in their F1 between 430-730 Hz, while all other parameters were kept constant (F0 = 100 Hz, F2 = 1090 Hz, F3 = 2350 Hz). From the continuum, 5 vowels varying in equal acoustic steps were extracted with a duration of 100ms each. The spectrograms of the 5 stimuli (Vowel 1 to Vowel 5) are shown in Figure 1. Perceptual identification of such speech sounds tends to be categorical such that the morphed vowels are perceived as belonging to discrete phonetic categories when using an identification task (Liberman et al., 1967); in this example, the vowels were identified as either 'u' or the vowel category 'a', while one of the middle stimuli was ambiguous

between the two categories, yielding inconsistent category judgments (Bidelman et al., 2014; Bidelman & Alain, 2015).

## ----- insert Figure 1 about here -----

In order to assess to what degree the stimuli were typical examples of English vowels and were acoustically different from Canadian French vowels, we compared the F1 and F2 of the two vowels at the ends of the continuum (i.e., the / $\upsilon$ / and the /a/ or Vowel 1 and Vowel 5) to reference parameters reported in the literature. As can be seen in Table 2, the English reference values recorded from male speakers (Peterson & Barney, 1952) are very similar to the stimulus material we have used, while several differences could be established from vowels in Canadian French: (1) The / $\upsilon$ / does not have phonemic status in Canadian French, (2) the F1 and F2 of the /u/ acoustically close to / $\upsilon$ / are lower in Canadian French than in English and therefore more different from the / $\upsilon$ / than is the English /u/, and (3) the F2 of the /a/ is higher in Canadian French than in English (Arnaud et al., 2018). In sum, Vowels 1 and 5, the two vowels at the ends of the continuum, reflect typical English vowels, while they are distinguishable from Canadian French vowels in their acoustic properties.

----- insert Table 2 about here -----

## 2.3. Behavioral task

In a behavioral task, each of the five vowels was presented 40 times in random order at an intensity of 67 dBA via EARLINK tube ear inserts (Neuroscan, El Paso, TX, USA). Participants performed a forced choice categorization task while listening to the vowels and were asked to categorize each stimulus into either category 'u' or 'a' as quickly as possible, by pressing the left or right arrow, respectively, on the keyboard. The inter-stimulus interval (ISI) was set to 500 ms.

We extracted reaction times (RTs) from each listener's identification responses across the 40 trials for each vowel. RTs shorter than 250 ms and longer than 1200 ms were excluded from the analysis. Furthermore, we extracted the slope of the individual vowel categorization scores in order to compare the steepness of the category boundary between the two vowel categories ('u' and 'a') between the groups. To compute the slope, we fitted a logistic function to the individual data using the quickpsy package (Linares & Lopez-Moliner, 2016) running in R version 3.4.0 (https://www.R-project.org/).

## 2.4. Auditory brainstem responses recording

For each vowel, frequency following responses (FFRs) were recorded in a separate block with 2000 trials each, with an ISI of 150 ms (Bidelman et al., 2014). The stimuli were presented at an intensity of 86 dB SPL through the same EARLINK insert earphones as for the behavioral task. Participants were instructed to ignore the vowels and stay in a wakeful and calm state for the recording. Subcortical responses were recorded at an electrode placed on cervical vertebrae 7 (C7) and referenced against the right mastoid, while the grounds (CMS/DRL) were placed on the left side of the forehead next to each other. Electrode impedance was kept below  $\leq 5 \text{ k}\Omega$  and was digitized using a sampling rate of 16,384

Hz (Biosemi running under "Active two"). Offline, data was pre-processed using EEGLAB (v14.1.1) running in Matlab 2017a. The data was filtered with a bandpass filter of 80-2500 Hz (Bidelman et al., 2013, 2014) and epochs of 140 ms phase-locked to stimulus onset were generated for each polarity and baseline corrected with respect to a 40 ms pre-stimulus baseline. The epoched data was further detrended and artifacts > 40  $\mu$ V were automatically removed. Epochs were then averaged across each polarity for each vowel. In keeping with Bidelman et al. (2014), we further computed the following analysis steps: 1) For visual inspection, the FFR responses for each vowel and for each of the three groups were plotted; 2) To assess the peak amplitudes of the harmonics of the brainstem responses, fast Fourier transforms (FFTs) were calculated in the time window of 0-100 ms of each epoch between 0-1000 Hz for each group and vowel; 3) For each of those FFTs, the neural sensitivity to the F0 and the F1 of the stimuli was estimated as follows: For the F0, the peak amplitude of the spectral neural response at 100 Hz (i.e., the voice pitch corresponding to the F0 of our vowels) was quantified for each FFT. Furthermore, to quantify the neural sensitivity to the varying voice timbre of the vowels (i.e., the varying F1 across the five vowels) in the brainstem spectra, the amplitudes of the spectral envelope were estimated using the envelope function in Matlab in the relevant frequency range between 400 and 750 Hz (i.e., the F1 range of our stimuli) (Bidelman et al., 2013, 2014).

## 2.5. Statistical analyses

All dependent variables of this study (i.e., reaction times for categorization task, brainstem responses to F0, and brainstem responses to F1) were analyzed using repeated measures ANOVAs to compare the three groups (3 levels: SIM, L1-English, L1-French) and stimuli (5 levels: Vowels 1-5) using R version 3.4.0. The slope of the categorization responses to the behavioral task was compared across the three groups with a one-way ANOVA. For all analyses, an alpha level of  $\alpha = .05$  was utilized unless otherwise indicated. Post-hoc tests were corrected for multiple comparisons via Tukey's test and effect sizes are indicated using  $\eta^2$ .

## 3. Results

## 3.1. Behavioral responses to categorization task

Behavioral responses to the categorization task and the fitted logistic functions to the vowel identification are shown in Figure 2. The functions indicate relatively consistent perceptual identification for Vowels 1 and 2 (as /u/), as well as Vowels 4 and 5 (as /a/), with Vowel 3 yielding inconsistent identification, reflecting the category boundary. After careful inspection of the raw data, we excluded the slope of one L1-English participant from statistical analyses because the slope was more than 5 SD smaller than the mean. Excluding that outlier, the slopes of the vowel identification functions (SIM: M=-2.2, SD=.61, L1-English: M=-2.73, SD=.98, L1-French: M=-1.88, SD=.97) showed a trend (F(2,37)=3.08, p=.058,  $\eta$ <sup>2</sup>=.14) towards a group difference between the L1-English and

the L1-French groups (p=.05). Thus, the group comparison indicates that individual differences in the age of first exposure to English shapes the category boundaries of typical English vowels even in adulthood. In other words, those participants who had extensive exposure to English in their first few years of life exhibited somewhat sharper category boundaries for these English vowels than those who started to learn English later.

