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FREQUENCY FOLLOWING RESPONSE (FFR): EXAMINING THE VARIABILITY OF THIS COMPLEX RESPONSE

by
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A thesis
Presented to the faculty of
Towson University
in partial fulfillment
of the requirements for the degree
Doctor of Audiology

Department of Audiology, Speech-Language Pathology and Deaf Studies

Towson University
Towson, Maryland 21252

May, 2017

TOWSON UNIVERSITY

OFFICE OF GRADUATE STUDIES

THESIS APPROVAL PAGE

This is to certify that the thesis prepared by Laura Grinstead entitled Frequency Following Response (FFR): Examining the Variability of this Complex Response has been approved by the thesis committee as satisfactorily completing the thesis requirements for the degree Doctor of Audiology (Au.D)

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Acknowledgements

I would like to thank my thesis committee for their continued support throughout this process. I'd especially like to thank Dr. Ananthakrishnan, whose instruction and guidance has been instrumental in this project, as well as my graduate school career. I would also like to thank my family and friends for keeping me grounded through the stress. I'd like to thank my cohort for always providing motivation and encouragement. Lastly, I'd like to thank my thesis partner, Lindsay, for being available for late night thesis sessions and for holding me accountable throughout this process. I can't thank you all enough for sticking with me through these few years.

Abstract

Frequency Following Response (FFR): Examining the
Variability of this Complex Response

Laura Grinstead

Objectives

The goal of the current study was to investigate the subject-related variations seen in the Frequency Following Response (FFR) in normal hearing listeners. We examined the subject factors of musical experience and gender to determine what, if any, effect these had on FFR amplitudes recorded in response to low frequency pure tone stimuli.

Methods

Thirty normal hearing adults were recruited as participants in the study. FFRs were recorded for each participant using three stimuli: 250 Hz, 500 Hz, and 1000 Hz pure tones. Participants were grouped based on length of musical training and by gender. Fast Fourier Transform (FFT) analysis was performed on FFR waveforms reflecting brainstem neural representation of temporal fine structure (FFR_{TFS}). Analyses of Variance (ANOVAs) were performed to investigate the effect of stimulus frequency on brainstem neural representation of pitch, and to investigate the effect of musical training and gender on the brainstem neural representation of a 250 Hz pure tone stimulus.

Results

Significant differences in FFR amplitudes were seen between each stimulus frequency (250, 500, and 1000 Hz), such that larger FFR amplitudes were seen for lower

frequency stimuli. For FFRs recorded in response to the 250 Hz stimulus, no significant effects of gender or musicianship were seen.

Conclusions

Examination of subject-related variability within normal hearing listeners is an important step toward introducing the FFR into the audiology clinic. Further research on the variability in the FFR across the lifespan, in individuals with normal hearing and hearing loss, in varying stages of sleep and attention, is necessary before the FFR can become a clinical tool.

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Introduction

Evoked potentials provide objective information regarding neurologic function in response to stimuli, by measuring bioelectrical signals through electrodes usually placed on the skin, tympanic membrane, or round window of the cochlea (Burkard & McNerney, 2009; Hood, 1998; Nuwer, 1998; Paulraj, Subramaniam, Yaccob, Bin Adom, & Hema, 2015). Visual, somatosensory, or auditory stimuli may be used to evoke these potentials (Nuwer, 1998). Responses obtained from visual stimulation can aid in the diagnosis of neurodegenerative diseases and demyelinating disorders, including multiple sclerosis (Nuwer, 1998). Somatosensory evoked potentials use electrical stimulation to assess the integrity of motor nerve pathways (Newer, 1998). Auditory evoked potentials (AEPs) can be used to evaluate functioning of the auditory nervous system at varying levels of the auditory nerve pathways (Robinson, 2015).

The Frequency Following Response (FFR) is one such AEP, recorded from the level of the rostral brainstem, that provides reliable and objective information regarding neural encoding of pitch (Skoe & Kraus, 2010). The FFR is becoming a promising electrophysiological clinical tool; however it is highly susceptible to influences of subject variability. We are interested in studying the effects of subject-related factors on the strength of the FFR. We will discuss AEPs broadly, before focusing on the FFR in detail.

Literature Review

Auditory evoked potentials are elicited through auditory stimulation and measured using electrodes placed at varying locations on the scalp. Clinically, AEPs can be used for a variety of purposes including: estimation of auditory thresholds, determining type of hearing loss (conductive, sensorineural, or retrocochlear), and intraoperative monitoring (Burkard & McNerney, 2009). While the Auditory Brainstem Response (ABR) is the most clinically useful AEP currently, there are a number of AEPs that give us further information regarding the integrity of the auditory pathways (Burkard & McNerney, 2009).

In order to organize the discussion of AEPs, we can classify responses according to type of response (transient, sustained, or perceptual; endogenous or exogenous), latency, anatomical generator, and type of recording (near-field or far-field). Picton, Woods, Baribeau-Braun, and Healey (1977) developed three main classifications for type of response: transient, sustained, and perceptual. Transient and sustained responses are most commonly used for objective audiological testing, as they do not necessitate conscious action from the patient. Transient evoked potentials are sensitive to a change in stimulus presentation, such as the onset, while sustained potentials respond to continual stimulus presentation (Hood, 1998; Picton et al., 1977). An example of a transient response is the ABR, as the neural activity is measured from neurons that are sensitive to the onset of a stimulus (Hood, 1998). The cochlear microphonic (CM) is an example of a sustained response, due to its ability to reflect the physical properties of the stimulus (Hood, 1998). Transient and sustained auditory evoked potentials may further be classified according to latency, or the timing of the response post-stimulus presentation (Picton, 2010).

Picton (2010) has outlined these latency classifications, which divide evoked potentials into first, fast, middle, slow, and late responses. First potentials, such as the cochlear microphonic, have latencies of 0-5 ms. Fast potentials, including the ABR and the

Frequency Following Response (FFR), have latencies of 1-15 ms. Middle latency evoked potentials, such as the Middle Latency Response, occur 10-50 ms post-stimulus presentation. Slow potentials have latencies of 30-500 ms, while late potentials, such as the Mismatch Negativity (MMN), occur 200-1000 ms after stimulus presentation (Picton, 2010). Due to the overlap of latency values across classifications, a three-tiered system can also be used, with fast potentials occurring between 0 and 10 ms post-stimulus presentation, middle potentials occurring at latencies of 10-50 ms, and late potentials occurring greater than 50 ms post-stimulus presentation (Picton et al., 1977).

Based on variations seen in this response according to where the electrodes are placed, we are able to determine the anatomical site of generation of AEPs (Picton, Hillyard, Krausz, and Galambos, 1974). AEPs are able to measure contributions from the entire auditory pathway, from the cochlea to the auditory cortex (Picton et al., 1974). The AEPs with the shortest latencies correspond to the more caudal auditory structures, while AEPs with longer latencies reflect responses from more rostral structures in the auditory nervous system (Burkard & McNerney, 2009). The early AEPs, specifically Wave I of the ABR, are thought to reflect activity of the distal auditory nerve (Burkard & McNerney, 2009; Picton et al., 1974). Waves II through IV of the ABR are thought to have generators located in the cochlear nucleus as well as the superior olivary complex (Picton et al., 1974). The exact origin of Wave V of the ABR is unclear, but the primary response is likely a result from either the lateral lemniscus or the inferior colliculus (Picton et al., 1974). Middle latency AEPs have components generated in the thalamus, the primary auditory cortex, and the auditory association areas (Picton et al., 1974). The AEPs with the longest latencies appear to reflect responses originating in the frontal cortex (Picton et al., 1974).

AEPs can also be described as endogenous or exogenous, depending on the nature of the response (Hood, 1998; Landa, Krpoun, Kolarova, & Kasparek, 2014). Early AEPs,

including the ABR, tend to be classified as exogenous, or sensory potentials, because they are dependent on the properties of the presented stimulus, such as stimulus intensity (Hood, 1998; Landa et al., 2014; Picton, 2010). Subject attention or cognitive state does not have any effect on the measurement of the response for exogenous potentials (Picton, 2010). In contrast, endogenous AEPs are typically those with longer latencies and depend on the subject's cognitive status (Hood, 1998; Landa et al., 2014; Picton, 2010). In these endogenous late AEPs, response waves (specifically waves N1 and P2) have been shown to be enhanced when subjects consciously attend to the stimuli as compared to ignoring the same stimuli (Picton, 2010).

A final characterization of AEPs is how they are measured/recorded. Electrodes can be placed at near-field locations, which are close to the anatomical site of generation though invasive techniques (Picton, 2010). For near-field recordings of cochlear potentials, needle electrodes can be placed through the tympanic membrane and placed on the round window to measure electrical responses (Picton, 2010). This technique is sometimes used for electrocochleography, in order to enhance the magnitude and stability of the recording (Picton, 2010). In contrast, far-field recordings, where electrodes are placed on the surface of the skin, are more commonly used in clinical settings, as they are non-invasive (Picton, 2010). ABRs and FFRs are commonly measured as far-field recordings. These scalp-recorded AEPs record measurements from multiple anatomical generators, as they receive input from a range of deeper structures, which can be greater than 10 cm away from the electrode placement (Jewett & Williston, 1971; Picton, 2010).

Based on these classifications, we can describe the FFR as a scalp-recorded, exogenous, sustained, fast AEP generated at the level of the brainstem (Hood, 1998). Auditory electrophysiology (including that which underlies the FFR) represents an intersection of acoustics, speech science and auditory neurophysiology. Therefore, a strong

understanding of acoustics and neural encoding is necessary before further discussion of the FFR.

Acoustics

Acoustics is defined as the study of sound, including its production, propagation, detection, and perception (Borden, Harris, & Raphael, 1994; Rossing, 2014). Sound travels through an acoustic medium, such as air, in the form of *longitudinal* waves (Emanuel & Letowski 2009; Johnson, 2011). Longitudinal waves are comprised of areas of compression and rarefaction, or areas of greater and lesser densities, respectively, as particles move in the same direction as the traveling wave (Borden et al., 1994; Emanuel & Letowski, 2009).

Sound waves are measured according to 3 basic features: frequency, period, and amplitude. Frequency is the number of cycles in a wave that occur in one second (Emanuel & Letowski, 2009). Frequency is usually reported in hertz (Hz), where 1 Hz is equal to one cycle per second (Emanuel & Letowski, 2009). Frequency is related to the perception of pitch; an increase in waveform frequency is perceived as an increase in pitch (Borden et al., 1994). The frequency of a waveform is inversely related to the period (Emanuel & Letowski, 2009). The period of a waveform is the length of time taken to complete one full cycle (Emanuel & Letowski, 2009). Figure 1 shows an example of a sine wave with a period of 4 seconds. The purple arrow denotes one cycle, which is completed in 4 seconds. Frequency can be calculated from the period of a wave; in this example the inverse of 4 seconds is ¼, or 0.25 Hz. The last feature, amplitude, is the height of the waveform from the baseline to the point of maximum displacement (Emanuel & Letowski, 2009). In Figure 1, the blue arrow illustrates the amplitude of the sine wave.

Sound waves can be classified as aperiodic or periodic (Johnson, 2011). Periodic sounds are comprised repeating waveforms, while aperiodic sounds do not have a discernible repeating pattern (Johnson, 2011). Aperiodic sound waves are random in nature and can be

classified as noise or transient (Johnson, 2011). White noise contains equal energy across frequencies and sounds like static (Johnson, 2011). Transient sounds are fast, impulse sounds that create a sudden disturbance, such as a clap (Johnson, 2011).

Waves can also be classified as simple or complex (Johnson, 2011). Simple periodic waves result from simple harmonic motion and resemble sine waves (Johnson, 2011). Figure 1 shows an example of a simple periodic wave. When two or more sine waves are combined, the result is a complex wave (Borden et al., 1994; Johnson, 2011).

