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*Neural entrainment to native and non-native  
linguistic rhythms in 4 to 5 month old infants*

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*Thesis submitted in partial fulfilment of the requirements of the degree for*

*Master in Brain and Cognition*

*13<sup>th</sup> of July 2020*



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*Abstract*

During the first year of life, infants use temporal components of the speech signal to help facilitate language acquisition, but the neural mechanisms that characterize this learning process are not well understood. Based on behavioral studies of native language discrimination, we hypothesized that infants use a complex neural process to assess prosodic components of the speech signal around the 4-5 month age range, however no study has yet explored speech entrainment extensively in infancy. In typical adult populations, neural oscillatory entrainment to the speech envelope has been postulated as a means for optimal analysis of the speech signal, with a emphasis on theta playing a major role in entrainment at the syllable rate level. It is not known if infants perform similar oscillatory entrainment when listening to natural speech. Using electroencephalography (EEG), speech brain entrainment was assessed in a cohort of 6 participants, 2 infants and 4 adults, while hearing a native language and subsequently a foreign one in passive listening tasks. Defining theta as our frequency range of interest (3-8 Hz), we measured differences in the participants entrainment to native and foreign speech by calculating phase-locking values (PLV's) between EEG and speech signals. Preliminary results indicate that both adults and infants showed stronger PLVs in the theta frequency for native language, with adults showing a stronger PLV in contrast to infants. Assessing infant language acquisition further through a temporally focused neurological lens could be combined with previous behavioral studies to better explain infants' attrition of prosodic and syllable speech elements. Specifically, neural entrainment may be one mechanism that bootstraps language learning and could be essential for accurate encoding of prosodic and syllable patterns of speech during infants' critical period for language discrimination. Clearly defining such developmental trajectories is important for optimization of language learning in childhood, both receptively and expressively.

*Keywords:* neural entrainment; language discrimination; infant language acquisition; phase locking value

### *Introduction*

A critical component to assess language learning is to identify ways in which infants understand and develop speech in the first year of life. Previous research has shown that even newborns discriminate native and non-native languages through self-activation (Moon et al., 1993). Additional behavioral studies confirm that monolingual infants discriminate speech of different rhythmic classes at 4-5 months of age (Bosch & Sebastian-Galles, 1997; 2001; Nazzi, Jusczyk & Johnson, 2000). These results suggest a nuanced capability to differentiate language patterns from quite a young age. It also is feasible to assess language discrimination from a neurological perspective, which could be combined with current behavioral data to provide a robust model of the developmental trajectory of speech perception. The present research aims to fill this gap and assess infant speech brain entrainment using electroencephalogram (EEG) in passive listening tasks with young infants in the first 6 months of life.

### ***Infant Behavioral Studies of Language Discrimination***

An essential aspect for language learning is the ability to decipher, or discriminate, a language from a differing one. Different studies have shown that infants can discriminate some languages from others already at birth, although they do not have this ability with every two languages. Discrimination of distant languages is intact at birth, if the infant is familiar with one of the two languages or if the languages are quite different (Mehler, Nazzi & Bertoncini 1998; Byers-Heinlein, Burns, & Werker, 2010). For example, newborns have no difficulties in noticing that Japanese and Dutch sound different, yet they do not classify English and Dutch as different because of overlapping language characteristics (Moon, Cooper & Fifer, 1993; Mehler, Nazzi & Bertoncini 1998). (Ramus, Nespor, & Mehler, 1999) proposed that infants are sensitive to differences in linguistic rhythm, defining language rhythm as related to the syllable structure of each individual language. These studies suggest that language differences play an imperative role, but the impact of prior exposure to languages has not been well explored. One study of prenatal infants exposed to two languages conducted by (Byers-Heinlein, Burns, & Werker, 2010) showed that monolingual English and bilinguals English-Tagalog could both discriminate the two languages as newborns. In addition, similar language discrimination capacities found in newborns have been observed in other species, namely cotton top tamarin monkeys

(Ramus et al., 2000) and Long-Evans rats (Toro, Trobalon, & Sebastian-Galles, 2003). Both of these studies suggest an innate capacity for language discrimination.

Languages have previously been grouped into three rhythmic groups based on the said language's timing, e.g. grouped into syllable-timed languages (ex. Spanish), stress timed (English, Dutch) or mora-timed (Japanese) (Abercrombie, 1967).

Languages such as stress timed ones like English allow for very complex syllable structures whereas a mora timed language (Japanese) use simple syllable structures. Abercrombie's model has been modified slightly by Ramus' 1999 proposal to include syllable complexity distribution of each language.

This classification system fits nicely with the idea that early language discrimination abilities in infancy have largely been attributed to prosodic information derived from speech. Mehler, Dupoux, Nazzi, & Dehaene-Lambertz (1996) proposed a model suggesting that infants would use the prosodic representation of the speech to discriminate languages. Called the Time Intensity and Grid Representation (TIGRE) model, it proposes that infants initially use the prosodic elements of speech to compute speech representations, but as infants proceed throughout the first year of life, they will use other elements of the speech to discriminate speech in a more elegant fashion. Further, the TIGRE model outlines a timeline for infant language discrimination abilities, detailing at birth that they can discriminate languages of different rhythmic classes, but not-within class languages. Hypothetically, within class discrimination will be possible months later as their language experience has increased.

This proposal has been solidified by experimental results in a number of studies. Ramus showed that newborns can discriminate, based on rhythm only, when the phonemic information is removed from the speech signal. As evidence of the switch to within-in class discrimination, Nazzi, Jusczyk & Johnson (2000) found partial within class discrimination in 5 month English exposed monolingual infants. The said sample was able to distinguish English and Dutch, but not Dutch and German, which they attributed to the infants familiarity with their native English. Bosch & Sebastian Galles (1997) showed that Spanish and Catalan could be discriminated at 4-5 months of age, even when the speech was low pass filtered to remove the phonetic cues. In sum, infants seem to initially use rhythmic qualities of speech to discriminate language whereas more complex phonetic language information begins to become significant after the first 6 months of life.

The presented empirical findings suggest that at around 4 to 5 months of age infants recognize and segment speech but the neurological mechanisms that facilitate discrimination remain unclear. From a behavioral standpoint, monolingual infants are faster at orienting to a native language than a foreign one (Dehaene-Lambertz & Houston 1998; Bosch & Sebastian-Galles, 1997). However, at the neurological level, it remains unclear what driving forces are behind this ability. Behavioral measures cannot suffice to answer our questions about what is happening in the brain between the onset of an auditory stimulus and the infants' processing of the said stimulus. Neurological measurements using electrophysiology (EEG) can inform us about more exact temporal aspects of speech processing. Attempts to define the neural systems supporting speech perception during infancy have yielded preliminary findings that indicate emerging speech-brain entrainment. In typical adult populations, strong evidence of neural entrainment to the speech signal has been found, providing potential directions for child development language acquisition neural mechanisms.

### *Adult neural entrainment*

In adult speech perception, the brain first processes the speech into phonological units that combine into larger ones. In other words, the hierarchical nature of speech allows smaller elements to be combined into larger structures in a causal relationship. Elements at one level constrain, or modify, elements at another lower level. Recent adult neural models of speech processing exemplify the brain performing a similar hierarchy in that neural oscillations in the auditory cortex entrain to modulation patterns in the speech amplitude envelope on strikingly similar timescales. Coined "speech brain entrainment" in typically developing adult populations, the phase of the brain signal at the lower frequency bands (theta and delta) aligns with slower components of the speech envelope amplitude. (Bourguignon et al., 2013; Giraud & Poeppel, 2012; Gross et al., 2013; Peelle & Davis, 2012; Poeppel, 2003). This mechanism has been argued to mirror linguistic concepts such as parsing and segmentation of the hierarchical linguistic constructions of speech.