----- insert Figure 2 about here -----

The analysis of the reaction times did not reveal differences across groups (F(2,190)=.73, p=.48,  $\eta^2$ =.01), suggesting that all groups categorized the vowels with the same speed. There was a main effect of vowel (F(4,190)=7.34, p<.001,  $\eta^2$ =.13), demonstrating, not surprisingly, that categorization was slower for Vowel 3 (the ambiguous stimulus) than for all other vowels (all p's <.08) due to the uncertainty associated with its identification. There was no group x vowel interaction (F(8,190)=.27, p=.98,  $\eta^2$ =.01).

## 3.2. Brainstem data

For visual inspection, Figure 3 shows brainstem responses in both time and spectral domains. Statistical analysis of the neural response to the F0 of the stimuli (i.e., the amplitude of the peak of the spectral neural response at 100 Hz) revealed a main effect of group (F(2,200)=6.34, p=.002,  $\eta^2$ =.06), as illustrated in Figure 4A. Post-hoc t-tests further suggested that the neural response to the F0 was lower in the L1-French group compared to the L1-English group (p=.001), with a trend toward a difference relative to the simultaneous group (p=.06). No main effect of vowel (F(4,200)=1.87, p=.12,  $\eta^2$ =.03) and no group x vowel interaction (F(8,200)=1.04, p=.40,  $\eta^2$ =.04) was found. The repeated measures ANOVA for the neural responses to the F1 of the stimuli (i.e., the mean envelope of the spectral neural response between 400 and 750 Hz) revealed similar effects, namely a significant main effect of group (F(2,200)=3.15, p=.04,  $\eta^2$ =.03), but no main effect of vowel (F(4,200)=.77, p=.55,  $\eta^2$ =.01) and no group x vowel interaction (F(8,200)=.32, p=.96,  $\eta^2$ =.01). For the brainstem response to F1, the response was lower in the L1-French group compared to the L1-English group (p=.03) (see Figure 4B).

----- insert Figure 3 about here -----

----- insert Figure 4 about here -----

Because the bilingual groups differed in their English language proficiency (based on the English sentence repetition task), we also investigated whether the group differences in the brainstem responses could be explained by participants' English proficiency at the time of testing rather than by their early language experience alone. We computed two Pearson's correlations between the English sentence repetition performance and the 1) neural response to the F0 of the stimuli, averaged across the neural responses elicited by all five vowels, because there were no significant differences between them) and the 2) neural response to the F1 of the stimuli, also averaged for brainstem responses to all

five vowels. No significant effects were found, either for the F0 responses (r=.07, p=.66) or for the F1 responses (r=-.06, p=.71), suggesting that English proficiency at the time of testing does not explain the variance in the brainstem responses to the English vowels. We interpret this finding to indicate that early childhood exposure to English during the first few years of life shapes the brainstem representations of sounds in the English language, with effects lasting until adulthood.

## 4. Discussion

Our study was the first to address the extent to which AoA and the amount of exposure to a specific language (here English) during early childhood shapes long-term neuroplasticity in the auditory brainstem. In order to investigate this issue, we went beyond the traditional bilingual versus monolingual comparison and instead focused on differences among three bilingual groups. We examined three groups of bilinguals with different AoA of English as well as different quantitative experience with English during early childhood and compared their FFRs to English vowels as well as perceptual identification of the same vowels in adulthood. We hypothesized that we would find results supporting an association between early AoA and larger FFR amplitudes as well as sharper perceptual boundaries (i.e., larger FFR amplitudes and sharper perceptual boundaries in SIM and L1-English as compared to L1-French). Furthermore, we also expected larger FFR amplitudes and sharper category boundaries in L1-English compared to SIM, which would suggest that not only the AoA, but also the amount of exposure to English during very early childhood (presumably about 50% in SIM versus 100% for L1-English) changes the way the brainstem processes sounds of English.

Our findings generally supported these hypotheses, showing marginally sharper category boundaries as well as larger FFR amplitudes evoked by the F0 as well as the F1 of the English vowels in the L1-English compared to the L1-French group. Furthermore, there was a trend towards larger FFR amplitudes evoked by the F0 of the English vowels in SIM compared to L1-French participants. Thus, overall, the findings support the hypothesis that earlier AoA leads to stronger neuroplasticity in the auditory brainstem and improved perception in the early-acquired language. Notably, these effects were stable even though we controlled for verbal fluency and cognitive abilities at the time of testing during adulthood, while some differences in sentence repetition remained. However, our data did not support the hypothesis that more extensive language exposure in early childhood leads to more robust processing of that language in adulthood, as reflected in the brainstem evoked responses, as we did not find a difference between the SIM and L1-English groups, who differed primarily in terms of amount of exposure to English during the first five years of life (along with exposure to an L2, of course).

Thus, analogous to Krizman et al. (2015), who investigated bilingual children, our findings demonstrate that earlier AoA leads to more robust neural responses in the auditory brainstem to sounds in the respective language, lasting until at least early adulthood. Similarly, neuroimaging studies using resting-state fMRI have shown that earlier L2-AoA leads to stronger functional

connectivity within and between auditory-related networks involved in language processing (Liu et al., 2017). Also, structural MRI studies have shown effects in gray matter plasticity as a function of L2-AoA (Grogan et al., 2012; Klein et al., 2014; Mechelli et al., 2004; for an overview see Li et al., 2014). Notably, some of the results regarding gray matter plasticity as a function of L2-AoA suggest different neuroplastic effects than the functional effects reported in the brainstem. For example, the study by Klein et al. (2014) demonstrated that cortical thickness was greater in the left interior frontal gyrus (IFG) and thinner in the right IFG in adults who had a later L2 acquisition compared to those with early L2-AoA (i.e., 8-13 years vs. 4-7 years of age). These results might reflect the greater difficulty of mastering an L2 in late learners, who also had lower L2-proficiency, compared to early learners. Thus, AoA effects might be manifested differently in low-level acoustic processing as compared to higher-level processes such as executive control or attentional control associated with learning a second language. Furthermore, there seems to be a discrepancy in studies investigating functional compared to structural neuroplasticity as discussed above. Future research should address this issue by using multimodal neuroimaging methods.