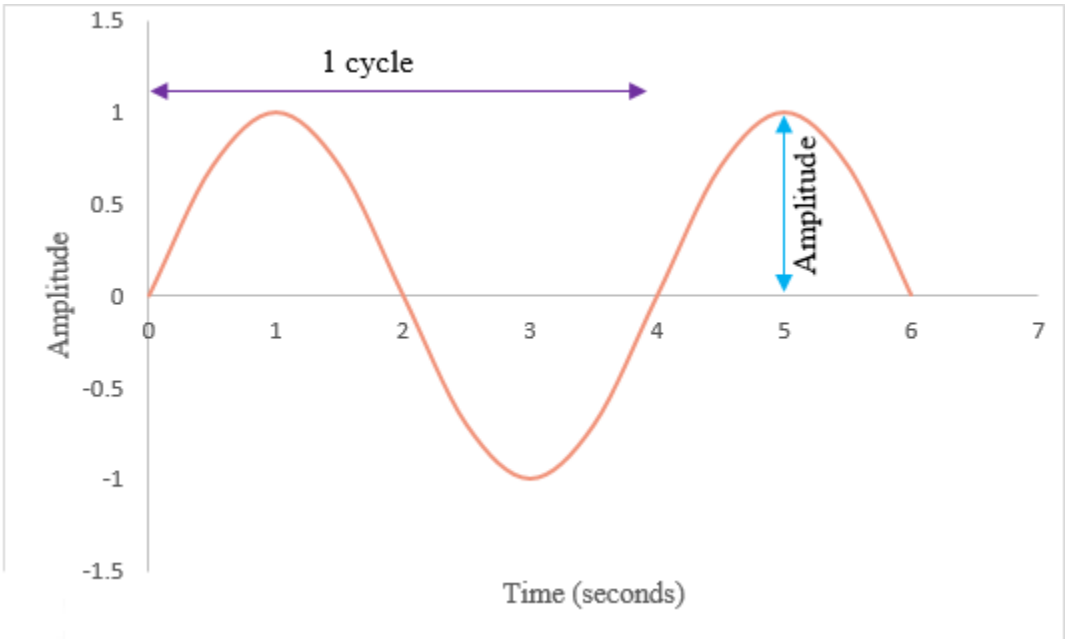

Figure 1. An example of a simple, harmonic wave with a period of 4 seconds, a frequency of 0.25 Hz (cycles per second), and an amplitude of 1.

Complex waves can be broken down through waveform analysis, allowing us to se the components of the waveform (Emanuel & Letowski, 2009). The lowest frequency of a waveform is referred to as the fundamental frequency (Borden et al., 1994; Emanuel & Letowski, 2009). If the components of a waveform are whole number multiples of the fundamental frequency, they are called harmonics (Borden et al., 1994; Emanuel & Letowski, 2009). Complex periodic waves are produced when the components are whole number multiples of the fundamental frequency, or harmonics (Walker, Bizley, King, & Schnupp, 2011). Panel (A) of Figure 2 gives an example of a complex periodic wave, which is characterized by a clearly repeating pattern. Speech is an example of a periodic, complex stimulus. Complex aperiodic sounds, such as percussion instruments, are the result of components that are not whole number multiples of the fundamental frequency. An example of a complex aperiodic sound is shown in Panel (B) of Figure 2.

Frequency, period, and amplitude are convenient ways to measure waveforms given as a function of time (Emanuel & Letowski, 2009). However, waveforms in the time domain do not allow visualization of the frequency components present in the wave (Emanuel & Letowski, 2009). In order to view the frequency components present in a waveform, a Fast Fourier Transform (FFT) must be performed to produce a spectrum in the frequency domain (Emanuel & Letowski, 2009). An example of a complex periodic waveform is shown in Figure 3A; with frequency components after FFT was performed shown in (B).

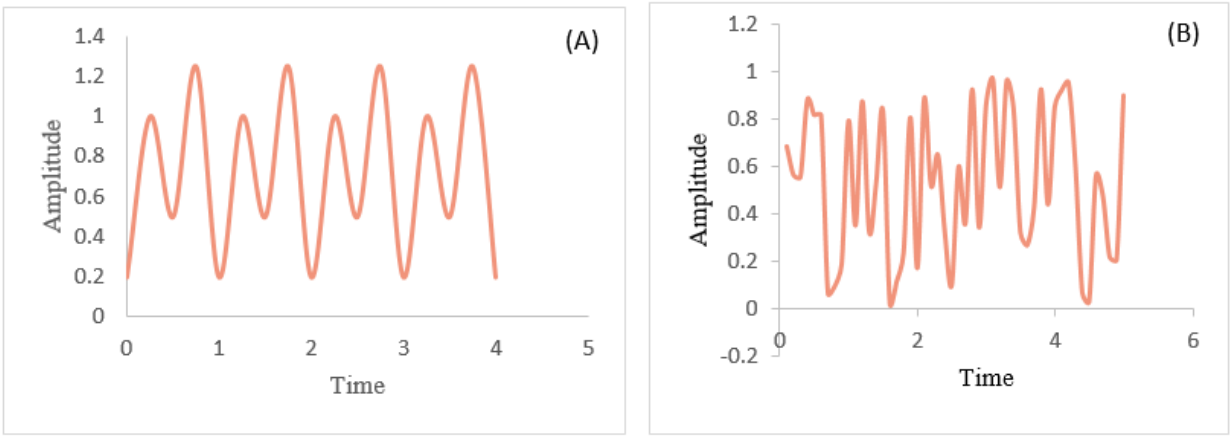

Figure 2. Examples of complex waves. (A) A complex periodic wave, resulting from components that are harmonics of the fundamental frequency. (B) A complex aperiodic wave, resulting from components that are not harmonics of the fundamental frequency.

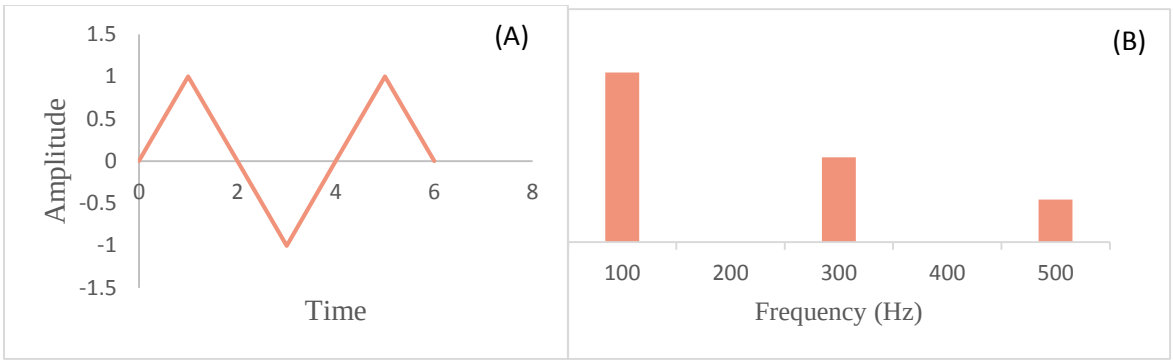

Figure 3. Two ways of viewing a triangle wave, in the time domain and the frequency domain. (A) An example of a triangle wave, shown in the time domain. Harmonics of this wave include odd whole number multiples of the fundamental frequency only. (B) Example of an FFT of a triangle wave.

As indicated earlier, speech is an example of a complex, periodic waveform. Speech is produced through the expiration of air and vibration of the vocal folds (Borden et al., 1994). The frequency of vocal fold vibration is related to the fundamental frequency of speech (Borden et al., 1994). The air is shaped by the vocal tract as it is expelled, which allows us to produce different sounds (Borden et al., 1994). Energy is enhanced at certain frequencies, based on the shape of the vocal tract (Borden et al., 1994). Those frequencies associated with greater energy due to the shape of the vocal tract are called resonance frequencies (Borden et al., 1994).

In order to view the resonances, or formants, of speech sounds, spectrograms are often used. Spectrograms allow us to see the amount of energy present at different frequencies over a period of time, allowing us to visualize the variation in speech production (Borden et al., 1994). An example of a spectrogram, from an FFR recording, is shown in Figure 4. Clear bands of energy are visible at various frequency marks throughout the given time window. Regions of yellow and purple represent areas of greater energy; which are seen at around 20 Hz, 250 Hz, and 500 Hz for 0 ms to 150 ms. Bright blue regions are areas that do not contain much energy.

Depending on the frequencies of interest, acoustic filters may be applied to eliminate regions of frequencies that are not needed (Johnson, 2011). A low-pass filter allows frequencies lower than a specified cut-off frequency to pass, while blocking higher frequencies (Emanuel & Letowski, 2009; Johnson, 2011). In contrast, a high-pass filter blocks frequencies lower than the cut-off frequency, and allows higher frequencies to pass (Emanuel & Letowski, 2009; Johnson, 2011). A band-pass filter combines the characteristics of a high-pass and a low-pass filter, and sets the two cut-off frequencies (Emanuel & Letowski, 2009; Johnson, 2011). The frequencies between the cut-off frequencies are allowed to pass, while those outside are blocked (Emanuel & Letowski, 2009; Johnson,

2011). A band-reject filter also has two cut-off frequencies, but in this case, the frequencies between the cut-off frequencies are rejected, while those frequencies outside are allowed to pass (Emanuel & Letowski, 2009).

All stimuli can be classified as time varying vs. steady state. Time-varying stimuli, such as speech, are characterized by a change in frequency over time. Panel A of Figure 5 shows a time-varying stimulus, with a frequency of 100 Hz, rising to 250 Hz at 30 ms. In contrast, steady-state stimuli do not vary in frequency, such as a continuous pure tone signal. Figure 5, panel B shows a steady state stimulus, such as a pure tone, with a frequency of 200 Hz.

Each type of stimulus described above, including simple and complex waves, and time varying and steady state stimuli, can be used to elicit AEPs. Now that the major acoustical properties of sound stimuli have been discussed, the next step is to describe the physiological representation of these acoustical properties. The following section describes neural encoding of auditory stimuli.

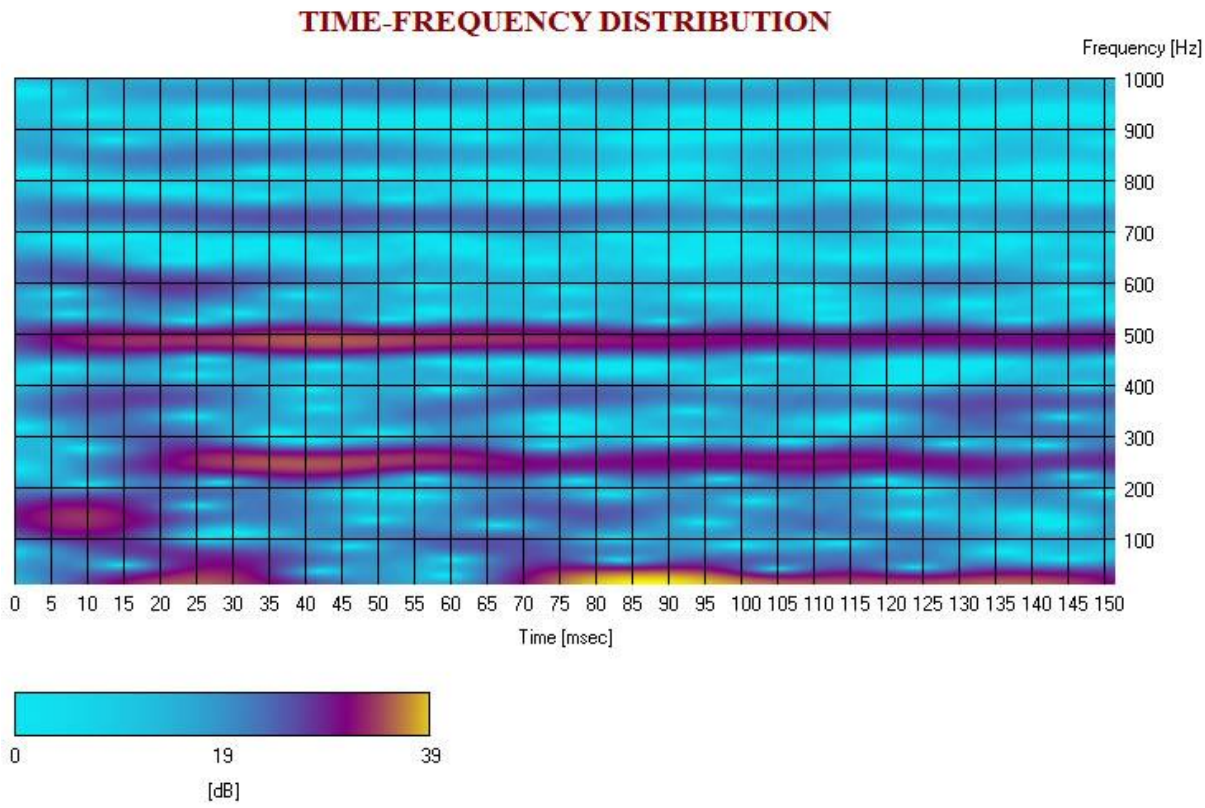

Figure 4. Example of a spectrogram, with time in milliseconds on the x-axis and frequency on the y-axis. Light blue represents low energy areas, with darker purple, red, and yellow representing areas of greater energy.