Theoretical models of entrainment have suggested that the asymmetric sampling in time hypothesis (AST; Poeppel, 2003), which explains the functional role of low frequency rhythms, could be linked to prosody and syllable information in the linguistic speech signal. In adult populations, speech brain entrainment occurs most

strongly in theta rhythms and is associated with syllable rate patterns of utterance. (Howard & Poeppel, 2012; Peelle, Gross, & Davis, 2013). Theta is known as a “master” oscillator, referring to theta’s hierarchical modulation of faster rate oscillators, mostly common gamma, through phase amplitude coupling. A number of current research studies have predicted that speech information integration at both slower and faster rates may occur because of low frequency bands features. This concept illustrates that the amplitudes of slow features of speech, e.g. amplitude fluctuations of syllable and prosody units may be the basis for which faster oscillatory processes rely on, such as the processing of phonemes.

As mentioned previously, some studies have analyzed low frequencies in response to speech, e.g. to the slow acoustic modulations of speech. A handful of studies examining increases in theta power in adults have found a relation between increases in theta for native but not for non-native contrasts (Pérez, Carreiras, Gillon-Dowens & Duñabeitia, 2015).

Expanding upon the theta oscillators, oscillation based models of speech perception postulating a computational principle of such decoding have recently provided evidence of an analogous role of delta at the phrasal level. (Rimmele, Poeppel, & Ghitza, 2020). Using MEG, participants showed strong delta entrainment for phrasal “chunks” of language, whereas periodicities were lacking for language chunk rates outside of the delta range. Taken altogether, theta and delta oscillators may play a significant role in syllable and phrasal level segmentation, respectively.

Although this evidence is compelling, it may be that adult and infants’ oscillatory responses are difficult to compare as adults use higher order processes for language comprehension, with speech coherence playing a role in the degree of entrainment in low frequency bands (Bourguignon et al., 2013). To begin to mitigate the gap between adult and infant entrainment research, a couple studies have been conducted in neural entrainment in children. Ríos-López and colleagues (2018) used a longitudinal design and found an increase in delta entrainment to the slow aspects of speech in 4-7 year old school age children. Following these children for three timepoints, they found delta coherence in the temporal electrodes at all timepoints and increasing coherence to speech as the children got older. An additional study (Power et al., 2016) also found evidence of right lateralized delta entrainment in 13-year-old children listening to natural continuous speech. Evidence of delta entrainment in early

childhood may be indicative of entrainment occurring at even earlier developmental milestones. As the adult neural entrainment research becomes solidified and school age children begin to be explored, there is a growing investment in infant studies of neural entrainment to auditory stimuli.

### ***Infant evoked neural responses***

Human infants process speech in a different way than other acoustic input long before they are able to produce speech themselves. Evidence in neural responses in infants has produced conflicting results, which can most likely be attributed differences in methodological choices. Studies measuring hemodynamic responses, using fNIRS and fMRI in infants, have shown that infants process the prosodic components of speech in the right hemisphere, both during natural speech and non-language acoustic input (Dehaene-Lambertz, Dehaene, & Hertz-Pannier, 2002; Telkemeyer et al., 2009). However, when oscillatory responses are measured using EEG, as in adult populations, there are no differences in lateralization (Pena, Pettuluga & Mehler, 2010; Telkeyemer et al., 2011) or left ward lateralization has been found (Kalashnikova et al., 2018).

There is converging evidence of young infants' ability to entrain to acoustic stimuli. Telkemeyer and colleagues (2011) showed that newborns' brains can be attuned to the temporal structure of speech. In this study, newborns were presented with frequency-modulated non-speech stimuli corresponding to the phonological and the lexical speech rates, as their electrophysiological and hemodynamic responses were recorded. Infants neural responses showed synchronization with the non-speech modulations and were considered to be equivalent to the adult auditory steady-state response (ASSR), which is phase-locked to the amplitude envelope of auditory input. As promising as these findings are, hemodynamic studies leave little to be compared with adult studies of neural entrainment because of the differences in methodological uses: hemodynamic responses in infants are difficult to be compared with studies of brain phase correlation in adults that general use phase locking value or coherence methods in EEG. In general, fNIRS or fMRI studies lend little information about the temporal components of speech infants. In search of more exact timescales, a handful of studies have utilized EEG.

The few studies that have used EEG in infants in the first year of life have yielded mixed results for oscillatory responses to speech input. Evidence found by



Ortiz-Mantilla, Hämäläinen, Realpe-Bonilla & Benasich (2013) saw that the power of gamma oscillations in 6-month-old infants was increased during the processing of native phoneme contrasts, but not during processing of non-native contrasts, which suggests gamma oscillatory mechanisms play a role in infants acquiring native phonemes. However, the researchers used isolated syllables to compare native and non-native speech reactions, which limits extendibility to infants' receptive comprehension of natural speech.

Additionally, Pena, Pittaluga & Mehler (2010) investigated language discrimination at 3 and 6 months of age in both preterm and full-term infants, using three different languages of naturally produced speech, (i.e. 1 native language and 2 foreign languages). The authors found differences in the gamma band, as well as in the P200 component. However, differences in these electrophysiological responses were found across age groups, with the P200 component peaking earlier in the 6 month old's than in 3 months old. Additionally, they found differences in the gamma band oscillations that were exclusive for the native language at 6 months of age, but not at 3 months. The authors attributed this to the ability to discriminate their native language as further brain maturation occurred in the 6 month olds, but not the preterm 6 month olds (adjusted age 3 months). More specifically, they thought that the developmental change they witnessed could be the consonant repertoire beginning to solidify at 6 months of age.

In an additional study of brain specialization patterns in infancy, Nacar, Guerrero-Mosquera, Colomer & Sebastian-Galles (2018) sought to replicate Pena & colleagues using a sample of both monolingual and bilingual 4 to 5 month old infants. Utilizing similar stimuli, the research additionally sought to assess differences between monolingual and bilinguals evoked responses to the speech signal. The researchers analyzed the speech signal in two windows and found that the P200 component peaked earlier for the native language for monolingual infants, whereas bilingual infants showed no differences across languages in P200. In the second window of analysis, bilinguals showed higher power synchronization in theta for native language when compared to unfamiliar languages, whereas monolinguals showed no differences in within class languages. These conflicting pattern of results provides evidence of differences in language discrimination for bilinguals and monolinguals but leaves more questions about how brain oscillations are related to language discrimination in the first year of life.

To date, the only study that attempted to assess theta level entrainment to the speech envelope in infants found larger responses over the left as compared to the right hemisphere when listening both to infant- and adult-directed speech (Kalashnikova et al., 2018). The researchers' analyses however were focusing on assessing enhanced coherence to infant directed speech (IDS) in comparison to adult directed speech (ADS), rather than specifically to neural entrainment in varying frequency bands.

### ***Current study: experimental predictions and considerations***

Although a small number of infant studies have studied brain oscillatory patterns to speech through EEG, they have yielded mixed results with samples that differ significantly in age during the first year of life. It is worth exploring speech brain entrainment through a finer lens to determine characteristics of neural response in the first 6 months of infancy. As mentioned earlier, adult studies of speech brain entrainment place an emphasis on theta's role in entrainment of the neural and speech signal (Luo & Poeppel, 2007), as well as an emerging role for delta in entrainment. It is not known if oscillatory speech parsing also occurs early in infant development, and assuming it does, whether infants' and adults' profile of neural entrainment mirrors each other or differs significantly across differing speech timescales.