With regard to low-level phonetic learning, which we investigated in this study, there is substantial evidence for a "sensitive period", a critical time window during early childhood in which phonetic learning is boosted more strongly than during other times across the lifespan (for a review see P. K. Kuhl, 2010). Before this period is over, infants across the world show similar phonetic perception regardless of the language environment to which they are exposed. They are able to discriminate phonetic contrasts regardless of their auditory experience with specific languages (P. Kuhl et al., 1992). Then, by approximately 6 months of age, infants' perception of phonetic cues starts to alter depending on the specific languages to which they are exposed (P. Kuhl et al., 1992). Between the ages of approximately 6 and 12 months, studies show a decline in the ability to discriminate nonnative phonetic contrasts such as American English /r-l/ in Japanese infants, while there is an increase in perception of these contrasts in native American English babies (P. K. Kuhl et al., 2006). Thus, during this sensitive period for phonetic learning over the second half of the first year of life, infants' brains start to commit to their native phonetic properties. Interestingly, these effects of linguistic experience on phonetic perception in infants have also been shown to be reflected in the auditory brainstem, namely in earlier latencies of onset peaks evoked by phonetic contrasts in native monolingual listeners as compared to non-native listeners (Zhao & Kuhl, 2018). This research, in line with our results, suggests that it is important to be exposed to a language during the phonetic sensitive period for robust and automatic bottom-up encoding in the brainstem of the respective language. Our data extends previous research by showing that such early exposure to phonetic contrasts of a language shapes the representation of these contrasts in the auditory brainstem not only during childhood but lasting until at least young adulthood.

More recent research has further investigated the sensitive period hypothesis in bilingual children. For example, Bosch and Sebastiàn-Gallés (2003) have shown that the change from language-universal to language-specific phonetic processing might be different in bilinguals compared to monolinguals. They compared 4-month- and 8-month-old Spanish and Catalan monolinguals as well as Spanish-Catalan bilinguals on Catalan /e/-/E/ vowel contrast discrimination. At 4 months, all three groups of infants were able to discriminate the phonetic contrasts, while at 8 months, only the Catalan monolinguals were able to perform the task. Thus, the simultaneous bilinguals were not performing like the monolinguals in discriminating phonetic contrasts of one of their native languages, suggesting that, unlike in our study, the amount of exposure to the specific phonetic contrast might shape the ability to discriminate them at that young age. However, in a second experiment, they also compared similar monolinguals and bilingual infants at 12 months of age and found that the bilinguals were now able to discriminate the phonetic contrast in a manner similar to monolinguals. These data suggest that simultaneous bilinguals show a distinct developmental pattern of perceptual reorganization compared to monolinguals across the first year of life, while they converge at approximately 12 months of age. Another study using magnetoencephalography (MEG) found similar patterns, but also extended previous findings (Ferjan Ramírez et al., 2017). The study demonstrated that 11-month-old monolinguals were sensitive to their native language, while simultaneous bilinguals were sensitive to both of their native languages (Ferjan Ramírez et al., 2017). Furthermore, they also showed that the MEG signals reflecting the transition from acoustic to phonetic sound processing in the brain were slower in the bilinguals compared to the monolinguals, potentially because of the increased variability of the sounds in their environment. However, even though some of the auditory processing was slower in bilingual infants, their sensitivity to phonetic contrasts non-native to either language was higher at 10-12 months compared to monolinguals (Petitto et al., 2012).

Thus, similar to our results, after approximately 12 months of age, it seems that the amount of exposure to a language during the sensitive period does not matter in terms of phonetic perceptual abilities or brainstem encoding, even though the development of language-specific perceptual abilities and their neuroplastic correlates during the first year of life might be different in simultaneous bilinguals and monolinguals. In sum, our research fits well with the literature in that we do not find differences in the robustness of phonetic perception and brainstem encoding in a specific language to which exposure varied between full-time and approximately 50% during the first years of life. However, it remains to be investigated what the minimum amount of exposure is to influence behavioral and neural sensitivity to specific phonetic categories of a language. Future research should therefore examine this issue by comparing infants with large variation in their amount of exposure to a language during early childhood.

One limitation of this study is that we did not collect actual values of the amount of L2 exposure during early childhood. However, such data are usually assessed retrospectively after many years and therefore limited in their predictive nature. Still, future research should try to develop novel ways to reliably collect data on the actual amount of L2 exposure during early childhood in order to avoid speculation based on the difference between simultaneous and sequential bilingual experience, which we used as a proxy for L2 exposure in this study. Another limitation is the relatively small sample size, especially of the L1-French group. However, it remains a challenge to find bilinguals who fit all of the inclusion and exclusion criteria, even in a bilingual city such as Montréal. Future research with larger samples as well as novel measures of L2 exposure during early childhood will be important in the investigation of perceptual and auditory brainstem plasticity as a function of simultaneous bilingual exposure to language sounds.

To conclude, our study extends previous literature by showing that a) exposure to language during the first years of life leads to more robust perceptual abilities as well as encoding of the language-specific phonetic contrasts until at least young adulthood. Furthermore, we were able to show that b) the amount of exposure to the language (estimated at 100% versus 50% because of a bilingual language environment) does not lead to differences in the stability of the encoding. Overall, our data therefore speak for long-term experience-dependent neuroplasticity in perception as well as the auditory brainstem as a function of early language exposure.

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## **Conflict of interest**

The authors declare no competing financial interests.

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## **Table Descriptions**

*Table 1*: Demographics (means and standard deviations in brackets) of the three bilingual groups tested in this study. Furthermore, this table shows bilingual group differences in cognitive variables as well as in language proficiency at timepoint of testing.

*Table 2*: F1 and F2 frequency of the two vowels used in this study at the end of the continuum and F1 and F2 of reference vowels recorded from English (Peterson & Barney, 1952) and Canadian French speakers (Arnaud et al., 2018).