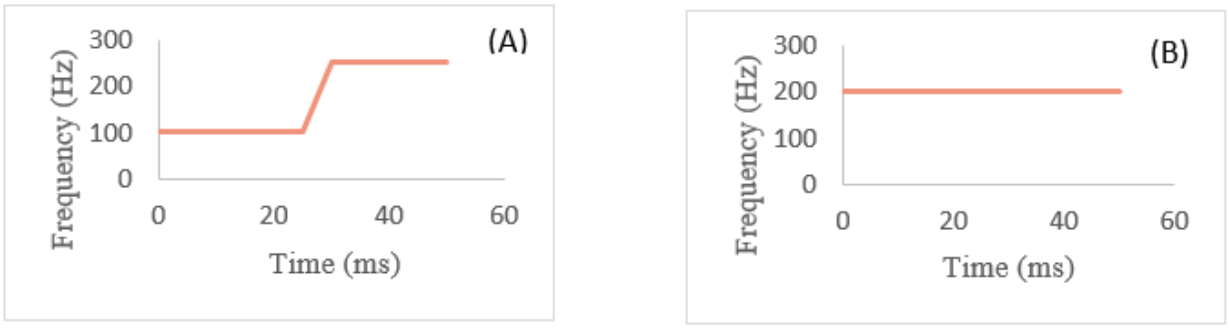

Figure 5. Example of a time varying stimulus and a steady-state stimulus. (A) A time-varying stimulus with a frequency of 100 Hz rising to 250 Hz at 30 ms post-stimulus onset. (B) Example of a steady-state stimulus with a consistent frequency of 200 Hz.

Neural Encoding

There are two primary ways the auditory nervous system encodes information regarding auditory stimuli: place mechanisms and temporal mechanisms (Moore & Glasberg, 1989; Walker et al., 2011). Place mechanism refers to the auditory stimulus activating a specific location along the basilar membrane, corresponding to the stimulus frequency (Walker et al., 2011). Temporal encoding involves auditory neurons firing based on timing information in the stimulus, due to phase locking (Moore & Glasberg, 1989; Reichenbach & Hudspeth, 2012; Walker et al., 2011). When working appropriately, neurons will fire on the positive-facing peaks of the stimulus, corresponding to the period of the stimulus as seen in Figure 6 (Moore, 2008). These auditory neurons correspond with the tonotopic organization of the basilar membrane, with neurons sensitive to high frequency stimuli located at the base of the cochlea, and low frequency-sensitive stimuli located at the apex (Henry & Heinz, 2013).

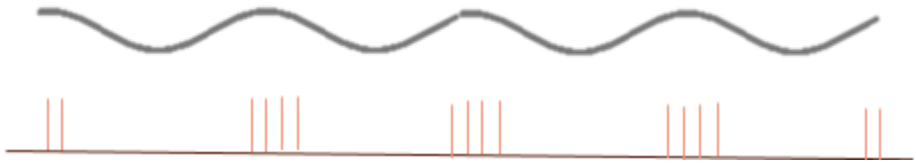

Figure 6. Representation of neural phase-locking to a sine wave signal. Neural firing (shown by orange lines) corresponds to positive-going peaks of an auditory stimulus.

Temporal encoding through neural phase-locking occurs at multiple levels of the auditory system. As you ascend the auditory pathway, the upper frequency limits of phase-locking decrease (Picton, 2010). Moore and Sęk (2009) reported effective phase-locking at the level of the basilar membrane through approximately 5000 Hz. At the level of the brainstem, neurons are able to phase-lock for frequencies up to 1000-1500 Hz or greater (Picton, 2010). Cortical neurons are only able to phase-lock to frequencies up to 50 to 100 Hz (Picton, 2010). The brainstem is the anatomic level of most importance to our present discussion, as this is where the FFR is generated. Aiken and Picton (2008) found FFR recordings were not measurable when the frequency of a stimulus exceeds approximately 1500 Hz, which reflects activity at the level of the brainstem. It is important to note that as we analyze higher portions of the auditory system, neurons have longer refractory periods and have an ability to phase-lock to only lower frequency stimuli.

As previously discussed, neural encoding of auditory signals is a complex process. Figure 7 displays the process of neural encoding for both a simple stimulus (Panel A) and a complex stimulus (Panel B). Consider a simple 200 Hz pure tone stimulus arriving at the basilar membrane, after passing through the outer and middle ears. The basilar membrane can be thought of as a series of overlapping bandpass filters, each tuned to a specific characteristic frequency (Henry & Heinz, 2013; Sayles & Winter, 2008). High characteristic frequency filters are located toward the basal end of the cochlea, while low characteristic frequency filters are located at the apical end. Pure tone stimuli are made up of a single component, which will pass through its designated filter alone, without any other components. As shown in Panel A of Figure 7, a single component reaches the basilar

membrane filters, and reaches the output stage as a single sine wave, or a *resolved* harmonic (Sayles & Winter, 2008).

In contrast, consider a complex stimulus arriving to the basilar membrane. As discussed, complex stimuli are comprised of multiple pure tone stimuli. The harmonic components of the stimulus are broken down and each harmonic arrives at the bandpass filter on the basilar membrane corresponding to its frequency. It is important to note that the cochlear bandpass filters are logarithmically-spaced along the basilar membrane (Henry & Heinz, 2013). Due to linearly-spaced harmonics, fewer individual frequencies will pass through low characteristic frequency filters than high characteristic frequency filters (Sayles & Winter, 2008). Low frequency components generally pass through individual filters, as described above for simple stimuli, resulting in resolved harmonics, as shown in purple in Panel A of Figure 7. However, multiple high frequency harmonic components may arrive at the same broadly tuned high characteristic frequency filter, as shown in Panel B of Figure 7. If multiple components pass into a single filter, they are combined, and the filter output is a complex wave (Sayles & Winter, 2008), made of *unresolved* harmonics, as shown in purple in Panel B of Figure 7. The complex wave is made of slow amplitude variations or "envelope" superimposed on rapid oscillations known as "temporal fine structure" (TFS). The overall shape of the fluctuations of the temporal fine structure or TFS is shown in purple in Figure 7 and the spectral envelope of the stimulus is shown in blue (Moore, 2008). This envelope reflects the fundamental frequency (F0) of the stimulus (Ananthakrishnan, Krishnan, & Bartlett, 2016). Neurons are able to lock onto the peaks of both the TFS and the spectral envelope (Aiken & Picton, 2008).

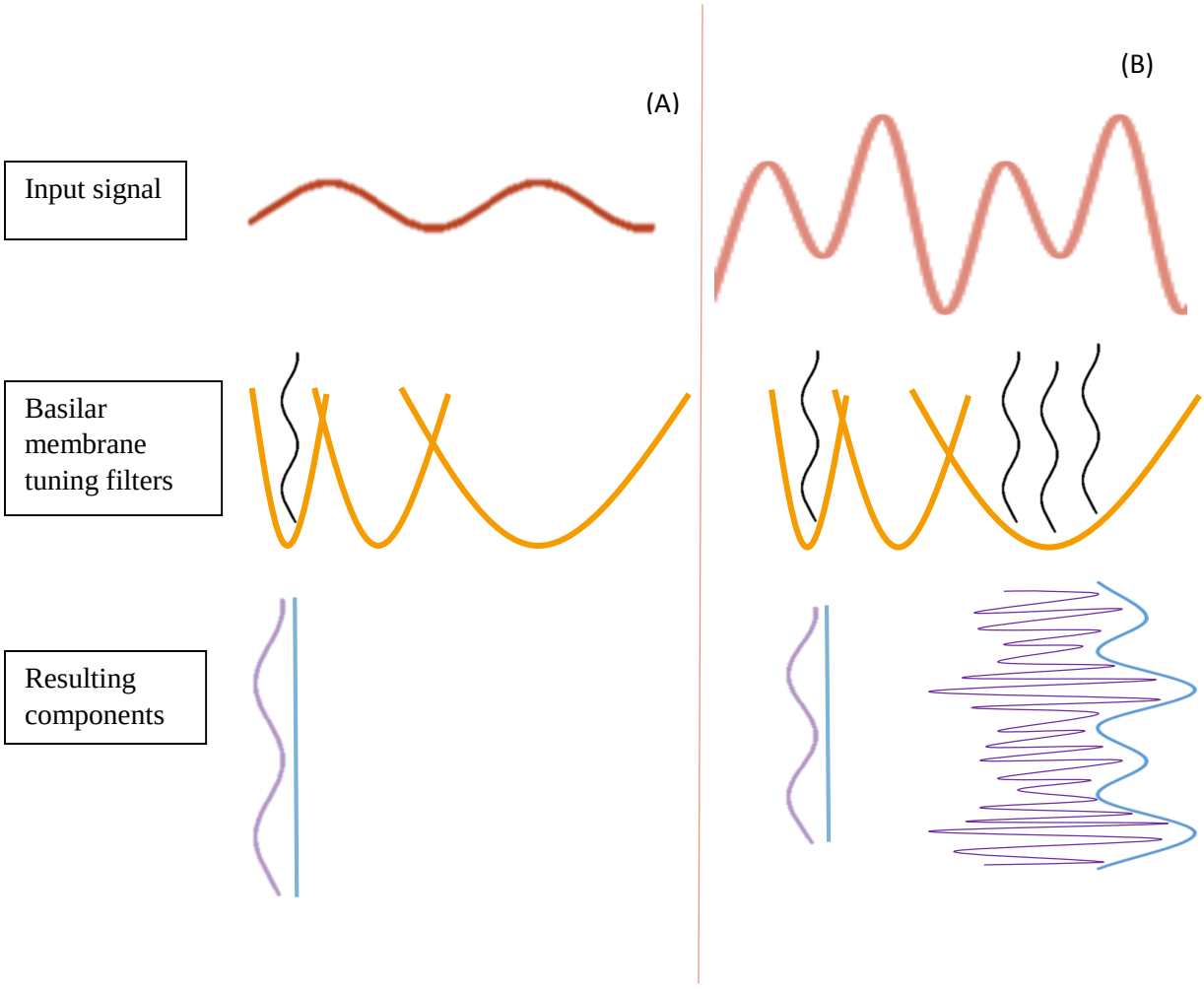

Figure 7. Incoming stimuli shown in red, overlapping cochlear filters (orange) and the resulting stimulus waveforms. TFS shown in purple; spectral envelope shown in blue. Low frequency components are shown (resolved) on the left side, higher frequency components are shown (unresolved) on the right. Panel A shows a simple, pure tone stimulus, with its resolved component. Panel B shows a complex stimulus with resulting output, including one resolved component and unresolved components.

History of the FFR

At approximately 40 years old, the FFR is a fairly young electrophysiological response. The discovery of the FFR stemmed from studies exploring neural pitch encoding mechanisms in the animal model. The idea that neural response firing patterns could reflect the frequency of an incoming auditory stimulus was first described by Wever and Bray in 1930, in the cat model. Specifically, Wever and Bray (1930a) transmitted pure tone stimuli to the cat's ear, and measured the electrical response through electrodes placed on the auditory nerve. Results of this study revealed that the frequency of the neural response corresponded to the frequency of auditory stimulation (Wever & Bray, 1930a). Measured responses were found to accurately reproduce the frequency of the pure tone stimulus, which lead to further investigation as to whether this response was a distant measurement of the cochlear microphonic (CM) potential, or a separate evoked potential (Marsh & Worden, 1968; Moushegian, Rupert, and Stillman, 1973).

Further research in cats supported the existence of the frequency following response as a new electrophysiological potential. Marsh and Worden (1968) investigated separating the FFR from the CM in the cat model using simple, pure tone stimuli. Their research revealed that varying electrode placements in the cochlear nucleus resulted in different patterns of amplitude changes for the FFR compared to the CM (Marsh & Worden, 1968). Results were also shown to vary in the FFR compared to the CM after euthanization of the cat, as the FFR was eliminated post-euthanization and the CM remained for a significantly longer period of time (Marsh & Worden, 1968). From this data, it was determined that the FFR was a unique response that could be recorded at the level of the cochlear nucleus, and not related to the CM (Marsh & Worden, 1968).

By 1973, research by Moushegian, Rupert, and Stillman, demonstrated that this evoked potential could be recorded in humans using scalp electrodes. Moushegian and colleagues (1973) recorded the FFR from five normal hearing subjects, using pure tone stimuli ranging from 250 Hz to 2000 Hz. Results revealed neural responses with peaks corresponding to the period of the frequency of the auditory stimulus (Moushegian et al., 1973). For example, a 250 Hz stimulus resulted in a response with peaks occurring every 4 ms; a 500 Hz auditory stimulus resulted in a neural response with peaks every 2 ms (Moushegian et al., 1973). Further investigation into these findings was necessary to ensure validity of the response (Moushegian et al., 1973). Latencies of these FFR recordings were found to occur approximately 6 ms post-stimulus onset, which is longer than would be expected for the CM potential, or for artifact caused by an acoustical leak from the earphone (Moushegian et al., 1973). Moushegian and colleagues (1973) also investigated the effect of continuous white noise masking presented simultaneously with the sine wave tone bursts, which resulted in an inability to measure the FFR, indicating that audibility of the stimulus was imperative for measurement of the response.