As the adult neural entrainment research becomes solidified, there is a growing investment in infant studies of neural entrainment to speech. Studying brain oscillations, by decomposing the EEG signal into specific different frequency bands, may shed more light on the cognitive processes that occur as infants process language in the first 6 months of life. It is quite surprising, given the functional relevance that the speech brain entrainment concept may have for language acquisition, that no study has specifically examined it in infancy, particularly at the age of behavioral language discrimination.

Research lines such as those conducted by Pena, 2010 and Nacar, 2018 have begun to characterize neural oscillations in the brain, but leave much to ponder, as their investigations in infancy led to discrepant results. Among other obstacles in this research line, infant auditory stimuli have differed widely across these studies. For example, as mentioned above, Kalashnikova & colleagues studied infants auditory response to the speech signal and showed that preverbal infants possess an endogenous neural mechanism to entrain to incoming speech before they have any

established knowledge of phonemes or semantic properties of speech. However, this study used both infant directed and adult directed speech stimuli to compare differences in entrainment, as a function of infants' preference, which makes it difficult to compare to Pena, Pittaluga & Mehler (2010) and Nacar, Guerrero-Mosquera, Colomer & Sebastian-Galles (2018)'s results using natural speech. Taking into account differences in stimuli length, it becomes even more complex to compare outcomes. Kalashnikova's stimuli was a constant stream of 8 minutes, whereas Pena and Nacar's were short phrases of 3 seconds in duration, presented with silent pauses between the natural speech sentences. Additionally, Kalashnikova's research analysis used a Time Response Function (TRF) to measure brain oscillations which may accurately assess evoked components of the speech, but does not necessarily target auditory entrainment (Doelling, et al., 2019). In sum, no experiment has identified stark similarities or differences between speech and neural synchrony in adult and infant entrainment. The neural mechanisms underlying infant language discrimination remain relatively unclear.

The present research aims to fill this gap and assess infant speech brain entrainment using EEG in passive listening tasks with young infants aged 4-5 months. The passive listening includes native and non-native linguistic rhythms in the form of continuous natural speech and focuses on low frequencies in the EEG. Primarily, we will use both native and novel languages with the participants to assess different entrainment patterns to differing languages. Specifically, we compare neural responses to the native language (either Spanish/Catalan) and a foreign unknown language of a different rhythmic class (English).

Expanding on Nazzi's behavioral model (2000), we anticipate language discrimination to be related to the rhythm and prosodic components of one's native language at this 4-5 month age range, as we focus on between-class discrimination. To truly target the rhythm and prosody of the speech input, resynthesized speech was used in the *saltanaj* format (Ramus & Mehler, 1999). This methodological choice was undergone to be able to isolate rhythm and expand our capacity to compare infant research results with adult entrainment research (unpublished work, Sebastian-Galles). Used previously in adult and infant behavioral studies, *saltanaj* changes fricatives into /s/, vowels into /a/, liquids into /l/, occlusives into /t/, nasals into /n/, and semi-vowels into /j/. In other words, *saltanaj* strips the lexical and syntactic information but preserves the duration, articulation, broad phonotactics and place of

each phoneme for the said language. Because the overall structure and organization of the syllables remains, syllabic rhythm is also intact. Importantly, because this age range is before the attrition of native phonemes, saltanaj should be sufficient to isolate the prosody and rhythm of the language and ensure that infants are not using other properties to differentiate the languages. We alternate the languages presented (e.g. native, foreign, native, foreign) during the testing period.

In order to link the temporal aspects of speech to the neural signal, it is important to assess different frequency bands that may illuminate differing cognitive functions that occur simultaneously to processing of different languages. Overall, we anticipate differences in the low frequency bands when listening to different languages because there is evidence of delta and theta's role in neural entrainment in adult and school age children populations (Rios-Lopez et al., 2019; Power et al., 2016; Pérez, Carreiras, Gillon-Dowens & Duñabeitia, 2015).

Using the majority of adult and child literature available, we predict differing modulations in the theta band when infants hear native and non-native speech. By honing in on evoked oscillatory responses at 4-5 months of age, we can study brain oscillations through the decomposition of the EEG signal and determine if infants differentiate the languages. We seek to know if infants' neural entrainment patterns differ when exposed to foreign and native speech sentences and we anticipate to see this difference in the power synchronization of theta to the speech envelope across different languages. We predicted infants would show more enhanced phase locking of the speech and neural signal (neural entrainment) for native speech in the theta frequency, as the previous literature stated found. We assess this by calculating phase locking values for the different languages presented (Lachaux et al., 1999). Overall, we hypothesize that enhanced entrainment in the native language will indicate that infants notice the difference between languages (e.g. that they discriminate them). In the foreign language case, we are less clear on the predictions for entrainment patterns because unknown languages are rarely investigated in the scant infant entrainment research. However, we anticipate that infant neural synchronization patterns will be clearly different for native and foreign continuous speech.

In sum, we can assess patterns in EEG evoked responses and determine how they contribute to language discrimination. Because speech brain entrainment may enable language learning and acquisition in young children (Leong & Goswami, 2015), it is both clinically and scientifically relevant to investigate how temporal

timescales for speech and neural patterns may facilitate language learning during the first year of life. This research proposal seeks to identify an endogenous neural mechanism for language discrimination and to show to what extent this is similar to adult models of speech brain entrainment. It is likely that infant speech brain entrainment is characterized by different features than adults. If preverbal infants have endogenous neural oscillations that entrain differently to different languages, before the attrition of phonetic and semantic information, what does that mean for language processing, learning and acquisition?

### *Methods and Materials*

#### *Participants*

Participants were tested at the Laboratori de Recerca en Infància at Universitat Pompeu Fabra. We intended to recruit infants aged 4 months 15 days to 5 months 15 days from the greater Barcelona, Spain area to participate. An adapted version of Bosch & Sebastian-Galles' language exposure questionnaire would be administered to establish the infants' language exposure and environment (Bosch & Sebastian-Galles, 2001) and we would recruit a monolingual sample. The exclusion criteria of the subjects included: exposure to another language (>20%), family history of intellectual or neurodevelopmental disorder, family history of dyslexia, endocrine disorders and regular usage of seizure medicine. The research was conducted in accordance with the principles expressed in the Declaration of Helsinki. The infants would be recruited from Hospital Quirón and Clínica Sagrada Família (two private hospitals in Barcelona). All parents of participants would sign written informed consent forms and would be given a child prize, either a shirt or bib, for participating. Given the circumstances of COVID-19, we piloted the experiment with four adult university students (2 males) and two 6-month old infants (2 males) from an additional experiment in the CBC before the closure of the CBC in March. Written informed consent was obtained from the participant or their caregiver before the experiment was conducted and participants were given an adult language questionnaire to gather information about language exposure and eliminate the possibility of dyslexia, as studies have shown people with dyslexia have atypical brain entrainment patterns (Power et al., 2016). The pilot participants were used for data analyses detailed below.