## **Figure Legends**

*Figure 1*: Oscillogram (top) and spectrogram (bottom) of the 5 vowels used as stimulus material in this study from  $/\upsilon/$  to /a/ from Vowel 1 to Vowel 5 (left to right). The red traces correspond to the automatic formant detection as performed by Praat. The red arrows are pointing to the F1 frequency, which was different between the 5 vowels.

*Figure 2*: Vowel identification accuracy and logistic functions for each group. A: Shows the likelihood of identification for vowel category 'u' and the fitted logistic functions separately for each group. B: Shows the group differences in the slope of the vowel categorization functions. C: Depicts the reaction time for categorization of each vowel and group. Error bars represent standard errors.

*Figure 3*: Figure 3 shows the brainstem data in the time domain (A) and in the spectral domain including the envelope of the spectral responses (B) for each vowel and group separately.

*Figure 4*: Figure 4 shows the neural responses to the F0 of the stimuli (i.e., the peak amplitude of the spectral neural response at 100 Hz) and the F1 of the stimuli (i.e., the mean envelope of the spectral neural response between 400 and 750 Hz) separately for each group. Error bars represent standard errors.

Earlier age of second language learning induces more robust speech encoding in the auditory brainstem in adults, independent of amount of language exposure during early childhood	
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#### Abstract

Learning a second language (L2) at a young age is a driving factor of functional neuroplasticity in the auditory brainstem. To date, it remains unclear whether these effects remain stable until adulthood and to what degree the amount of exposure to the L2 in early childhood might affect their outcome.

We compared three groups of adult English-French bilinguals in their ability to categorize English vowels in relation to their frequency following responses (FFR) evoked by the same vowels. At the time of testing, cognitive abilities as well as fluency in both languages were matched between the 1) simultaneous bilinguals (SIM, N=18); 2) sequential bilinguals with L1-English (N=14); and 3) sequential bilinguals with L1-French (N=11).

Our results show that the L1-English group show sharper category boundaries in identification of the vowels compared to the L1-French group. Furthermore, the same pattern was reflected in the FFRs (i.e., larger FFR responses in L1-English > SIM > L1-French), while again only the difference between the L1-English and the L1-French group was statistically significant; nonetheless, there was a trend towards larger FFR in SIM compared to L1-French.

Our data extends previous literature showing that exposure to a language during the first years of life induces functional neuroplasticity in the auditory brainstem that remains stable until at least young adulthood. Furthermore, the findings suggest that amount of exposure (i.e., 100% vs. 50%) to that language does not differentially shape the robustness of the perceptual abilities or the auditory brainstem encoding of phonetic categories of the language.

#### Words: 250

Key Words: auditory brainstem responses, sequential bilinguals, simultaneous bilinguals, vowels,

phonetic processing, speech processing, second language learning, age of language acquisition

## Highlights

- Exposure to a language during the first years of life induces lifelong functional neuroplasticity in the auditory brainstem
- Earlier age of second language acquisition induces more robust speech encoding in the auditory brainstem in adults
- Amount of <u>language</u> exposure (i.e., 100% <u>orvs.</u> 50%) to a <u>language</u> during first years of <u>lifecarly childhood</u> does not differentially shape the robustness of the perceptual abilities <u>orperception in adulthood</u>
- <u>Similarly</u>, no evidence that amount of language exposure changes the robustness of the auditory brainstem processing in <u>adulthoodadults</u>

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## Statement of significance

Previous studies have indicated that early age of second language (L2) acquisition induces functional neuroplasticity in the auditory brainstem during processing of the L2. This study compared three groups of adult bilinguals who differed in their age of L2 acquisition as well as the amount of exposure to the L2 during early childhood. We demonstrate for the first time that the neuroplastic effect in the brainstem remains stable until young adulthood and that the amount of L2 exposure (i.e., 50 vs. 100%) does not influence behavioral or brainstem plasticity. Our study provides novel insights into low-level auditory plasticity as a function of varying bilingual experience.

4

#### 1. Introduction

Previous research on bilinguals has suggested that the enriched linguistic experience, as well as the increased demands of controlling multiple languages, alters high-level processes such as executive control and attentional control (for a review, see Bialystok et al., 2012), as well as low-level perceptual encoding of sound in the brainstem and the cortex (for a review, see Hayakawa and Marian, 2019). For example, studies have indicated that individuals who were exposed to two languages from early childhood show more robust and stable brainstem responses, as measured by the frequency following response (FFR), to simple speech sounds compared to monolinguals (Krizman et al., 2012, 2014; Skoe et al., 2017). Typically, the observed FFRs are expressed in stronger neural encoding of the fundamental frequency (F0) of the speech sound and lower inter-trial variability (Krizman et al., 2012, 2014). Both parameters have been linked to language skills such that larger FFR amplitudes have been associated with better or faster speech-in-noise perception (Kraus et al., 2000; Yellamsetty & Bidelman, 2019) and better higher-order language abilities (Hornickel & Kraus, 2013). Thus, enhanced processing in the auditory brainstem may provide a platform for higher-order auditory processes in the cortex. This suggests that there is long-term learning-induced neuroplasticity in bilinguals that may build up over many years of bilingual experience. In sum, there is substantial evidence for functional neuroplasticity in the auditory brainstem that shapes the automatic encoding of speech sounds in bilinguals.

However, despite the growing evidence of the functional consequences of bilingual exposure, many questions about language development in a multilingual world remain unanswered, especially if they go beyond a bilingual versus monolingual comparison. It has been argued that research should emphasize bilingual variability in order to address the diverse language experience among bi- and multilinguals (Baum & Titone, 2014). In the present study, we adopted a different approach and compared different groups of bilinguals on whether the effects of the amount of L2 exposure in early childhood influences auditory processing in the auditory brainstem. In a study of auditory brainstem speech encoding in bilinguals who were exposed to two languages from birth, and sequential bilinguals, who learned their second language at around 4 years of age, Krizman and colleagues (2015) found larger F0-related FFR amplitudes to the synthesized syllables /ba/ and /ga/ and lower inter-trial variability to /ba/ in the simultaneous compared to the sequential group. Furthermore, the years of bilingual experience correlated positively with the magnitude of the two FFR parameters (Krizman et al., 2015). Thus, the observed neural enhancements in the auditory brainstem increased with longer bilingual experience, or, in this case, with lower age of acquisition (AoA) of the L2. Overall, these findings confirm that L2-AoA is one important factor contributing not only to

individual differences in the bilingual experience, but also to variability in brainstem neuroplasticity as a function of the enriched exposure to sounds.