Marsh and Worden (1968) also proposed that there are frequency limits at each level of the auditory pathway with the highest frequency limits at the level of the cochlear nucleus, and lower frequency limits at rostral structures. In cats, FFR recordings from the inferior colliculus were observable at frequencies of 1000 Hz and below (Marsh & Worden, 1968). However, in humans, the FFR was observable up through 2000 Hz (Moushegian et al., 1973). These limits of measuring the FFR lead to research regarding the neural generators of the response.

Neural Generators

The exact neural generator of the FFR is not fully known, though multiple experiments have investigated the site of origin (Gardi, Merzenich, & McKean, 1979; Smith, Marsh, & Brown, 1975; Snyder & Schreiner, 1984). As previously mentioned, FFR latencies measured from the human scalp were found to be approximately 6 ms. This gave some information regarding site of neural generation, as this latency corresponded to that of wave V of an ABR (Smith et al., 1975; Moushegian et al., 1973). Wave V of the ABR was identified as originating from the inferior colliculus (IC), with a latency of approximately 5-6 ms (Smith et al., 1975).

Smith and colleagues (1975) investigated FFR latencies directly from different brainstem nuclei in cats, in order to compare these latencies to scalp-recorded FFRs in both cats and humans. Latency, amplitude, and phase information obtained from this experiment lead to the theory that the scalp-recorded FFR originated from a single neural generator in the brainstem, likely the IC (Smith et al., 1975). In order to further investigate the IC as the underlying neural generator, Smith and colleagues (1975) also bilaterally cooled the IC in cats, and found reduction or elimination of the FFR recorded from the IC and from the scalp. In contrast, cooling of the medial superior olive did not have any effect on the FFR (Smith et al., 1975). From these experiments, it was determined that the IC was the primary neural generator of the scalp-recorded FFR (Smith et al., 1975).

Contrasting findings were established by Gardi, Merzenich and McKean (1979), who examined underlying neural activity using needle electrodes and ablation procedures in the auditory brainstem of cats. Through ablation and sectioning of the auditory brainstem nuclei,

it was found that multiple generators contribute to the scalp-recorded FFR in the cat, including cochlear nuclei, cochlea, and superior olivary nuclei (Gardi et al., 1979).

Further research in cats investigated the differences between the characteristics of the cochlear microphonic and the scalp-recorded FFR and the auditory nerve neurophonic (Snyder & Schreiner, 1984). Similarities in response to forward masking conditions, changes in stimulus intensity, and spectral composition have lead Snyder and Schreiner (1984) to categorize the scalp-recorded FFR with the auditory nerve neurophonic, measured directly from the auditory nerve, rather than with the cochlear microphonic.

Multiple anatomical sites have been cited as the origin of the FFR, but Marsh, Brown, and Smith (1974) have also showed the presence of two separate pathways for the transmission of the FFR though the auditory nervous system. One pathway follows the direct connection of auditory fibers to the lateral lemniscus (LL) and IC (Marsh et al., 1974). The second pathway involves synapses from the superior olivary complex to the LL and IC (Marsh et al., 1974).

Results of these studies lead us to believe that the FFR is neural in origin, likely resulting from primary contributions from the IC, as well as other auditory brainstem nuclei. Due to similarities observed between the auditory system of cats and that of humans, results of these experiments in cats can be generalized to help us understand the functioning of the human auditory nervous system.

Subject Parameters

As previously discussed, a variety of AEPs can be utilized to provide information regarding the functioning of the auditory system. As each AEP evaluates a different part or

function of the system, there are varying parameters related to subject characteristics, stimuli, recording, and equipment set-up that should be considered when testing.

Sleep state and attention. The goal for obtaining the most robust response for all AEPs is to maximize the signal to noise ratio (SNR) when possible (Picton, 2010). One way to reduce the background noise to improve SNR is to encourage subjects to sleep during testing (Picton, 2010). Some AEPs require subjects to be awake and alert during testing, or to respond to stimuli (Picton, 2010). The sustained portion of the frequency following response is reported as "insensitive to sleep and… general anesthesia" (Varghese, Bharadwaj, & Shinn-Cunningham, 2015). Although the FFR is not affected by sleep state, subjects are often encouraged to sleep during testing, in order to reduce EEG noise or muscle artifact (Skoe & Kraus, 2010). If subjects remain awake for testing, attention is usually directed away from the stimulus through watching a silent movie or reading a book (Skoe & Kraus, 2010).

Hoormann and colleagues (2000) conducted a series of experiments to specifically study the effects of attention on the amplitude and latency of the FFR. The first two experiments utilized multiple stimulus modalities, to investigate the effect of visual and somatosensory stimuli on FFR recordings (Hoormann et al., 2000). Their first experiment used auditory stimuli (two tone burst frequencies) and visual stimuli (two colors of flashing lights) (Hoormann, Falkenstein, & Hohnsbein, 2000). In this experiment, participants were instructed to press a button after the presentation of the target stimulus, which varied with each presentation block (Hoormann et al., 2000). A second experiment in this study used auditory, visual, and somatosensory stimuli, presented in pairs (Hoormann et al., 2000). Participants were instructed to attend to the first presentation of each pair, regardless of stimulus modality (Hoormann et al., 2000). Attention was only directed to the second

stimulus in each presentation pair if the modality matched that of the first stimulus (Hoorman et al., 2000). The following experiments focused on auditory stimuli only (Hoormann et al., 2000). The third experiment used exclusively auditory stimuli, presented monaurally or binaurally (Hoormann et al., 2000). Participants were instructed to attend to stimuli in one ear only, while ignoring the stimuli presented in the opposite ear, when present (Hoormann et al., 2000). The last experiment contained two different auditory stimuli presented monaurally, with masking noise presented to the opposite ear (Hoormann et al., 2000). In this experiment, participants were tasked with identifying the number of target stimuli in each block, which were deviant from the standard stimuli presented (Hoormann et al., 2000). The results of this study concluded no significant effect of attention on FFR amplitude (Hoormann et al., 2000). The fourth experiment resulted in shorter FFR latencies when attention was directed toward the stimulus, but the authors have concluded that this effect originated in the cochlea, rather than the brainstem (Hoormann et al., 2000). In general, the FFR is not affected by subject sleep or attention (Skoe & Kraus, 2010).

Age. A second factor that should be considered is subject age. Age is closely related to anatomical maturity. Anatomical maturity should be considered when testing any AEP in young infants and children (Picton, 2010). Latency, amplitude, and morphological changes are seen in some AEPs with maturation of the auditory system, and increases in nerve myelination (Picton, 2010). After approximately age 2 years, maturation of the auditory brainstem is complete, and recordings from the brainstem are expected to remain stable until later in life (Skoe, Krizman, Anderson, & Kraus, 2013). Older adults may also display differences in AEP recordings, specifically in latency and intensity values (Burkard & McNerney, 2009). As the FFR provides a window into neural pitch encoding abilities, and

older adults tend to have increased difficulty with pitch perception, significant research has focused on the effects of age on this response.

Skoe and colleagues (2013) examined the effects of age on the behavior of brainstem-originating AEPs, in a group of 586 subjects ranging in age from 3 months to approximately 72 years of age. FFR amplitudes were found to increase dramatically throughout the first 5-8 years of life, when maximum amplitude values were reached (Skoe et al., 2013). After this age group, amplitudes were found to decrease slightly through older adulthood (Skoe et al., 2013). This decrease is hypothesized to be the result of changes in peripheral auditory functioning (Parthasarathy, Lai, & Bartlett, 2016). It has been speculated that a reduction in functioning of cochlear hair cells and auditory neuron fibers may be responsible for the changes observed in AEPs in older adult populations (Parthasarathy et al., 2016). Research specifically focusing on FFR recordings comparing young adults to older adults found that older adults tended to have reduced phase-locking abilities, as shown by reduced periodicity of FFR waveforms (Clinard & Cotter, 2015). AEP research in older adults proves to be difficult, as the possible effect of hearing loss is a confounding factor (Burkard & McNerney, 2009).

Hearing status. In addition to age, hearing loss also has an effect on the measurement and recording of AEPs. Sensorineural hearing loss (SNHL) may cause changes in the amplitude, latency and morphology of auditory evoked potentials such as the ABR (Don & Kwong, 2009). In individuals with hearing loss, neural responses are reduced, as shown primarily by reduced periodicity in FFR recordings (Ananthakrishnan, Krishnan, & Bartlett, 2016; Daly, Roeser, & Moushegian, 1976; Plyler & Ananthanarayan, 2001). Hearing loss may also cause a reduction in phase-locking abilities, especially in stimuli that

vary across time (Plyler & Ananthanarayan, 2001). Phase-locking responses to both the envelope and TFS have been found to be reduced in individuals with SNHL compared to individuals with normal hearing (Ananthakrishnan et al., 2016).

Gender. Some research has also investigated differences seen in evoked potential responses between male and female subjects. Anatomically, females have shorter cochlea lengths compared to males, which results in shorter latencies and larger amplitudes for some AEPs (Burkard & McNerney, 2009). Earlier peaks, corresponding to the onset of the auditory signal, have been shown in both the ABR and the FFR of female subjects (Ahadi, Pourbakht, Jafari, Shirjian, & Jafarpisheh, 2014; Krizman, Skoe, & Kraus, 2012). For the FFR, stronger formant representation was observed in female subjects (Ahadi et al., 2014; Krizman et al., 2012). Results of some experiments show responses from females have larger amplitudes and greater encoding of the fundamental frequency, while other studies show no difference between males and females (Ahadi et al., 2014; Krizman et al., 2012). Although anatomical differences have been established, conclusive data on the effect of gender on FFR results remains to be seen.

Experience and training. In addition to biologic factors, subject experience and training related to pitch perception, such as music or language instruction, may have an effect on the FFR. Neural plasticity is seen in musicians and tonal language speakers from the level of the brainstem through the level of the cortex (Bidelman, Weiss, Moreno & Alain, 2014). Music or language training enhances neural encoding abilities, represented by stronger and more periodic FFR waveforms (Krishnan, Gandour, & Suresh, 2016).

A review of AEP responses in musicians described enhanced responses at the level of the brainstem through the cortex for individuals with musical training (Sanju & Kumar,

2016). Individuals with either vocal or instrumental music training have greater responses to single and combination tones, as shown by larger FFR amplitudes, than individuals without musical training (Lee, Skoe, Kraus, & Ashley, 2009). This study also found a trend of increased FFR amplitude correlating with increased number of years of musical training (Lee et al., 2009).

A study of musicians and non-musicians by Parbery-Clark and colleagues (2009) investigated differences in their FFRs in quiet and noise conditions. Results of this study found better phase-locking ability and larger amplitudes in musicians than non-musicians in background noise (Parbery-Clark, Skoe, & Kraus, 2009). These results correlated with better results for the musicians on behavioral measures of understanding speech in noise (Parbery-Clark et al., 2009).

In addition to musical training, perceptual differences in speech, such as are present in tonal languages, may enhance a listener's sensitivity to changes in pitch, at the level of the auditory brainstem before the signal reaches the auditory cortex (Krishnan & Gandour, 2009). When comparing native tonal language speakers to native non-tonal language speakers, results reveal stronger FFR recordings with better pitch perception for tonal language speakers (Krishnan, Xu, Gandour, & Cariani, 2005). Effects of music training are also seen in the recording of AEPs, as both brainstem and cortical measures are enhanced corresponding to the length of musical training (Krishnan et al., 2005).

A study by Bidelman and colleagues (2011) compared the FFR results of musicians, non-musicians, and native Chinese speakers. Using musical chords as stimuli, results of this study found enhanced FFRs for musicians with greater than 10 years of musical training and native speakers of Chinese (Bidelman, Gandour, & Krishnan, 2011). Both groups had

greater neural representation of the stimuli than non-musicians who were not native speakers of a tonal language (Bidelman et al., 2011). From this data, it has been hypothesized that combined effects of musical training and tonal language instruction will result in further increased neural encoding of stimuli (Bidelman et al., 2011).