### ***Stimuli***

We used audio recordings of neutral sentences (~3 seconds in duration each) playing from speakers located behind a screen in the testing room as a static child friendly image of clouds was presented. Of note, the audio consisted of a native language (either Spanish or Catalan) to all participants and then a novel, foreign language of the different rhythmic class (English) during the trials. We selected 20 utterances in each language that were each between 2,800 ms and 3,200 ms in duration. The speech stream was adult directed speech, recorded by native speakers, as to be able to compare to adult studies of evoked oscillatory responses. The speech stimuli were resynthesized to *saltanaj* (Ramus & Mehler, 2000) to retain only rhythm and prosodic cues. Speech stimuli *saltanaj* examples can be found in Appendix I. One block was comprised of a native language presented for ~3 second intervals of 20 phrases. The novel language was then presented following the exact same structure. This was then repeated, so that in total each participant was presented with ~2.5 minutes of audio stimuli from both the native and foreign language. Triggers were time-locked to stimulus (utterance) onset for all languages, as well as the onset of inter-trial periods. Within each language block, utterances were separated by 500 ms pauses, and between block silences were 1000 ms in duration. All experimental stimuli were created in PsychoPy3 (Pierce, 2007).

### ***EEG recording***

The two infant participants were held by their seated caregiver in a dimly lit, sound-attenuated, electrically shielded room while a research assistant ensures the infant is calm comfortable. The caregiver was instructed to not speak to the child or provide any extra input. The four adult participants were seated on a comfortable chair and instructed to move as little as possible, including limiting eye and facial movements. Continuous EEG was recorded for 7 to 10 minutes during the passive listening task as a static child friendly image was projected on the center of the computer screen. EEG data was collected with a 128- channel Hydrocel Geodesic Sensor Nets (Electrical Geodesics, Inc., Eugene, OR) with a NetAmps 300 amplifier and using NetStation 4.5.7 software (EGI Inc) at a sampling rate of 1000 Hz. EEG data were rereferenced offline to a single vertex electrode (Cz) according to the international 10-20 placement system, with impedances kept below 100k $\Omega$ . The continuous EEG was

saved for offline analysis. Raw EEG data collected in NetStation were exported to MATLAB (version R2017a) for preprocessing and subsequent analysis.

### ***Data processing and analyses***

After much experimentation with infant pediatric EEG processing pipelines, all data processing and analyses were performed in Matlab (Mathworks, 2014b) and using EEGLab functions (Delorme & Makeig, 2004). Please see attached coding scripts in Appendix II for more specific analyses steps.

### ***Preprocessing***

EEG was preprocessed using the Maryland Analysis of Developmental EEG pipeline (Debnath et al., 2020), which is a MATLAB and EEGLab based platform for processing high artifact, developmental EEG data with EGI Net Station caps. Please see Figure 1 for a schematic of the pipeline's steps. The continuous data was sampled

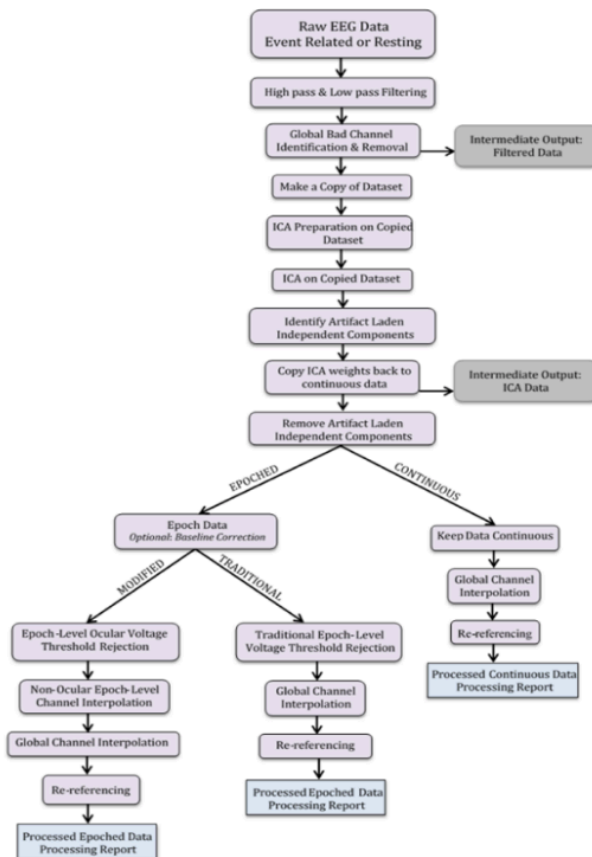


Figure 1: Schematic representation of the MADE pipeline (Debnath et al., 2020). Independent component analysis is abbreviated to ICA.

at 1000 Hz and then low and high pass filtered at 50 Hz and .3 Hz using the FIRfilt plugin in EEGLab. This choice was undertaken because empirical research has shown that data recorded with EGI systems tends to have high impedance with leaves it vulnerable to ultralow frequency artifacts (Kappenman & Luck, 2010). Following MADE pipeline guidelines, the outermost ring of channels was removed because they

tend to cause noisy recordings in infants and can bias the detection of bad channels in the next step. Next, bad channels were detected, using the ‘channel\_properties.m’ function from the FASTER EEGLAB plugin (Nolan et al., 2010).

An independent component analysis (ICA) was performed with the remaining 106 channels. An ICA hybrid approach was employed because ICA’s have been shown to perform better on data with certain level of stationarity. This hybrid approach includes making a “copy” of the data, utilizing a 1 Hz high-pass filter and removing data segments that contain excessive EMG or high/low amplitude data from the copy, running ICA on the copied dataset and then eventually copying the ICA weights (because they contain the information needed to identify artifacts) back to the original dataset that has not been adjusted with the 1 Hz high-pass filter (Viola et al., 2010). The “bad” ICS were then identified and removed with modified ADJUST EEGLAB plugin scripts (Mognon et al., 2011; Debnath et al., 2020). This process allows low frequency information, that typically can have excessive EMG activity, to be retained.

The EEG was subsequently epoched, with the duration of each epoch corresponding to the longest audio stimuli and the epochs were looped through for a final stage of artifact rejection that may have been missed during the ICA. Epochs containing more than 10% of data that exceeded voltage threshold parameters were rejected and for the remaining epochs, individual channels exceeding this threshold are subsequently interpolated at the epoch level. Finally, we interpolated missing channels using the spherical spline interpolation procedure and re-referenced the data to average reference. Please see article for more detailed justification and steps of the preprocessing. (<https://doi.org/10.1111/psyp.13580>). After preprocessing, we retained 1 adult participant (adult2) and 1 infant participant (infant 2) for further analyses. Due to the high impedance of the EGI cap, only these 2 participants had a sufficient number of epochs to meet MADE’s requirements for data retention. See Figure 2 below for an example spectroplot of one participant after preprocessing.