Nonetheless, it remains a puzzling question how the auditory brainstem becomes more sensitive to speech sounds with greater bilingual experience, while it goes hand in hand with getting less exposure to the sounds of each language system (i.e., the "native" language L1 and the L2) compared to monolinguals (assuming that bilingual parents speak a similar amount of time with their children as monolingual parents (Costa & Sebastián-Gallés, 2014)). Previous research has mainly used languageneutral speech stimuli (Krizman et al., 2012, 2014, 2015; Skoe et al., 2017) that do not permit disentanglement of the effects of bilingualism on the subcortical neural processing of a distinct language such as the L1 or the L2 specifically. Thus, in order to investigate the degree to which the amount of exposure to one language --in the present study English-- induces neuroplastic effects in the auditory brainstem, we compared FFRs to representative English sounds across three groups of bilinguals differing in their amount of English-exposure during early childhood. The three bilingual groups were: 1) simultaneous bilinguals who were presumably exposed to English approximately 50% of the time during early childhood; 2) sequential bilinguals with L1-English and L2-French, being exposed only to their L2 (French) for the first time between 2 and 6 years of age (L1-English, N=14) and 3) sequential bilinguals with L1-French and L2-English, who were exposed to their L2 (English) for the first time between 2 and 6 years of age (L1-French, N=11).

In addition to the passive recording of the FFR responses to typical English vowels, participants also performed a vowel identification task using the same speech stimuli used in the FFR task to determine the perceived sharpness of categorical boundaries (Bidelman et al., 2014). Speech sounds such as vowels or syllables, which have been used in FFR recordings (Bidelman et al., 2014; Krizman et al., 2012, 2014, 2015; Skoe et al., 2017), are typically perceived categorically in a categorization task, meaning that they are perceived as belonging to a distinct phonetic category (Bidelman et al., 2014). Such categorical perception has been shown to be influenced by language experience with sharper functions associated with more language exposure (P. Kuhl et al., 1992), suggesting that it is prone to learning-induced plasticity similar to the auditory brainstem responses.

Thus, we investigated to what degree AoA, as well as the amount of language experience during early childhood, shape neuroplastic changes in the auditory brainstem. If AoA is a driving factor of long-term neural plasticity in the auditory brainstem, it can be expected that earlier English-AoA leads to more robust neural encoding of the F0 and the first formant (F1) (i.e., larger neural responses) as well as sharper category boundaries for English speech sounds. Thus, we expected to find a response pattern showing more robust responses in simultaneous bilinguals and L1-English sequential bilinguals compared to L1-French sequential bilinguals. Furthermore, if the amount of exposure to

English during the first years of life is also reflected in long-term subcortical auditory plasticity, we also expected to find more robust neural responses and sharper category boundaries for English sounds in those with more exposure to English during early childhood (i.e., L1-English sequential bilinguals > simultaneous bilinguals > L1-French sequential bilinguals). Thus, based on the results we find, we can, for the first time, disentangle the effects of AoA and quantitative exposure to a language to assess the degree to which each of them shapes the functioning of the auditory brainstem in adulthood.

#### 2. Materials and Methods

#### 2.1. Participants

For this study, Canadian English – French bilinguals with an age ranging from 18 to 36 years were recruited, including 18 simultaneous bilinguals (SIM), 14 early bilinguals with English as their L1 (L1-English), and 11 early bilinguals with Canadian French as their L1 (L1-French). Simultaneous bilinguals were defined as having learned English and French from birth. Early bilinguals had an age of L2 acquisition (AoA) between 2 and 6 years. Participants had no functional knowledge of a third language. As shown in Table 1, the three groups did not differ in chronological age, gender, or cognitive abilities (i.e., verbal working memory and nonverbal inhibition). Furthermore, the groups did not differ in fluency of English and French at the time of testing, which was based on an average score resulting from a phonemic (English letters: F, A, and S; French letters: P, F, and L) and a semantic (English: animals; French: fruits) fluency task. However, sequential bilinguals (but not simultaneous bilinguals) performed worse in a sentence repetition task when administered in their L2 relative to their L1, suggesting that, at the time of testing, participants were more proficient in their L1 than in their L2, while simultaneous bilinguals had equal proficiency in both languages (see Table 1).

#### ----- insert Table 1 about here -----

All participants were healthy, right-handed young adults with normal hearing as assessed by pure-tone average (PTA) thresholds at 500, 1000, 2000, and 4000 Hz (< 20 dB HL). Professional musicians were excluded from participating in this study. Participants gave written informed consent and were given monetary compensation for their participation.

#### 2.2. Stimulus material

For this study, an English vowel continuum was used, which has been described in previous studies (Bidelman et al., 2014; Bidelman & Alain, 2015). The steady-state stimuli were synthesized along a continuum from /o/ to /a/, varying in their F1 between 430-730 Hz, while all other parameters were kept constant (F0 = 100 Hz, F2 = 1090 Hz, F3 = 2350 Hz). From the continuum, 5 vowels varying in equal acoustic steps were extracted with a duration of 100ms each. The spectrograms of the 5 stimuli (Vowel 1 to Vowel 5) are shown in Figure 1. Perceptual identification of such speech sounds tends to be categorical such that the morphed vowels are perceived as belonging to discrete phonetic

categories when using an identification task (Liberman et al., 1967); in this example, the vowels were identified as either 'u' or the vowel category 'a', while one of the middle stimuli was ambiguous between the two categories, yielding inconsistent category judgments (Bidelman et al., 2014; Bidelman & Alain, 2015).