Hidden hearing loss. There are some situations in which neural encoding of stimuli is reduced, rather than increased (Kujawa & Liberman, 2009). In cases of excessive exposure to loud sounds, temporary threshold shifts may result in a loss of cochlear neuron function, in the presence of intact inner and outer hair cells (Kujawa & Liberman, 2009). In the case of this hidden hearing loss, individuals tend to have normal audiometric thresholds, but report difficulty with suprathreshold speech understanding especially in the presence of background noise (Kujawa & Liberman, 2009). The FFR provides a window in to the neural functioning at the level of the brainstem that traditional audiometry cannot provide.

Stimulus Parameters

Stimulus complexity. As previously discussed, the FFR is able to faithfully encode frequency and timing information present in a stimulus (Skoe & Kraus, 2010). This feature of the FFR is observed in simple and complex stimuli with frequency content present below 1500 Hz (Skoe & Kraus, 2010). In simple pure tone stimuli, brainstem neurons phase-lock onto the stimulus, and the resulting response reflects the period of the pure tone (Skoe & Kraus, 2010). The same process occurs with more complex stimuli, and the FFR reflects fundamental frequency information as well as formant structure information (Skoe & Kraus, 2010). Complex speech stimuli are often used to elicit the FFR, as speech can provide practical information on an individual's neural phase-locking abilities (Skoe & Kraus, 2010).

Many studies have used consonant-vowel syllable stimuli, such as /dɑ/, in order to evoke the FFR (Skoe & Kraus, 2010). This syllable, which contains a stop consonant and a sustained vowel, can be analyzed in segments as an onset response similar to an ABR, and a sustained FFR measurement (Skoe & Kraus, 2010). Specifically, the sustained portion of this response provides information on speech encoding abilities at the level of the auditory brainstem (Skoe & Kraus, 2010). The FFR is also able to measure the brainstem's ability to phase-lock to stimuli with changing frequency content, such as pitch contours and diphthongs (Skoe & Kraus, 2010). This provides information on the capacity of the brain to encode discrete changes in auditory information (Skoe & Kraus, 2010). Depending on the experimental population and the question at hand, researchers may use different types of stimuli to evoke the FFR, including pure tones, vowels, syllables, or music notes, which may be manipulated using acoustic filtering or added distortion (Skoe & Kraus, 2010). In order to isolate and investigate variation seen amongst individuals, pure tone stimuli will be used for the present experiment. Pure tone stimuli will also allow for eventual comparison to audiometric thresholds in the clinic.

Frequency. The frequency range of the stimulus should be considered when choosing stimuli to elicit the FFR, as phase-locking abilities are limited at rostral levels of the auditory nervous system (Skoe & Kraus, 2010). FFR recordings are strongest when the fundamental frequency (F_0) of a speech signal is between 80 and 300 Hz (Skoe & Kraus, 2010). This range allows for inclusion of speech formants within the range of brainstem phase-locking ability (Skoe & Kraus, 2010). The upper limit of phase-locking at the level of the brainstem is 1500 Hz, and should be considered when choosing simple stimuli (Skoe & Kraus, 2010). In addition, phase-locking ability tends to decrease as this upper limit is approached (Skoe &

Kraus, 2010). In the current experiment, we will use low frequency stimuli, to ensure adequate ability of neurons to encode information contained in the stimuli.

Recording Parameters

Intensity. Stimuli used to elicit the FFR must be presented at supra-threshold levels in order to elicit a clear response (Skoe & Kraus, 2010). Stimuli must be 40-50 dB greater than an individual's audiometric threshold in order to elicit a clear FFR (Davis & Hirsh, 1976). Krishnan (2002) studied the effects of intensity on the FFR responses and found that in general, amplitudes increase as stimulus intensity increases, although not necessarily as a linear increase. Typical intensities used are 60-85 dB SPL (Skoe & Kraus, 2010). For this study, we will use a stimulus intensity of 80 dB SPL, in order to maximize visualization of the response.

Monaural vs. binaural stimulation. Binaural auditory stimulation is perceived as louder than monaural stimulation (Skoe & Kraus, 2010). Studies comparing ears for monaural stimulation have found differences between recordings when the right ear is stimulated compared to the left (Ballachanda, Rupert, & Moushegian, 1994; Skoe & Kraus, 2010). Ballachanda and colleagues (1994) found that differences between ear recordings varied depending on stimulus intensity, with right ear stimulation producing more robust responses at some frequencies, and left ear stimulation producing more robust responses at others (Ballachanda et al., 1994). From this information, they concluded that asymmetries in neural encoding are present at the level of the auditory brainstem (Ballachanda et al., 1994). Monaural right ear stimulation has been shown to result in stronger formant frequency representation, possibly due to stronger left hemisphere encoding (Hornickel, Skoe, & Kraus, 2008). For this experiment, we will use monaural stimulation in the right ear.

Stimulus polarity. Stimuli may be presented in rarefaction or condensation polarities, or alternating between the two. Adding opposite polarity recordings together will allow for visualization of the neural envelope response (Skoe & Kraus, 2010). Subtracting opposite polarity recordings will show the neural spectral response, or the neural TFS (Skoe & Kraus, 2010). Recording alternating polarities allows rarefaction and condensation to be recorded in the same block of time. We will record FFRs to alternating polarities.

Sweeps. As previously discussed, the goal of any electrophysiologic test is to minimize artifact noise in order to maximize SNR. A greater number of sweeps will generally allow for a lower noise floor, a greater SNR, and therefore a more robust response (Skoe & Kraus, 2010). FFR recordings to tonal stimuli can be reliably measured in as little as 1000-2000 sweeps (Skoe & Kraus, 2010). For FFR recordings to complex stimuli, 1000 to 6000 sweeps are recommended in order to obtain a reliable response (Skoe & Kraus, 2010). For this experiment, stimuli will be presented in blocks of 2000 sweeps.

Analysis window. When selecting the appropriate length of the analysis window, stimulus duration must be considered. The analysis window must be long enough to encompass the entire duration of the stimulus, in order to visualize the entire response (Skoe & Kraus, 2010). A pre-stimulus period and a post-stimulus period should also be included in the analysis window, in order to record baseline measurements (Skoe & Kraus, 2010). The post-stimulus window should extend approximately 10-50 milliseconds after the stimulus ends (Skoe & Kraus, 2010). Our analysis window will be set to view 0-300 milliseconds of the FFR recording, in order to capture the response to our 265-millisecond stimuli.

Electrode montage. To record the FFR, a one-channel or two-channel electrode montage can be used. For both recording setups, an active electrode is placed on the midline

(either at Cz or Fz), and at least one reference electrode is placed on the earlobes or mastoids (Ananthakrisnan et al., 2016; Skoe & Kraus, 2010). If used, the second recording channel measures from the active midline electrode to a separate reference electrode, which may be placed on the 7th cervical vertebra (C7) (Ananthakrishnan et al., 2016). A one-channel or two-channel electrode montage measuring from midline to earlobes, mastoid, or C7 is referred to as a vertical montage (Bidelman, 2015). A vertical electrode montage reflects activity from rostral generators (Galbraith, 1994). A second, less common, electrode montage involves measuring between the right and left mastoids or earlobes, and is referred to as a horizontal montage (Bidelman, 2015). A horizontal electrode montage reflects neural information generated in more caudal structures (Galbraith, 1994). All configurations must also contain a common ground electrode, placed elsewhere on the forehead, or on an unused earlobe (Ananthakrishnan et al., 2016; Skoe & Kraus, 2010). An example of electrode placement for a vertical two-channel recording setup, similar to the placement to be used in the current experiment, is shown in Figure 8.

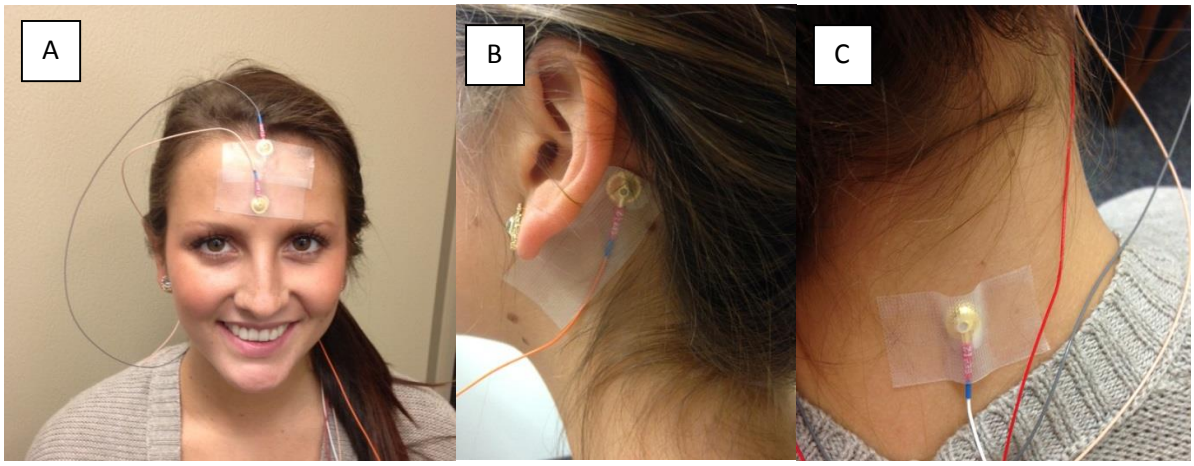

Figure 8. Depiction of electrode placement for a two-channel vertical recording montage. Panel A shows the common ground electrode on the lower forehead, as well as the active electrode placed at Fz. Panel B shows an inverting electrode placed on the mastoid. Panel C shows placement of an inverting electrode on the 7th cervical vertebra (C7).

What Information Does the FFR Provide?

The FFR provides valuable information regarding auditory brainstem function. Multiple studies have established that the FFR can be recorded in response to pure tone, speech, and noise stimuli. The underlying neural activity generating the FFR in response to auditory signals of varying complexity provides information on how the brainstem processes and encodes the varying acoustic characteristics contained in these stimuli (Bidelman 2015; Skoe & Kraus, 2010). As previously discussed, the FFR reflects neural phase-locking to acoustic features such as frequency in pure tone stimuli, and the fundamental and formant frequencies in complex stimuli. It is well-known that auditory brainstem neurons are capable of phase-locking to stimulus frequencies up to ~1500 Hz (Skoe & Kraus, 2010). For lower frequency stimuli, brainstem neurons are capable of phase-locking to both the neural envelope (representing lower frequency components of the stimulus such as the stimulus F0) as well as the TFS (representing higher frequency components of the stimulus such as the formant frequencies) (Skoe & Kraus). However, as stimulus frequency increases, and brainstem neural phase-locking reaches its limits, the neurons generating the FFR are unable to fire at the short intervals corresponding to the period of higher frequencies (Skoe & Kraus, 2010). In these situations, brainstem neurons phase-lock primarily to the lower frequency neural envelope, which typically represents lower frequency components of the stimulus such as the stimulus F0 (Skoe & Kraus, 2010). For pure tone stimuli, the neural envelope gives a response at twice the stimulus frequency, while the TFS produces a response at the frequency of the stimulus.

Extraction of brainstem neural representation of the envelope (FFR_{ENV}) and TFS (FFR_{TFS}) involves the addition and subtraction of the FFR collected in opposing polarities.

Specifically, when using the Intelligent Hearing Systems data acquisition platform, the FFR recorded in alternating polarity can be split into two buffers representing the FFR collected in condensation and rarefaction polarities. An example of an FFR recorded in alternating polarity in response to a 250 Hz pure tone stimulus is shown in Figure 9. Splitting this recording into its component polarities (rarefaction and condensation) results in the waveforms seen in Figure 10 and Figure 11.

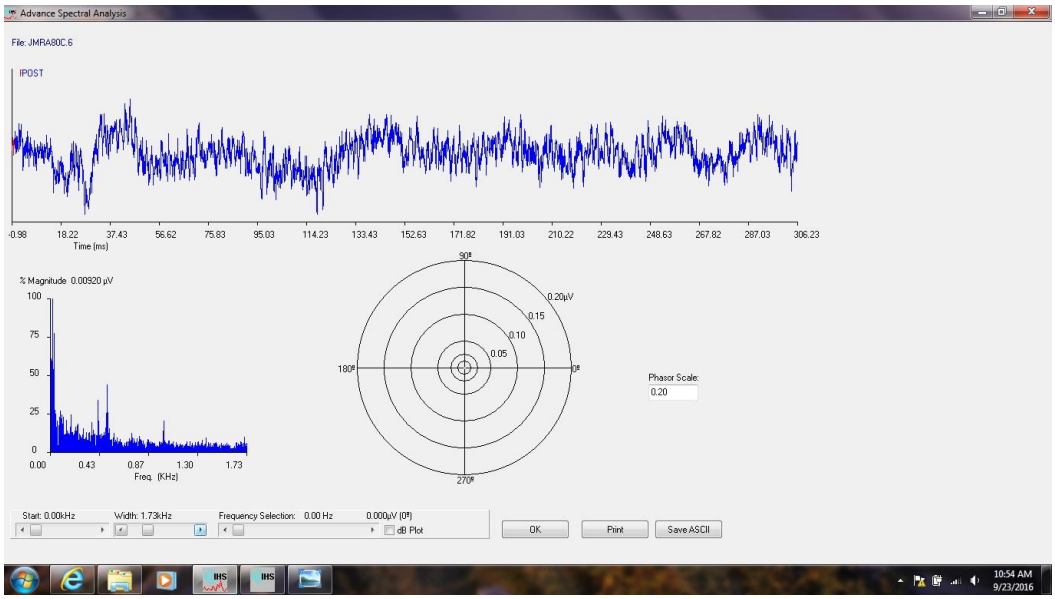

Figure 9. Example of an FFR waveform recorded to alternating polarity.