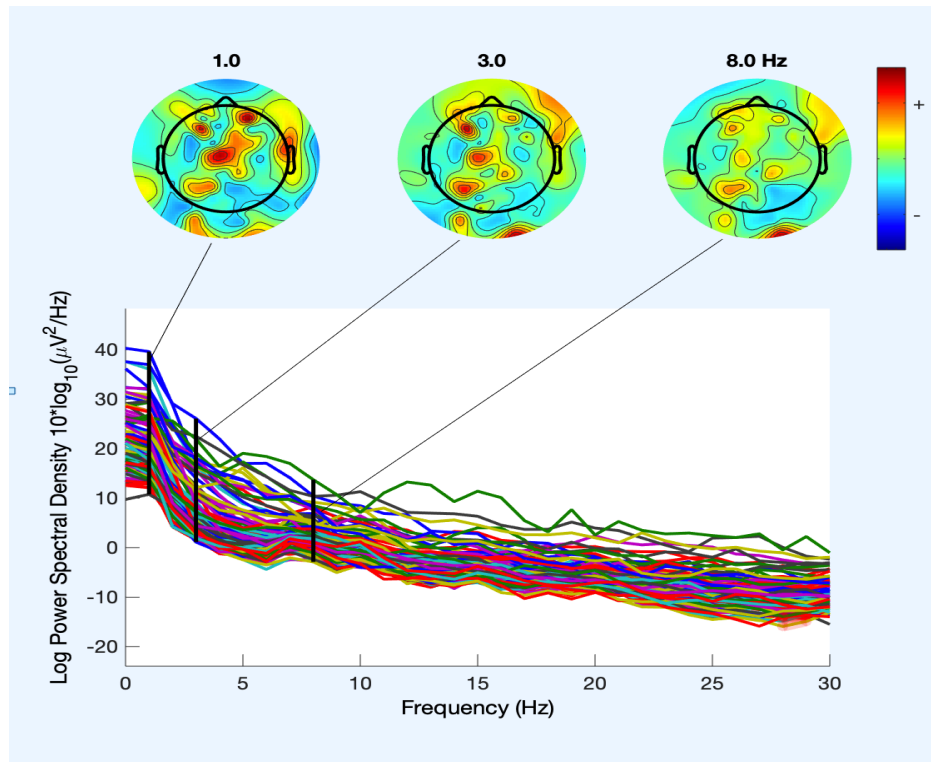


Figure 2: each colored line in the graph represents the frequency spectrum of different channel activity for the adult participant after preprocessing the EEG data. For example, the left scalp diagram shows the energy distribution of the scalp at 1 Hz, and it is mainly concentrated in the frontal midline. Other scalp diagrams show energy distributions at 3 Hz and 8 Hz for epochs retained after preprocessing.

## Results

### **Speech brain entrainment -Phase Locking Value (PLV)**

The preprocessed EEG data had zero padding applied to ensure no overlapping of epochs. The data were bandpass-filtered into the theta frequency with a Finite Impulse Response (FIR) filter with a Kaiser window from 3 to 8 Hz with a filter of minimal order applied twice: in the forward and then backward direction. Figure 3 below shows the EEG signal pre and post filtering for one participant.

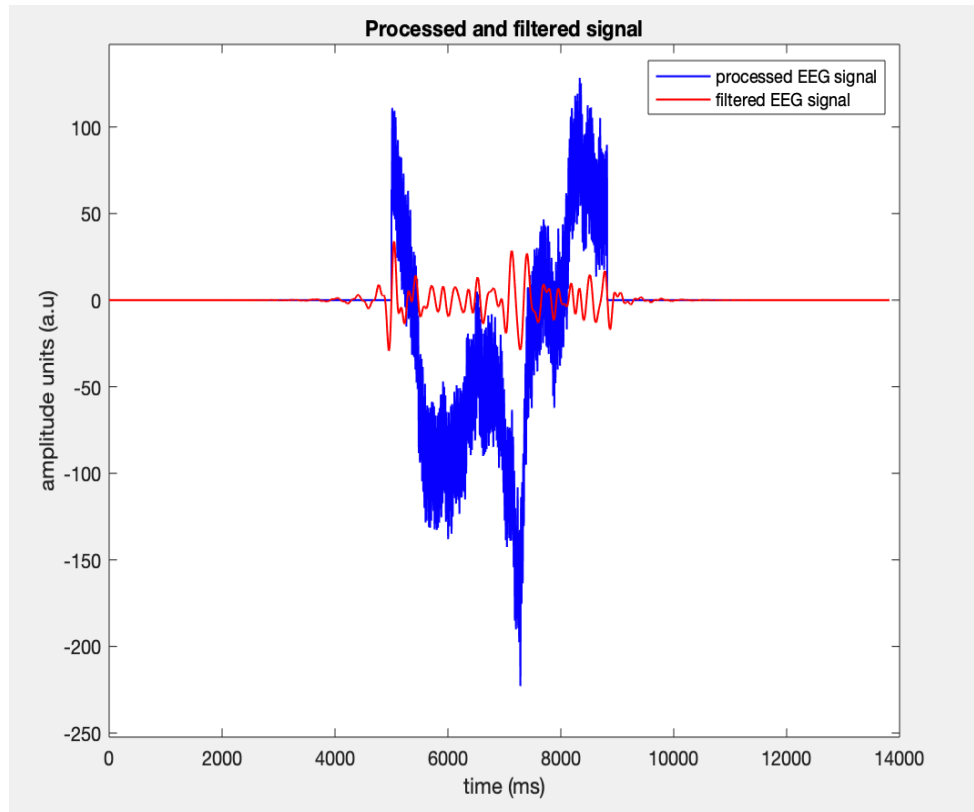


Figure 3: EEG signal after filtering into theta (3-8 Hz) with adult participant 2

Regarding the speech stimuli, the same FIR filter was applied to the audio signal, band pass-filtered between 3 and 8 Hz and then the audio and EEG signals were Hilbert-transformed to obtain the broadband amplitude envelope of the signals (Drullman, Festen, & Plomp, 1994). The phase series at the theta frequency was extracted from both the EEG signal and audio Hilbert transformed signal and divided into matching EEG and speech epochs of length 3.316s, corresponding to the longest audio stimuli duration.

Next, the coherence of the EEG and audio signals was calculated in order to determine the degree of neural entrainment during different speech input. Because previous studies in adults showing significant coherence between the speech and the brain signals at 3–8 Hz (Ding & Simon, 2012; others) we chose to concentrate our entrainment analysis on this frequency range. To calculate the relation (dependency) between the phase of EEG signals and the phase of speech signal, we can calculate coherence using a phase locking value (PLV; Lachaux et al., 1999). One PLV estimate for each epoch was computed in Matlab as shown below in Figure 4. To assess the degree of entrainment between the neural EEG signal and the speech

amplitude envelope the phase-locking value (PLV) was computed. The PLV takes values between [0, 1], where a value of 0 reflects the absence of phase synchrony and a value of 1 reflects perfect synchronization.

$$PLV_t = \frac{1}{N} \left| \sum_{n=1}^N \exp(j\theta(t,n)) \right|$$

In the PLV equation to the left (Leong et al., 2017),  $N$  is the data samples within each epoch and  $\theta$  is the phase difference

between the EEG signal and audio signal for each sample within the epoch at a specific time ( $t$ ).

Phase locking values were calculated for each participant and later for each block, and for each epoch by individual participant. The data was assessed for normality and found to be non-normal using Anderson-Darling tests, as well as qqplots (see Appendix IV for plots), thus Wilcoxon rank sum tests were utilized to assess differences between native and foreign neural responses. First, the average PLV over all epochs for each participant by block was pooled and used initially for analysis ( $p=0.10$ ). However, this small number of values of the averaged epochs is too small to be applied correctly with Wilcoxon rank tests. Given this and that the total number of average PLVs obtained per participant was only four, we decided to use permutation tests to run analyses. We performed permutations with 6 iterations, yielding 24 averaged PLV epochs from the original averaged PLV values (Nichols & Holmes, 2001). Next, using the permuted data, a Wilcoxon rank sum test was performed and showed a significant effect of language type on PLV with native language median=0.1147, foreign language median=0.1072 ( $Z = 5.18, p < 0.05$ ). Bearing this in mind, we conducted an alternate analysis to parse out further differences in PLV across the two language groups. In this third step, the data were split into individual subject to further explore the effect of language and each individual epoch was used with point to point PLVs. In other words, we compared vectors of PLV for each epoch signal at the individual sentence level between native and nonnative language for each participant.