#### ----- insert Figure 1 about here -----

In order to assess to what degree the stimuli were typical examples of English vowels and were acoustically different from Canadian French vowels, we compared the F1 and F2 of the two vowels at the ends of the continuum (i.e., the / $\sigma$ / and the /a/ or Vowel 1 and Vowel 5) to reference parameters reported in the literature. As can be seen in Table 2, the English reference values recorded from male speakers (Peterson & Barney, 1952) are very similar to the stimulus material we have used, while several differences could be established from vowels in Canadian French: (1) The / $\sigma$ / does not have phonemic status in Canadian French, (2) the F1 and F2 of the / $\mu$ / acoustically close to / $\sigma$ / are lower in Canadian French than in English and therefore more different from the / $\sigma$ / than is the English / $\mu$ /, and (3) the F2 of the /a/ is higher in Canadian French than in English (Arnaud et al., 2018). In sum, Vowels 1 and 5, the two vowels at the ends of the continuum, reflect typical English vowels, while they are distinguishable from Canadian French vowels in their acoustic properties.

----- insert Table 2 about here -----

#### 2.3. Behavioral task

In a behavioral task, each of the five vowels was presented 40 times in random order at an intensity of 67 dBA via EARLINK tube ear inserts (Neuroscan, El Paso, TX, USA). Participants performed a forced choice categorization task while listening to the vowels and were asked to categorize each stimulus into either category 'u' or 'a' as quickly as possible, by pressing the left or right arrow, respectively, on the keyboard. The inter-stimulus interval (ISI) was set to 500 ms.

We extracted reaction times (RTs) from each listener's identification responses across the 40 trials for each vowel. RTs slower than 250 ms and longer than 1200 ms were excluded from the analysis. Furthermore, we extracted the slope of the individual vowel categorization scores in order to compare the steepness of the category boundary between the two vowel categories ('u' and 'a') between the groups. To compute the slope, we fitted a logistic function to the individual data using the quickpsy package (Linares & Lopez-Moliner, 2016) running in R version 3.4.0 (https://www.R-project.org/).

#### 2.4. Auditory brainstem responses recording

For each vowel, frequency following responses (FFRs) were recorded in a separate block with 2000 trials each, with an ISI of 150 ms (Bidelman et al., 2014). The stimuli were presented at an intensity of 86 dB SPL through the same EARLINK insert earphones as for the behavioral task. Participants were instructed to ignore the vowels and stay in a wakeful and calm state for the recording. Subcortical responses were recorded at an electrode placed on cervical vertebrae 7 (C7) and referenced against the 8

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right mastoid, while the grounds (CMS/DRL) were placed on the left side of the forehead next to each other. Electrode impedance was kept below  $\leq 5 \text{ k}\Omega$  and was digitized using a sampling rate of 16,384 Hz (Biosemi running under "Active two"). Offline, data was pre-processed using EEGLAB (v14.1.1) running in Matlab 2017a. The data was filtered with a bandpass filter of 80-2500 Hz (Bidelman et al., 2013, 2014) and epochs of 140 ms phase-locked to stimulus onset were generated for each polarity and baseline corrected with respect to a 40 ms pre-stimulus baseline. The epoched data was further detrended and artifacts > 40 µV were automatically removed. Epochs were then averaged across each polarity for each vowel. In keeping with Bidelman et al. (2014), we further computed the following analysis steps: 1) For visual inspection, the FFR responses for each vowel and for each of the three groups were plotted; 2) To assess the peak amplitudes of the harmonics of the brainstem responses, fast Fourier transforms (FFTs) were calculated in the time window of 0-100 ms of each epoch between 0-1000 Hz for each group and vowel; 3) For each of those FFTs, the neural sensitivity to the F0 and the F1 of the stimuli was estimated as follows: For the F0, the peak amplitude of the spectral neural response at 100 Hz (i.e., the voice pitch corresponding to the F0 of our vowels) was quantified for each FFT. Furthermore, to quantify the neural sensitivity to the varying voice timbre of the vowels (i.e., the varying F1 across the five vowels) in the brainstem spectra, the amplitudes of the spectral envelope were estimated using the envelope function in Matlab in the relevant frequency range between 400 and 750 Hz (i.e., the F1 range of our stimuli) (Bidelman et al., 2013, 2014).

#### 2.5. Statistical analyses

All dependent variables of this study (i.e., reaction times for categorization task, brainstem responses to F0, and brainstem responses to F1) were analyzed using repeated measures ANOVAs to compare the three groups (3 levels: SIM, L1-English, L1-French) and stimuli (5 levels: Vowels 1-5) using R version 3.4.0. The slope of the categorization responses to the behavioral task was compared across the three groups with a one-way ANOVA. For all analyses, an alpha level of  $\alpha = .05$  was utilized unless otherwise indicated. Post-hoc tests were corrected for multiple comparisons via Tukey's test and effect sizes are indicated using  $\eta^2$ .

#### 3. Results

#### 3.1. Behavioral responses to categorization task

Behavioral responses to the categorization task and the fitted logistic functions to the vowel identification are shown in Figure 2. The functions indicate relatively consistent perceptual identification for Vowels 1 and 2 (as /u/), as well as Vowels 4 and 5 (as /a/), with Vowel 3 yielding inconsistent identification, reflecting the category boundary. After careful inspection of the raw data, we excluded the slope of one L1-English participant from statistical analyses because the slope was more than 5 SD smaller than the mean. Excluding that outlier, the slopes of the vowel identification

functions (SIM: M=-2.2, SD=.61, L1-English: M=-2.73, SD=.98, L1-French: M=-1.88, SD=.97) showed a trend (F(2,37)=3.08, p=.058,  $\eta^2$ =.14) towards a group difference between the L1-English and the L1-French groups (p=.05). Thus, the group comparison indicates that individual differences in the age of first exposure to English shapes the category boundaries of typical English vowels even in adulthood. In other words, those participants who had extensive exposure to English in their first few years of life exhibited somewhat sharper category boundaries for these English vowels than those who started to learn English later.

#### ----- insert Figure 2 about here -----

The analysis of the reaction times did not reveal differences across groups (F(2,190)=.73, p=.48,  $\eta^2$ =.01), suggesting that all groups categorized the vowels with the same speed. There was a main effect of vowel (F(4,190)=7.34, p<.001,  $\eta^2$ =.13), demonstrating, not surprisingly, that categorization was slower for Vowel 3 (the ambiguous stimulus) than for all other vowels (all p's <.08) due to the uncertainty associated with its identification. There was no group x vowel interaction (F(8,190)=.27, p=.98,  $\eta^2$ =.01).