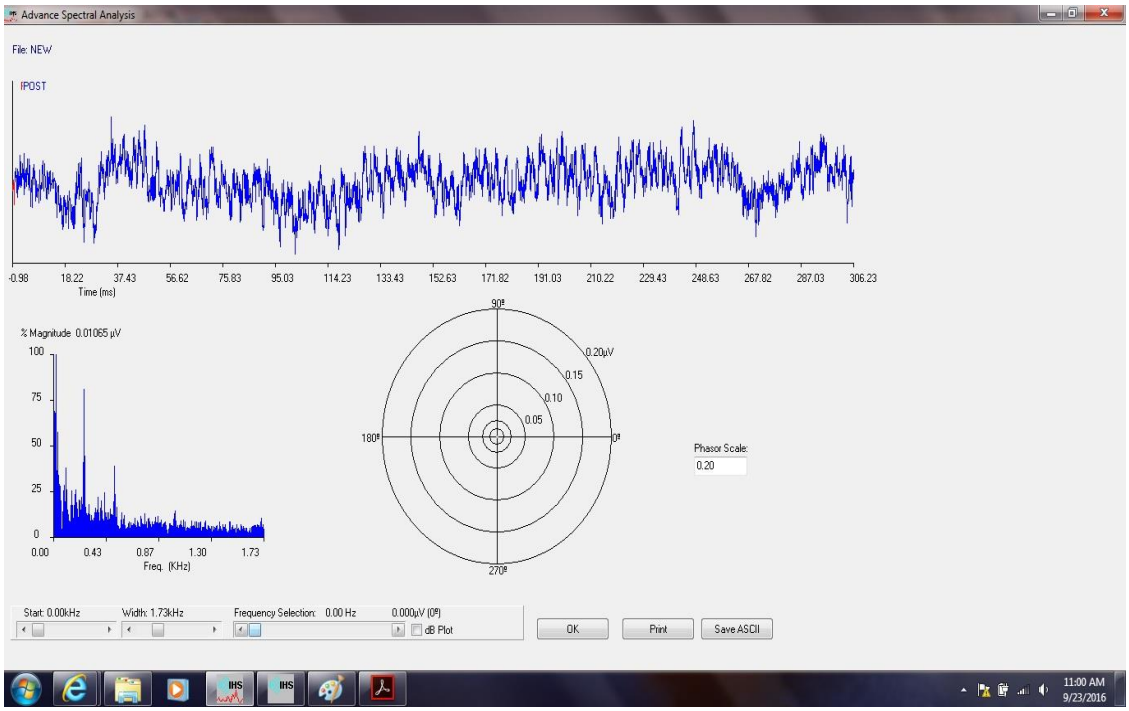

Figure 10. One polarity component (Buffer 1) of the FFR shown in Figure 9.

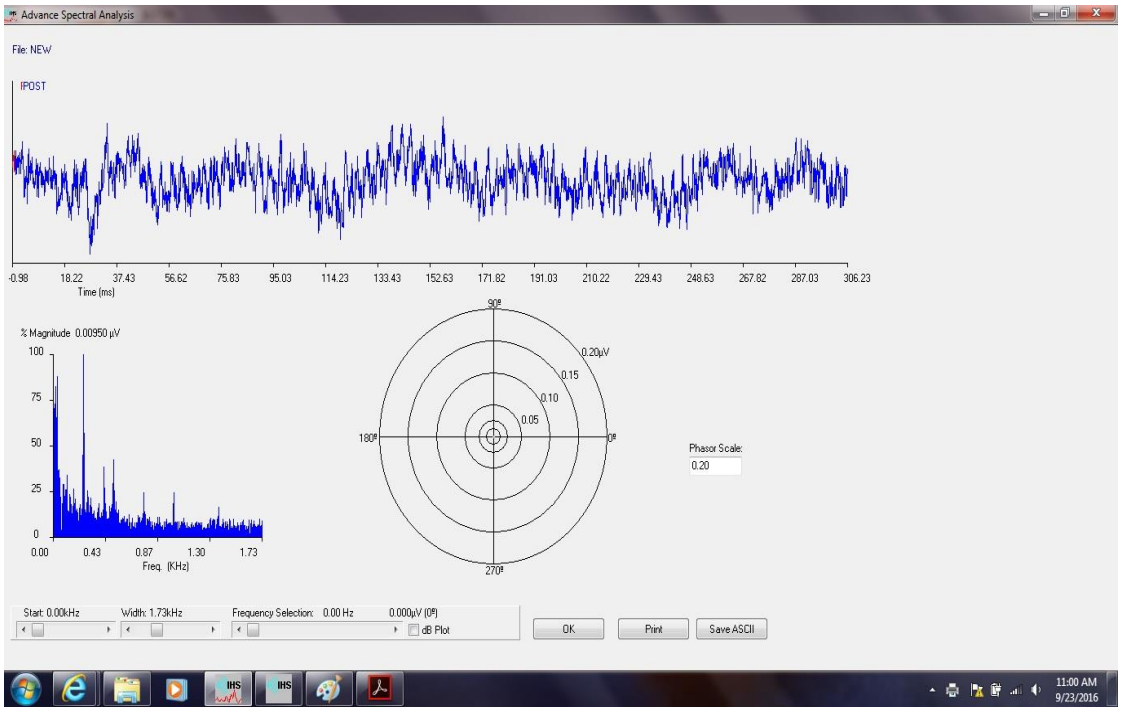

Figure 11. The second polarity component (Buffer 2) of the FFR shown in Figure 9.

Adding rarefaction and condensation polarities results in a waveform reflecting phase-locking to the neural envelope (Skoe & Kraus, 2010). An example of an FFR to the neural envelope is shown in Figure 12. The resulting FFT allows visualization of primary energy peaks of the response.

In contrast, subtraction of rarefaction and condensation polarities results in a waveform reflecting neural phase-locking to the TFS (Skoe & Kraus, 2010). An example of a response to TFS is shown in Figure 13. A predominant peak is seen in the FFT, at the fundamental frequency of the eliciting stimulus

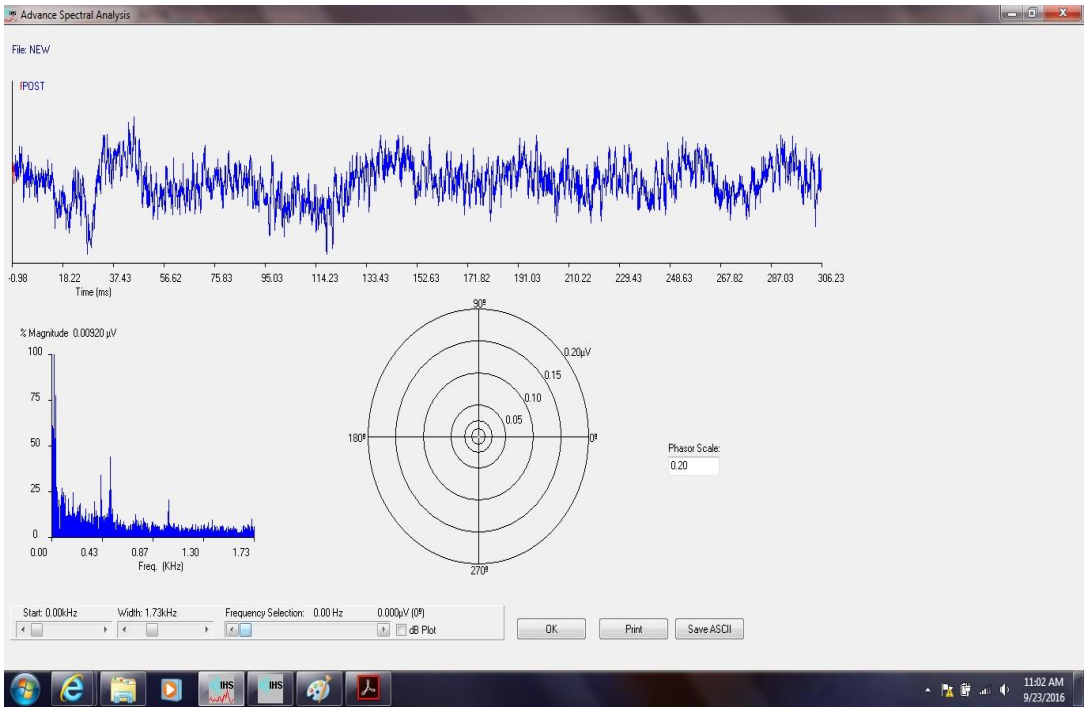

Figure 12. Depiction of an envelope response; created by adding rarefaction and condensation polarity recordings shown in Figure 10 and Figure 11.

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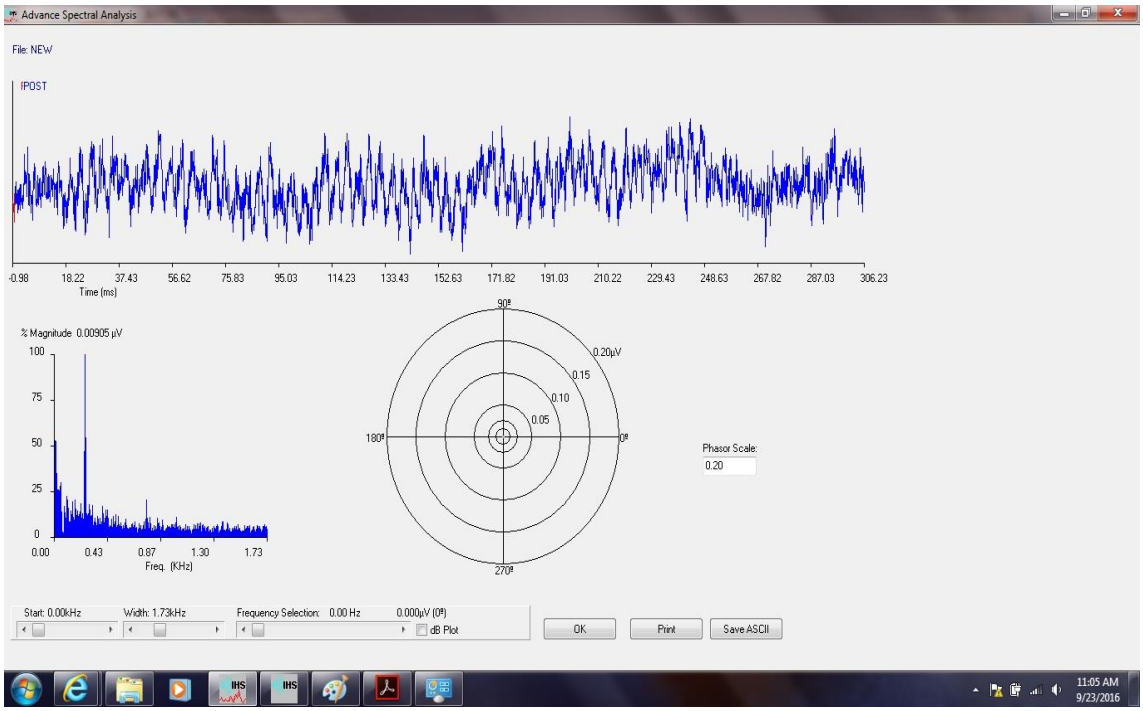


Figure 13. Depiction of phase-locking response to TFS; visualized by subtracting rarefaction and condensation polarities seen in Figure 10 and Figure 11.