Conducting Wilcoxon rank sum tests at the sentence level, we found that, for the infant, the medians of native language PLV and foreign language PLV were 0.111 and 0.108 ( $Z = -2.03, p = 0.043$ ). For the adult, the medians of native language PLV and foreign language PLV were 0.123 and 0.104, respectively ( $Z = -3.043, p = 0.002$ ). See Figures 4 and 5 below for a schematic of the average PLVs for each

participant. These extended analyses show that for both participants there was a significant effect of language for PLV at the single epoch level, although a stronger effect in the adult ( $p < 0.01$ ). Thus, PLV was also significantly stronger in the native language condition at the individual sentence level in comparison to the foreign language condition.

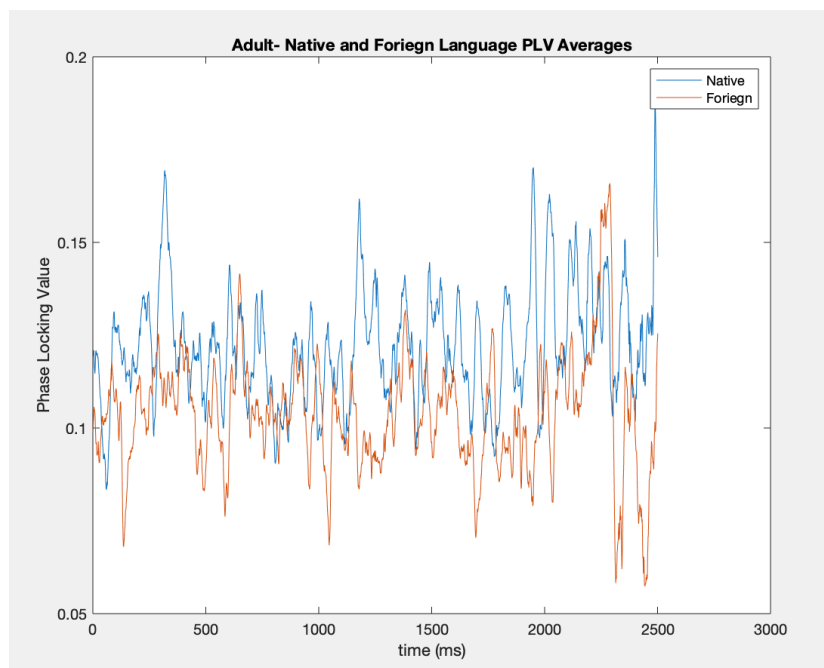


Figure 4: Adult participant-Phase Locking Values for the average of all epochs

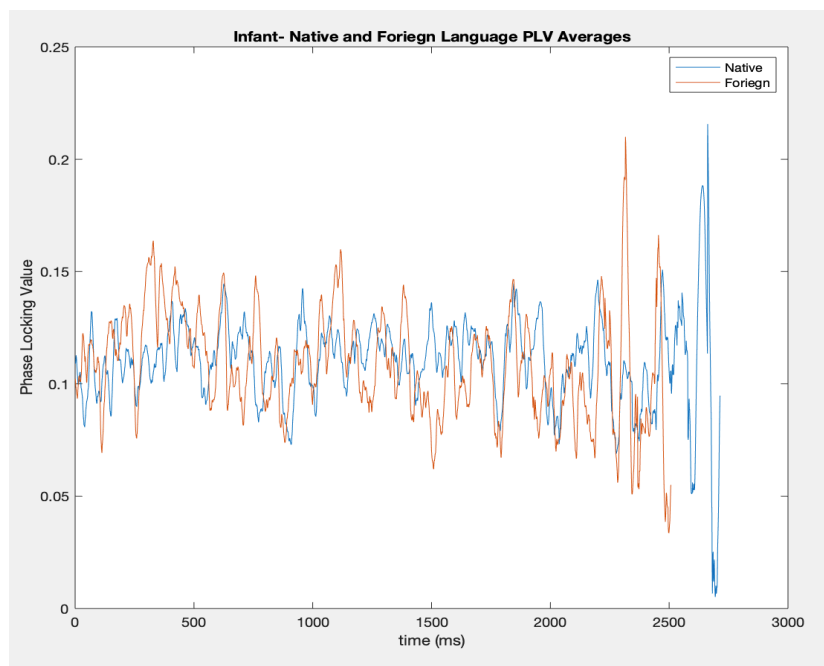


Figure 5: Infant participant-Phase Locking Values for the average of all epochs

### *Discussion*

Human infants make remarkable strides in language acquisition in the first year of life, although we know little about the neural mechanisms necessary to attain the early stages of language acquisition. Neural oscillatory entrainment to speech patterns has been researched thoroughly and is well developed as a mechanism for speech analysis in adults (Bourguignon et al., 2013; Giraud & Poeppel, 2012; Gross et al., 2013;). The research intention was to assess and begin to characterize infants' neural processing of the temporal structure of native and foreign natural speech and to compare their entrainment patterns in these two distinct languages. Our primary goal was to begin to ascertain the neural differences in language processing at 4 to 5 months of age and we particularly aimed to explore whether infants' neural response to their native language would be enhanced relative to their response to a foreign language in the theta frequency, corresponding to converging behavioral evidence that infants can use prosody to discriminate different languages at this age point (Bosch & Sebastian-Galles, 1997; 2001; Nazzi, Jusczyk & Johnson, 2000).

Bearing these goals in mind and using our available data sample, we found that there was a significant difference in PLVs for a native language in comparison to a foreign language, specifically in the theta frequency (3-8 Hz). This falls in line with previous adult research in speech brain entrainment and this frequency typically corresponds to syllable rates of utterance in said adult populations. However, with only 2 participants retained for analysis, one being an adult, these findings lead little extendibility to our intended infant population. However, postulating on the two participants' results, we initially notice that the entrainment, as calculated by PLVs, was stronger for the adult participant rather than the infant, indicating that entrainment may only be beginning to form in infancy. Intuitively, this finding supports both previous adult research strongly evidencing speech-brain entrainment and the still discrepant findings in infant neural explorations of speech acquisition.

Honing in on the infant data, we provide further evidence that infants use the prosodic aspects of speech to discriminate different languages, which links well with behavioral studies of language discrimination. As Bosch & Sebastian Galles (1997) showed that 4 to 5 month old infants can discriminate low pass filtered speech, which changes the speech stimuli to retain prosody and exclude phonetic information, our

saltanaj stimuli also isolated rhythm and prosody to ensure that infants could only use prosodic information to differentiate the native and foreign speech. The difference in phase locking values across language conditions indicates that infants noticed the change in language (e.g. discriminated the language), similarly to the infants investigated in Bosch & Sebastian Galles (1997) and Nazzi, Jusczyk & Johnson (2000).

Additionally, theta power has been linked to the processing of slow suprasegmental speech information, such as prosody and syllable properties of language in infancy (Telkemeyer, 2011) and strongly shown to play an essential role in speech brain entrainment in adults (Bourguignon et al., 2013; Giraud & Poeppel, 2012; Gross et al., 2013; Peelle & Davis, 2012; Poeppel, 2003). Other studies have linked increases in theta power (5-8 Hz) in infants to the perception of native/non-native contrasts in syllables and sentences in adults (Bosseler et al., 2013; Perez et al., 2015). These infant preliminary results also align well with Kalashnikova & colleagues' (2018) evidence for variations in the neural tracking of continuous speech in the theta frequency, dependent upon speech input. The differences found in theta neural entrainment here add credibility to theta's role in the development of effective speech perception in infancy and should be further investigated.