#### 3.2. Brainstem data

For visual inspection, Figure 3 shows brainstem responses in both time and spectral domains. Statistical analysis of the neural response to the F0 of the stimuli (i.e., the amplitude of the peak of the spectral neural response at 100 Hz) revealed a main effect of group (F(2,200)=6.34, p=.002,  $\eta^2$ =.06), as illustrated in Figure 4A. Post-hoc t-tests further suggested that the neural response to the F0 was lower in the L1-French group compared to the L1-English group (p=.001), with a trend toward a difference relative to the simultaneous group (p=.06). No main effect of vowel (F(4,200)=1.87, p=.12,  $\eta^2$ =.03) and no group x vowel interaction (F(8,200)=1.04, p=.40,  $\eta^2$ =.04) was found. The repeated measures ANOVA for the neural responses to the F1 of the stimuli (i.e., the mean envelope of the spectral neural response between 400 and 750 Hz) revealed similar effects, namely a significant main effect of group (F(2,200)=3.15, p=.04,  $\eta^2$ =.03), but no main effect of vowel (F(4,200)=.77, p=.55,  $\eta^2$ =.01) and no group x vowel interaction (F(8,200)=.32, p=.96,  $\eta^2$ =.01). For the brainstem response to F1, the response was lower in the L1-French group compared to the L1-English group (p=.03) (see Figure 4B).

- ----- insert Figure 3 about here -----
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Because the bilingual groups differed in their English language proficiency (based on the English sentence repetition task), we also investigated whether the group differences in the brainstem responses could be explained by participants' English proficiency at the time of testing rather than by their early language experience alone. We computed two Pearson's correlations between the English sentence repetition performance and the 1) neural response to the F0 of the stimuli, averaged across

the neural responses elicited by all five vowels, because there were no significant differences between them) and the 2) neural response to the F1 of the stimuli, also averaged for brainstem responses to all five vowels. No significant effects were found, either for the F0 responses (r=.07, p=.66) or for the F1 responses (r=-.06, p=.71), suggesting that English proficiency at the time of testing does not explain the variance in the brainstem responses to the English vowels. We interpret this finding to indicate that early childhood exposure to English during the first few years of life shapes the brainstem representations of sounds in the English language, with effects lasting until adulthood.

#### 4. Discussion

Our study was the first to address the extent to which AoA and the amount of exposure to a specific language (here English) during early childhood shapes long-term neuroplasticity in the auditory brainstem. In order to investigate this issue, we went beyond the traditional bilingual versus monolingual comparison and instead focused on differences among three bilingual groups. We examined three groups of bilinguals with different AoA of English as well as different quantitative experience with English during early childhood and compared their FFRs to English vowels as well as perceptual identification of the same vowels in adulthood. We hypothesized that we would find results supporting an association between early AoA and larger FFR amplitudes as well as sharper perceptual boundaries (i.e., larger FFR amplitudes and sharper perceptual boundaries in SIM and L1-English as compared to L1-French). Furthermore, we also expected larger FFR amplitudes and sharper category boundaries in L1-English compared to SIM, which would suggest that not only the AoA, but also the amount of exposure to English during very early childhood (presumably about 50% in SIM versus 100% for L1-English) changes the way the brainstem processes sounds of English.

Our findings generally supported these hypotheses, showing marginally sharper category boundaries as well as larger FFR amplitudes evoked by the F0 as well as the F1 of the English vowels in the L1-English compared to the L1-French group. Furthermore, there was a trend towards larger FFR amplitudes evoked by the F0 of the English vowels in SIM compared to L1-French participants. Thus, overall, the findings support the hypothesis that earlier AoA leads to stronger neuroplasticity in the auditory brainstem and improved perception in the early-acquired language. Notably, these effects were stable even though we controlled for verbal fluency and cognitive abilities at the time of testing during adulthood, while some differences in sentence repetition remained. However, our data did not support the hypothesis that more extensive language exposure in early childhood leads to more robust processing of that language in adulthood, as reflected in the brainstem evoked responses, as we did not find a difference between the SIM and L1-English groups, who differed primarily in terms of amount of exposure to English during the first five years of life (along with exposure to an L2, of course).

Thus, analogous to Krizman et al. (2015), who investigated bilingual children, our findings demonstrate that earlier AoA leads to more robust neural responses in the auditory brainstem to

sounds in the respective language, lasting until at least early adulthood. Similarly, neuroimaging studies using resting-state fMRI have shown that earlier L2-AoA leads to stronger functional connectivity within and between auditory-related networks involved in language processing (Liu et al., 2017). Also, structural MRI studies have shown effects in gray matter plasticity as a function of L2-AoA (Grogan et al., 2012; Klein et al., 2014; Mechelli et al., 2004; for an overview see Li et al., 2014). Notably, some of the results regarding gray matter plasticity as a function of L2-AoA suggest different neuroplastic effects than the functional effects reported in the brainstem. For example, the study by Klein et al. (2014) demonstrated that cortical thickness was greater in the left interior frontal gyrus (IFG) and thinner in the right IFG in adults who had a later L2 acquisition compared to those with early L2-AoA (i.e., 8-13 years vs. 4-7 years of age). These results might reflect the greater difficulty of mastering an L2 in late learners, who also had lower L2-proficiency, compared to early learners. Thus, AoA effects might be manifested differently in low-level acoustic processing as compared to higher-level processes such as executive control or attentional control associated with learning a second language. Furthermore, there seems to be a discrepancy in studies investigating functional compared to structural neuroplasticity as discussed above. Future research should address this issue by using multimodal neuroimaging methods.