Current Research

As discussed earlier, the FFR provides a window into how the brainstem processes pitch (Ananthakrishnan, et al., 2016; Skoe & Kraus, 2010). One of the puzzling aspects of the FFR is the tremendous variability seen in this response, even in a group of normal hearing adults (Ruggles, Bharadwaj, & Shinn-Cunningham, 2012). This variability may stem from a number of different reasons related to the subject, including experience- or training- induced plasticity, age, gender, "hidden hearing loss", or other unexamined factors (Skoe & Kraus, 2010). Additionally, review of prior FFR research indicates that the FFR is a versatile response in that it can be recorded in response to a variety of stimuli, ranging from simple, pure tone stimuli, to complex speech stimuli (Ananthakrishnan et al., 2016). As the FFR mimics the stimulus, the more complex the stimulus, the more complex the resultant FFR. Further, depending on the research question, the FFR can be analyzed in a number of different ways, including Fast Fourier Transform (FFT), spectral correlation, and phase coherence analysis techniques (Skoe & Kraus, 2010). When comparing across studies to paint a complete picture of the FFR, the subject-related variability of the FFR coupled with other sources of variability across studies (the wide range of stimuli, populations, and analysis techniques used by different researchers when recording the FFR) make it challenging to decipher this complex response. A potential way to better understand the nature of the FFR across subjects could be to minimize stimulus- and analysis-related variability, by measuring the strength of the neural response as reflected by spectral peaks in the FFT to simple stimuli such as pure tones in a group of normal-hearing adults.

Also, as previously discussed, the FFR has been used as an objective platform to study neural plasticity following music training (Skoe & Kraus, 2010). The effect of music

training on the FFR is well-established; stronger FFRs can be recorded in musicians v. non-musicians. However, these experience dependent effects have been noted only in response to certain stimuli. It is unclear if such training effects will be preserved in the neural response to non-musical stimuli such as pure tones. In this study, we investigate the effects of two subject variables: gender and music experience, on the strength of the FFR.

Hence, our aim for the current study is to collect FFRs in response to pure tones in listeners with normal hearing in order to examine subject-related variability, including musical training and gender, in brainstem neural encoding strength.

Methods

Participants

A total of thirty normal hearing adults (male = 12; female 18) between the ages of 19 and 29 years of age (mean age = 23.71, standard deviation = 2.15) with pure tone thresholds of 20 dB or better from 250 Hz through 8000 Hz bilaterally) were recruited to participate in the experiment. Participation was voluntary, and enrollment was based on the following pre-screening criteria: no reported or visible middle or outer ear deformity or disorder and no history of hearing loss. All participants were required to have normal middle ear function, defined by Jerger Type A tympanograms at the time of study enrollment. Participants completed a case history form prior to testing. On this case history form, if participants indicated any musical experience, their musical ability was assessed using a portion of the Goldsmiths Musical Sophistication Index, which can be found in Appendix A (Müllensiefen, Gingras, Musil, & Stewart, 2014). All participants gave informed consent in compliance with a protocol approved by the Institutional Review Board at Towson University. Participants were compensated $20 in the form of a gift card.

Stimuli

FFRs were elicited by pure tone stimuli with frequencies corresponding to audiometric octaves (250, 500, and 1000 Hz). These frequencies were chosen because they are below the upper limits for phase-locking in the brainstem, and correspond to measurable audiometric octave frequencies. Stimulus duration were 265 milliseconds. FFRs were recorded at 80 dB SPL to alternating polarity stimuli presented at a repetition rate of 3.13 per second. Stimuli were presented to the right ear through magnetically shielded ER-3A insert earphones. Stimulus frequency presentations were randomized across participants. Stimuli

were generated and recorded using an auditory evoked potential signal generation platform (Smart EP, Intelligent Hearing Systems [IHS], Miami, FL).

Recording

Participants sat in a recliner, and were instructed to sleep or relax and refrain from extraneous movement during testing. FFRs were recorded using a two-channel vertical electrode configuration. The positive (non-inverting) electrode was placed at Fz, on the upper forehead. Right and left mastoids (A2 and A1, respectively) served as the reference (inverting) sites for channel 1. The second channel recorded from the Fz electrode to a reference (inverting) electrode placed on cervical vertebra 7 (C7). An additional electrode was placed in the middle of the forehead (Fpz) to serve as the common ground. Electrode impedances measured below 3 kOhms. Response waveforms were obtained as averages to 2000 stimulus sweeps, using a 300-millisecond post stimulus analysis window. Testing took approximately 90 minutes.

Analysis

After FFR waveforms were obtained to alternating polarity stimuli, the buffers were split in order to visualize the rarefaction and condensation components. As previously discussed, addition of these components resulted in the FFR_{ENV} and subtraction resulted in the FFR_{TFS} (Ananthakrishnan et al., 2016). For the purpose of this study, analysis was performed on the subtracted waveforms, or FFR_{TFS} only. Fast Fourier Transform (FFT) analysis using MATLAB was used to measure the magnitude of the tallest spectral peak in each subject's response for each stimulus condition. Figures representing grand average waveforms and FFTs for each stimulus condition were generated.

Statistical Analyses

An analysis of variance (ANOVA) was performed to explore the variations seen in the FFR in response to all three stimulus frequencies (250, 500 and 1000 Hz). The independent variable was stimulus frequency, and the dependent variable was peak FFR amplitude. Next, the potential effect of musical training and gender was examined on the strength of FFR in response to just the 250 Hz pure tone. A two-way ANOVA was used to describe the effects of these subject variables on the FFR. The independent variables were musical experience (0 years, 1-5 years, or 6+ years) and gender (male or female). The dependent variables were peak FFR amplitude for the 250 Hz tone condition. Statistical analysis and figure generation were performed using SPSS Statistical Analysis Software and MATLAB.

Results

Grand averaged FFR response waveforms for all participants at each stimulus frequency (250 Hz, 500 Hz, and 1000 Hz) are presented in Figure 14. Overall response amplitude and periodicity decrease with increasing stimulus frequency. Grand averaged FFT responses for all participants at each stimulus frequency are presented in Figure 15. As can be seen, peak FFT amplitudes were greatest for the 250 Hz stimulus (in blue), and decrease with increasing frequency. Mauchly's Test of Sphericity indicated that the assumption of sphericity had been violated, $\chi^2(2) = 10.56$, $p = 0.005$, and therefore, a Greenhouse-Geisser correction was used. There was a statistically significant effect of frequency on the FFT amplitude, F (1.52,44.14) = 76.2, p <0.001. Bonferroni post-hoc comparisons revealed significantly larger amplitudes for 250 Hz compared to 500 Hz ($p < 0.001$) and 1000 Hz ($p < 0.001$), as well as 500 Hz compared to 1000 Hz ($p < 0.001$), which can be visualized in Figures 14, 15, and 16.

To investigate the effect of musical training on the FFR, participants were split into three groups: individuals who reported no musical training, individuals with 1-5 years of training, and individuals with 6 or more years. Here, we focus on the results of the FFR recorded to the 250 Hz stimulus only. A two-way ANOVA revealed no significant effect of musicianship (F (2, 24) = 0.132, $p = 0.877$) or gender (F (1, 24) = 0.46, $p = 0.504$) on FFT amplitude. Additionally, no significant interaction effects of gender and musicianship were seen, F (2, 24) = 0.888, $p = 0.425$. Figure 17 displays mean peak FFT amplitudes for males and females at each level of reported musical training; large variability is seen between groups. Table 1 displays mean peak FFT values for each of these groups.

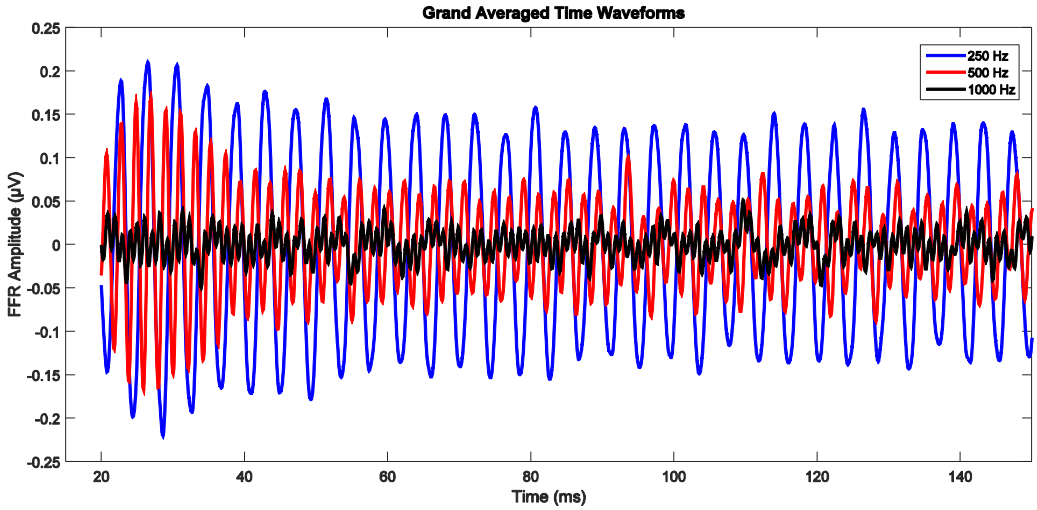


Figure 14. Grand averaged time waveforms for each of the 3 stimulus frequencies: 250 Hz (blue), 500 Hz (red), and 1000 Hz (black).

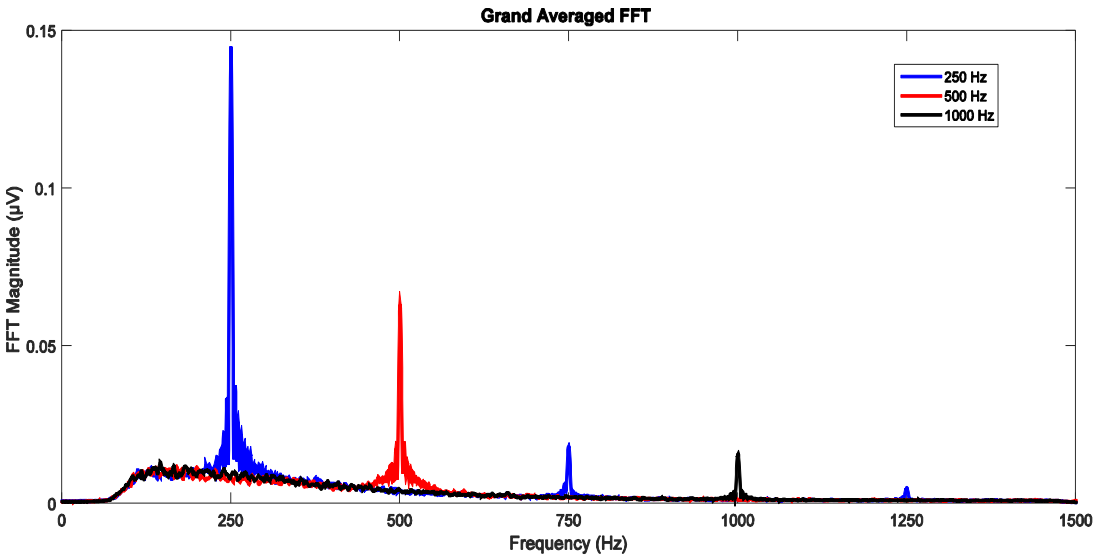


Figure 15. Grand averaged FFT for each of the 3 stimulus frequencies: 250 Hz (blue), 500 Hz (red), and 1000 Hz (black).