Because the naïve human perceptual system must perceive and encode multiple types of information simultaneously to analyze speech signals, understanding the neural basis behind decoding of the speech perception is essential. Measuring neural entrainment through the use of EEG and MEG provides a powerful tool to understand the neural hierarchies of speech processing, at both the syllable and phoneme levels. Acknowledging that this process may be more complex in infants still acquiring native language, we must employ both creative, yet rigorous tactics to measure these neural mechanisms.

The primary goal of our research was to understand the differences in neural processing of native and unknown foreign speech, and what specific role entrainment may play in those differences. Most significantly, there was a stronger correlation between neural oscillations and speech amplitude when infants listen to their native language rather than a foreign one. This suggests the earliest preliminary evidence of neural adaptations created by a naturalistic speech environment as perceptual narrowing begins to occur. By establishing a clear-cut relationship between endogenous neural and external speech patterns, we may begin to study the

development importance of entrainment in language disorders in early childhood, as well as expand research to investigate neural entrainment in children who grow up in bilingual environments.

### ***Future directions & limitations***

One limitation to the study is that, given the circumstances of COVID-19, we were unable to get an appropriate sample size of the targeted age group. We were only able to run the experiment with 2 infant participants and ran 4 adult participants after hearing that the CBC would close the same week due to the global pandemic. Future directions would ideally include a sample of at least 20 participants at the correct age range of 4-5 months old, corresponding to the age of language discrimination found in behavioral studies mentioned previously and providing clearer evidence of the possible connections between behavioral findings of language discrimination and simultaneous neural oscillatory patterns.

Another limitation of the study involves difficulty in recording EEG in pediatric populations and the high impedance frequently experienced with EGI Net Station EEG caps traditionally used in these infants. EEG signals recorded in infants typically are high artifact and only very short recordings can be obtained from infants, as they tend to move and become uncomfortable quickly. We attempted to compensate for these two factors through a rigorous preprocessing pipeline created specifically for infant EEG data obtained from EGI systems (Debnath et al., 2020).

Conversely, a positive of the EGI caps is the large number of electrodes used, which can provide expansive further research analysis opportunities. Expanding analyses to assess PLVs by different channel locations could provide more evidence about different brain areas' role in entrainment. It would be interesting to explore hemispheric dominance of response to speech as other infant studies have attempted to do. Slow delta and theta rates are typically better tracked over the right hemisphere in adults (Giraud et al., 2007) and it would be intriguing to assess if a similar pattern occurred in the first year of life, as mixed results have been found in infants. Previous hemodynamic studies in infancy have found right lateralized activations to speech prosody (Telkemeyer et al., 2009; 2011), whereas Kalashnikova et al., 2018 found leftward lateralized activations in 7 month old infants. Delving deeper, significant coherence to natural speech has been shown mostly in temporal and parietal channels using MEG in adult populations (Bourguignon et al., 2013; Gross et al., 2013).

Parsing out specific channels primarily located in the temporal and parietal areas and using only these for analysis could provide further evidence for brain area language specializations in infancy.

Lastly, expanding to different frequencies, the emerging role of delta in entrainment in school age children provides new research lines for infant neural entrainment research (Rios-Lopez et al., 2019). Using a similar research procedure as ours, potential future analyses could include an ANOVA with language, hemispheric location, and frequency as within subject factors. If an interaction were observed, post hoc Tukey HSD tests could then assess at which particular frequencies language group differences were significant at or not, and at which hemispheric location phase locking was stronger, as well as any frequency by hemisphere interactions. These potential future methodological choices and analyses could provide a better basis for characterizing evoked oscillatory responses to speech in infancy, at the age of behavioral language discrimination.

### ***Conclusions***

Multiple questions remain about how neural entrainment may play a role in language acquisition during the first year of life. We know that perceptual narrowing to native phonemes typically occurs by 10-12 months of age (Werker & Tees, 1984) while as early as 6 months of age, neural affinity for native prosodic rhythms is already in place (Pena et al., 2010). Behavioral studies have shown that infants can use speech rhythm patterns to decipher languages of different rhythmic class as early as 4 months of age (Mehler et al., 1998; Nazzi, Jusczyk & Johnson, 2000; Bosch & Sebastian-Galles, 2001). While no conclusions can be drawn due to the small data sample size, preliminary results indicate that oscillatory patterns in the brain are connected and altered by native prosodic rhythm patterns, as other research has found (Pena et al., 2010; Ortiz-Mantilla et al., 2013). Thus, speech brain entrainment is a proper basis to help facilitate infant language learning. More research is needed to ascertain the role that neural oscillations have in infant language acquisition and establish a clearer relationship between neural entrainment and the developmental phenotypic manifestation of language acquisition.



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### Statement of contribution

1. Conceptualization Ideas; formulation or evolution of overarching research goals and aims.

**Nuria Sebastian-Galles, Haley Medeiros & Konstantina Zacharaki**

2. Methodology Development or design of methodology; creation of models

**Nuria Sebastian-Galles, Haley Medeiros & Konstantina Zacharaki**

3. Software Programming, software development; designing computer programs; implementation of the computer code and supporting algorithms; testing of existing code components.

**Haley Medeiros, Konstantina Zacharaki, Ege Ekin Ozer, Silvana Silva Pereira & Francesc Xavier Mayoral**

4. Investigation Conducting a research and investigation process, specifically performing the experiments, or data/evidence collection.

**Haley Medeiros & Konstantina Zacharaki**

5. Formal Analysis Application of statistical, mathematical, computational, or other formal techniques to analyze or synthesize study data.

**Haley Medeiros & Konstantina Zacharaki**

6. Visualization Preparation, creation and/or presentation of the published work, specifically visualization/data presentation.

**Haley Medeiros**

7. Writing – Original Draft Preparation Creation and/or presentation of the published work, specifically writing the initial draft.

**Haley Medeiros**

8. Additional comments and acknowledgments:

*Conceptualization for the experiment came from Nuria Sebastian-Galles. Konstantina Zacharaki and I then had many conversations around the methodology of the experiment to be able to measure the proper hypotheses. We researched extensively any previous literature we could find, as there are not many entrainment studies in infant literature. We then discussed with fellow SAP members about the experimental setup and collaboratively designed the experimental setup based on the literature, both behavioural and neural and had Nuria Sebastian-Galles review our methodological experimental choices. Francesc Xavier Mayoral assisted with creating the experimental stimuli in Psychopy, as well as technological support when participants were being run with the EEG cap. Konstantina and/or I were present for all participants seen for the experiment. I researched and then applied or created codes for preprocessing/data analyses with Konstantina Zacharaki's input and advice. Ege Ekin Ozer and Silvana Silva Pereira assisted with EEG data processing. I am extremely grateful for the help I received from all of these people, but especially Konstantina Zacharaki, who mentored and assisted with me with the majority of the aspects of my thesis. I would like to thank Nuria Sebastian-Galles for the opportunity to work on such a research project, as well as her mentorship and guidance throughout the process as I gained*

*a vast amount of new research skills and knowledge. Lastly, I would like to thank all of the members of the Speech Acquisition & Perception Group for allowing me the opportunity to work and collaborate with them, and know that from these scintillating discussions and collaborations, I learned and grew very much.*

## Appendices

- I. Saltanaj stimuli link <http://www.lscp.net/persons/ramus/resynth/ecoute.htm>
- II. The scripts generated for preprocessing and analysis during the current study are available in the GITHUB repository:  
<https://github.com/haleymedeiros/TFM-scripts>

*Preprocessing methodological choices:* After much testing with different EEG preprocessing programs, such as EEGLab and Fieldtrip, we began investigating different infant EEG pipelines to use, as we learned more and more that pediatric EEG research differs greatly from adult EEG research. We initially used the Harvard Automated Preprocessing Pipeline for EEG (Gabard-Durham et al., 2018), but ultimately chose to use the Maryland Automated Preprocessing Pipeline (Debnath et al., 2018) because it contained more rigorous criteria for EEG data retention and was specifically created for pediatric EEG that used EGI Net Station caps, like our data sample.