With regard to low-level phonetic learning, which we investigated in this study, there is substantial evidence for a "sensitive period", a critical time window during early childhood in which phonetic learning is boosted more strongly than during other times across the lifespan (for a review see P. K. Kuhl, 2010). Before this period is over, infants across the world show similar phonetic perception regardless of the language environment to which they are exposed. They are able to discriminate phonetic contrasts regardless of their auditory experience with specific languages (P. Kuhl et al., 1992). Then, by approximately 6 months of age, infants' perception of phonetic cues starts to alter depending on the specific languages to which they are exposed (P. Kuhl et al., 1992). Between the ages of approximately 6 and 12 months, studies show a decline in the ability to discriminate nonnative phonetic contrasts such as American English /r-l/ in Japanese infants, while there is an increase in perception of these contrasts in native American English babies (P. K. Kuhl et al., 2006). Thus, during this sensitive period for phonetic learning over the second half of the first year of life, infants' brains start to commit to their native phonetic properties. Interestingly, these effects of linguistic experience on phonetic perception in infants have also been shown to be reflected in the auditory brainstem, namely in earlier latencies of onset peaks evoked by phonetic contrasts in native monolingual listeners as compared to non-native listeners (Zhao & Kuhl, 2018). This research, in line with our results, suggests that it is important to be exposed to a language during the phonetic sensitive period for robust and automatic bottom-up encoding in the brainstem of the respective language. Our data extends previous research by showing that such early exposure to phonetic contrasts of a

language shapes the representation of these contrasts in the auditory brainstem not only during childhood but lasting until at least young adulthood.

More recent research has further investigated the sensitive period hypothesis in bilingual children. For example, Bosch and Sebastiàn-Gallés (2003) have shown that the change from language-universal to language-specific phonetic processing might be different in bilinguals compared to monolinguals. They compared 4-month- and 8-month-old Spanish and Catalan monolinguals as well as Spanish-Catalan bilinguals on Catalan /e/ - /E/ vowel contrast discrimination. At 4 months, all three groups of infants were able to discriminate the phonetic contrasts, while at 8 months, only the Catalan monolinguals were able to perform the task. Thus, the simultaneous bilinguals were not performing like the monolinguals in discriminating phonetic contrasts of one of their native languages, suggesting that, unlike in our study, the amount of exposure to the specific phonetic contrast might shape the ability to discriminate them at that young age. However, in a second experiment, they also compared similar monolinguals and bilingual infants at 12 months of age and found that the bilinguals were now able to discriminate the phonetic contrast in a manner similar to monolinguals. These data suggest that simultaneous bilinguals show a distinct developmental pattern of perceptual reorganization compared to monolinguals across the first year of life, while they converge at approximately 12 months of age. Another study using magnetoencephalography (MEG) found similar patterns, but also extended previous findings (Ferjan Ramírez et al., 2017). The study demonstrated that 11-month-old monolinguals were sensitive to their native language, while simultaneous bilinguals were sensitive to both of their native languages (Ferjan Ramírez et al., 2017). Furthermore, they also showed that the MEG signals reflecting the transition from acoustic to phonetic sound processing in the brain were slower in the bilinguals compared to the monolinguals, potentially because of the increased variability of the sounds in their environment. However, even though some of the auditory processing was slower in bilingual infants, their sensitivity to phonetic contrasts non-native to either language was higher at 10-12 months compared to monolinguals (Petitto et al., 2012).

Thus, similar to our results, after approximately 12 months of age, it seems that the amount of exposure to a language during the sensitive period does not matter in terms of phonetic perceptual abilities or brainstem encoding, even though the development of language-specific perceptual abilities and their neuroplastic correlates during the first year of life might be different in simultaneous bilinguals and monolinguals. In sum, our research fits well with the literature in that we do not find differences in the robustness of phonetic perception and brainstem encoding in a specific language to which exposure varied between full-time and approximately 50% during the first years of life. However, it remains to be investigated what the minimum amount of exposure is to influence behavioral and neural sensitivity to specific phonetic categories of a language. Future research should

therefore examine this issue by comparing infants with large variation in their amount of exposure to a language during early childhood.

One limitation of this study is that we did not collect actual values of the amount of L2 exposure during early childhood. However, such data are usually assessed retrospectively after many years and therefore limited in their predictive nature. Still, future research should try to develop novel ways to reliably collect data on the actual amount of L2 exposure during early childhood in order to avoid speculation based on the difference between simultaneous and sequential bilingual experience, which we used as a proxy for L2 exposure in this study. Another limitation is the relatively small sample size, especially of the L1-French group. However, it remains a challenge to find bilinguals who fit all of the inclusion and exclusion criteria, even in a bilingual city such as Montréal. Future research with larger samples as well as novel measures of L2 exposure during early childhood will be important in the investigation of perceptual and auditory brainstem plasticity as a function of simultaneous bilingual exposure to language sounds.

To conclude, our study extends previous literature by showing that a) exposure to language during the first years of life leads to more robust perceptual abilities as well as encoding of the language-specific phonetic contrasts until at least young adulthood. Furthermore, we were able to show that b) the amount of exposure to the language (estimated at 100% versus 50% because of a bilingual language environment) does not lead to differences in the stability of the encoding. Overall, our data therefore speak for long-term experience-dependent neuroplasticity in perception as well as the auditory brainstem as a function of early language exposure.

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#### **Conflict of interest**

The authors declare no competing financial interests.

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#### **Table Descriptions**

*Table 1*: Demographics (means and standard deviations in brackets) of the three bilingual groups tested in this study. Furthermore, this table shows bilingual group differences in cognitive variables as well as in language proficiency at timepoint of testing.

*Table 2*: F1 and F2 frequency of the two vowels used in this study at the end of the continuum and F1 and F2 of reference vowels recorded from English (Peterson & Barney, 1952) and Canadian French speakers (Arnaud et al., 2018).

#### **Figure Legends**

*Figure 1*: Oscillogram (top) and spectrogram (bottom) of the 5 vowels used as stimulus material in this study from /o/ to /a/ from Vowel 1 to Vowel 5 (left to right). The red traces correspond to the automatic formant detection as performed by Praat. The red arrows are pointing to the F1 frequency, which was different between the 5 vowels.

*Figure 2*: Vowel identification accuracy and logistic functions for each group. A: Shows the likelihood of identification for vowel category 'u' and the fitted logistic functions separately for each group. B: Shows the group differences in the slope of the vowel categorization functions. C: Depicts the reaction time for categorization of each vowel and group. Error bars represent standard errors.

*Figure 3*: Figure 3 shows the brainstem data in the time domain (A) and in the spectral domain including the envelope of the spectral responses (B) for each vowel and group separately.

*Figure 4*: Figure 4 shows the neural responses to the F0 of the stimuli (i.e., the peak amplitude of the spectral neural response at 100 Hz) and the F1 of the stimuli (i.e., the mean envelope of the spectral neural response between 400 and 750 Hz) separately for each group. Error bars represent standard errors.