- III. Layout of Hydrocel EEG Net Station 128 electrode cap used for experiments:

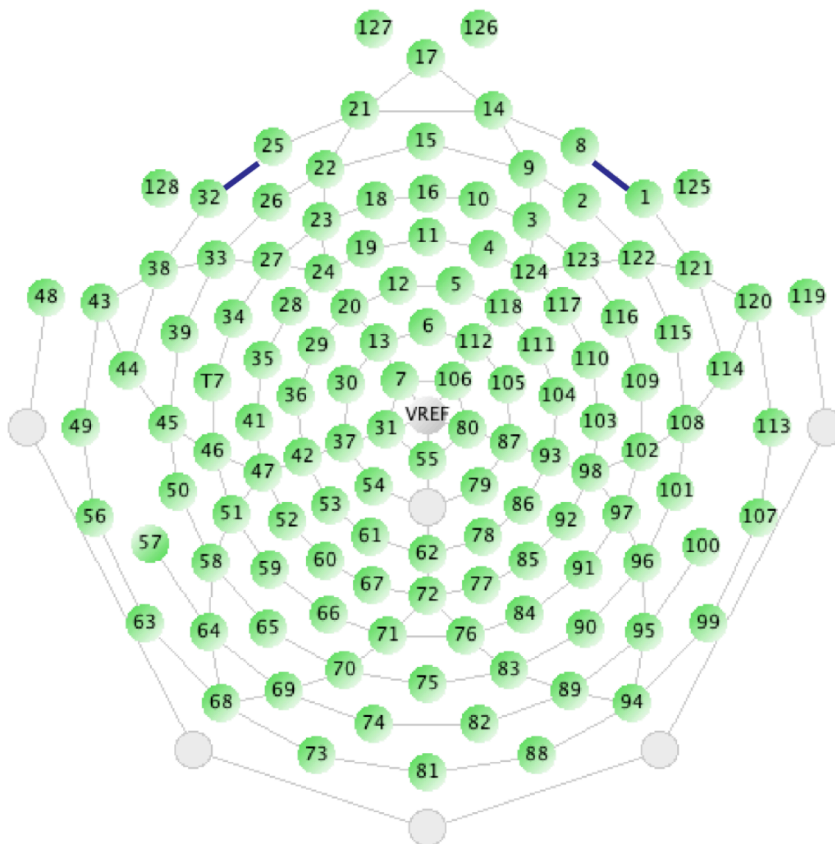
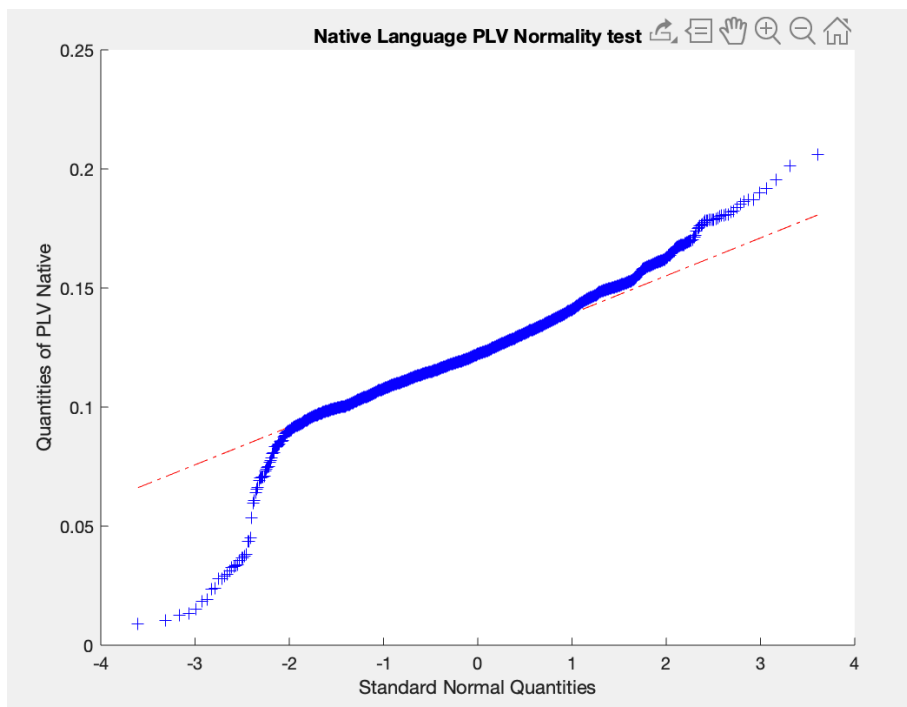
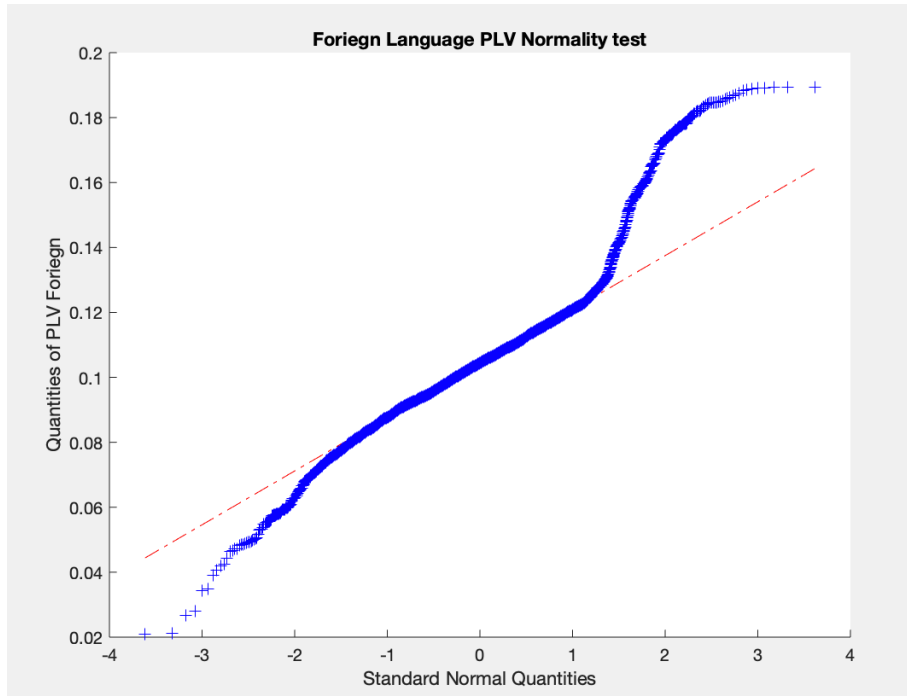


Figure B-3. 128-channel HCGSN v.1.0



IV. QQ plots to test normality for both participants retained after preprocessing  
 i. *Adult*



ii. *Infant*