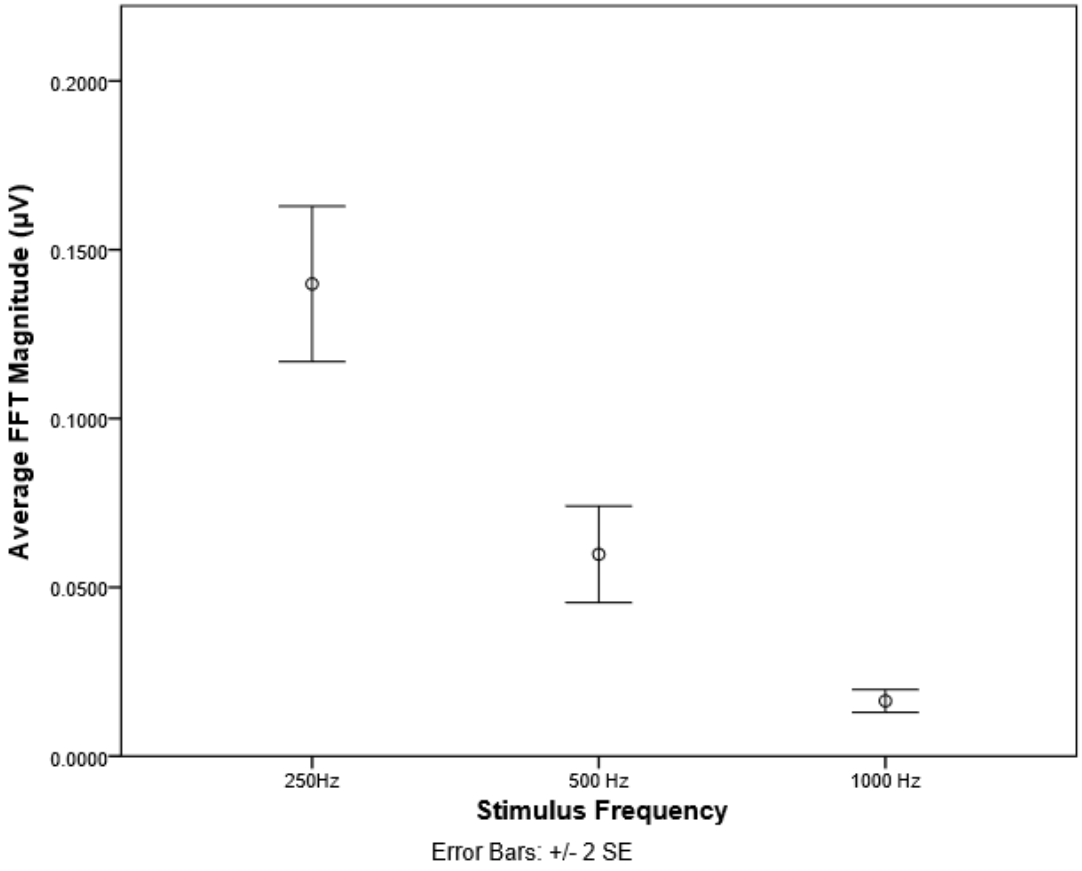


Figure 16. Mean peak FFT amplitudes for each stimulus frequency, for all participants. Amplitude decreases with increasing stimulus intensity.

Table 1

FFT Amplitudes by Gender and Level of Musical Experience

	Mean FFR Amplitude (μV)	Standard Error
Females		
No musical Experience	0.120	0.023
1-5 Years Musical Experience	0.148	0.038
6+ Years Musical Experience	0.173	0.025
Males		
No musical Experience	0.143	0.029
1-5 Years Musical Experience	0.126	0.033
6+ Years Musical Experience	0.120	0.038

Note. Mean FFR amplitudes and standard error presented for varying levels of musical experience, split by gender.

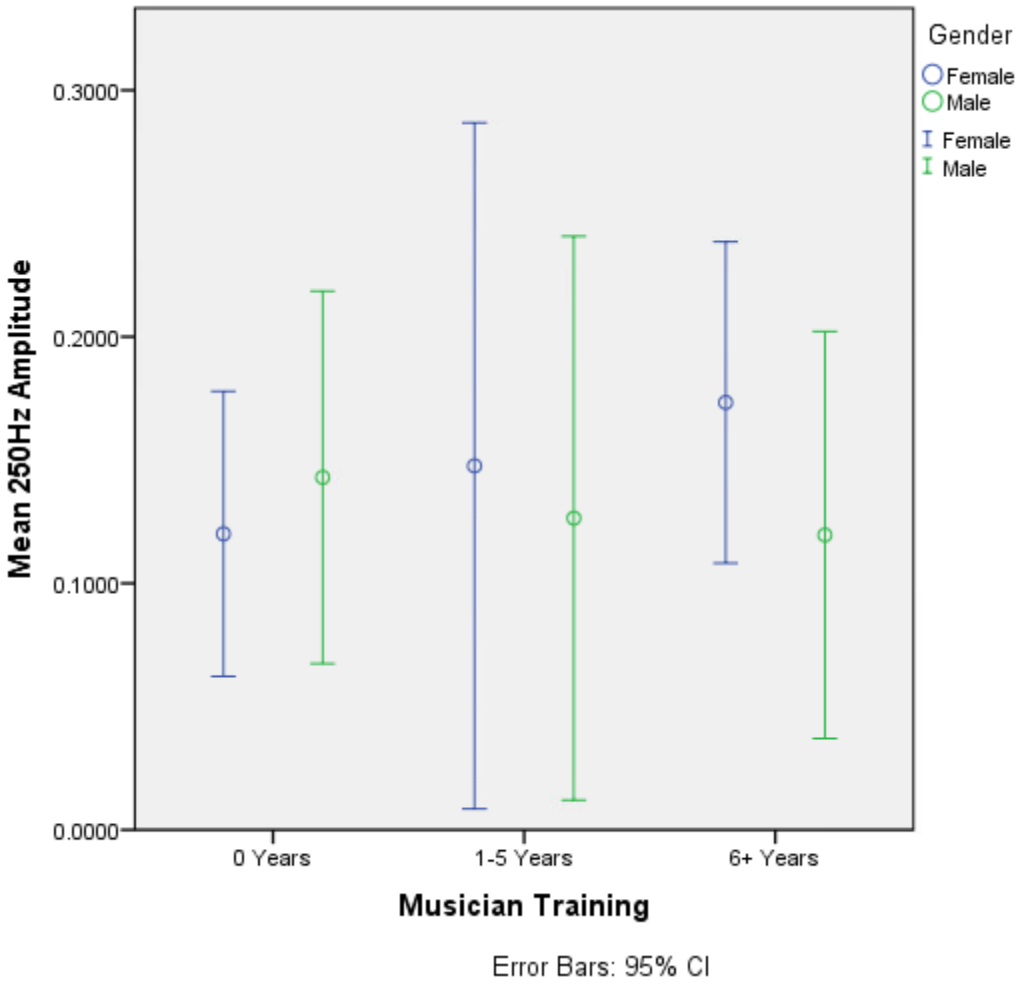


Figure 17. Mean FFT amplitudes (in μV) for each level of musical training, separated by gender. No significant effects of musical training or gender were seen in the current study. Error bars represent 95% confidence interval around the mean.

Discussion

The goal of the current study was to investigate possible differences due to subject-related factors such as age and musicianship in the pure-tone evoked FFR obtained in individuals with clinically normal hearing thresholds. No significant effects of musical training or gender were seen on the FFT amplitudes of the FFR recorded in response to a 250 Hz pure tone stimulus.

Effect of Frequency

Significant effects of stimulus frequency were observed in the current study, such that FFR strength decreased with increase in stimulus frequency. These findings are not unexpected, and are consistent with the FFR literature (Aiken & Picton, 2008; Picton, 2008). The FFR is known to decrease in strength as frequency increases, through approximately 1500 Hz, as it reflects neural phase locking abilities at the level of the auditory brainstem (Aiken & Picton, 2008; Skoe & Kraus, 2010). Based on the established phase-locking limits at the auditory brainstem, we expect the strongest neural encoding to occur at lowest frequencies (Skoe & Kraus, 2010). Our results were consistent with the literature in this case, as the FFR amplitudes were greatest for the 250 Hz stimulus condition, and lowest for the 1000 Hz condition. We did not measure stimuli above 1500 Hz, as neural phase-locking abilities would be limited beyond this point (Skoe & Kraus, 2010).

Effect of Gender

When responses for the 250 Hz stimulus were analyzed by gender, no significant differences were seen between males and females. The existing literature has not conclusively determined if gender effects are reflected in the FFR (Ahadi et al., 2014; Krizman et al., 2012). Anatomical differences have noted between males and females, with

females having shorter cochlea lengths, smaller head size, and less brain volume, and therefore may have shorter latencies and larger amplitudes for some AEPs (Ahadi et al., 2014; Burkard & McNerney, 2009). However, the effect of these anatomical differences has not been fully established for the FFR. Additional potential causes of gender differences include presence of estrogen, differences in body temperature, and ear canal size (Ahadi et al., 2014). Gender-related variations are limited for the sustained portion of the FFR, which we analyzed in the current study, rather than onset latencies (Ahadi et al., 2014; Krizman et al., 2012). The results of the current study do not indicate significant differences in FFR amplitudes between males and females, likely because we focused on the sustained portion of the response. Further analysis of onset latencies may reveal differences between males and females. Further research is needed to determine if effects of gender are reflected in FFR recordings to various stimuli.

Effect of Musical Training

Participants were separated according to self-reported level of musical training: no experience (0 years), 1-5 years of formal training, and 6 or more years. No significant effects of musical training were seen on brainstem neural representation of the 250 Hz pure tone stimulus. Large variability in FFR amplitudes was seen within each level of musical experience. Prior research indicates that individuals with musical training have enhanced neural encoding of pitch, evidenced by larger FFR amplitudes and more periodic FFR waveforms (Lee et al., 2009; Krishnan et al., 2016). Some studies have shown increased FFR amplitudes correlated with increased years of musical training, however our data does not support this trend (Lee et al., 2009). Although our data are inconsistent with the current literature regarding enhanced brainstem neural encoding in musicians, prior research utilized

more complex musical stimuli (Lee et al., 2009). It is possible that enhanced FFR waveforms in musicians is limited to musical stimuli, and cannot be generalized to pure tone stimuli. It is also possible that the sample size of the current study was a cause of the lack of effect of musical training. Participants were divided based on number of years of musical training, which resulted in group sizes of 7 to 13 participants, which may have affected statistical effects seen.

Additionally, differences in our results may be the result of the nature of the questionnaire, which counted total years of formal musical training regardless of how long ago this training took place. Further detail regarding musical training and the recentness of this training, could help establish a relationship between experience and the FFR. Further investigation into the effect of musical training on the FFR could consider measuring the length of time between musical training and FFR measurement, as well as using various stimulus types, including pure tones.

Future Directions

In the current small-scale study, we only investigated the effects of overall musical training and gender on FFR amplitude in a limited number of subjects. Our study did not show significant effects of musical training or gender on the amplitudes of FFRs to a 250 Hz pure tone stimulus. Although these data provide useful information regarding the impact of gender and musical training on brainstem neural representation of pitch as indexed by the FFR, further research is needed in this area. We cannot utilize the FFR in the clinical setting until we more fully understand the subject-related factors that influence this response. There are a vast number of variables, aside from gender and musical training, which can impact the response seen in the FFR, including hearing status, age, and training in tonal languages.

Future studies could consider further auditory testing in an attempt to correlate the FFR to measurements of hearing sensitivity. Additional testing into auditory system functioning could include extended high frequency pure tone testing, ABRs, otoacoustic emissions (OAEs), and/or speech perception testing. These behavioral and objective tests would provide more information on auditory ability from both the peripheral and central nervous systems, allowing possible correlation with FFR results, and a greater understanding of the effects of the functioning of auditory brainstem neurons.

Appendix A

Musical Questionnaire

Questionnaire adapted from:

Müllensiefen, D., Gingras, B., Musil, J., & Stewart L. (2014). The Musicality of Non-Musicians: An Index for Assessing Musical Sophistication in the General Population. PLoS ONE, 9(2): e89642. doi:10.1371/journal.pone.0089642

Please circle the most appropriate category:

1. I engaged in regular, daily practice of a musical instrument (including voice) for 0 / 1 / 2 / 3 / 4-5 / 6-9 / 10 or more years.

2. At the peak of my interest, I practiced 0 / 0.5 / 1 / 1.5 / 2 / 3-4 / 5 or more hours per day on my primary instrument.

3. I have played or sung in a group, band, choir, or orchestra for 0 / 1 / 2 / 3 / 4-5 / 6-9 / 10 or more years.

4. I have attended 0 / 1 / 2 / 3 / 4-6 / 7-10 / 11 or more live music events as an audience member in the past twelve months.

5. I have had formal training in music theory for 0 / 0.5 / 1 / 2 / 3 / 4-6 / 7 or more years.

6. I have had 0 / 0.5 / 1 / 2 / 3-5 / 6-9 / 10 or more years of formal training on a musical instrument (including voice) during my lifetime.

7. I can play 0 / 1 / 2 / 3 / 4 / 5 / 6 or more musical instruments.

8. I listen attentively to music for 0-15 min / 15-30 min / 30-60 min / 60-90 min / 2 hrs / 2-3 hrs / 4 hrs or more per day.

9. The instrument I play best (including voice) is _____ / Not applicable.

Case History Form

Frequency Following Response (FFR): Examining the Variability of This Complex Response

Principal Investigator: Laura Grinstead, B.S.

Case History Form

Age: _____

Do you have any concerns about your hearing? (Circle)	Yes	No
Have you ever seen a doctor who specializes in ears? (Circle)	Yes	No
Do you ever have difficulty understanding speech? (Circle)	Yes	No
Have you ever had your hearing tested? (Circle)	Yes	No
-If yes, what were the results? _____		
Do you have a history of ear infections? (Circle)	Yes	No
Do you have tinnitus (ringing, roaring, hissing, buzzing in your ears?) (Circle)	Yes	No
Do you have otalgia (ear pain)? (Circle)	Yes	No
Do you have any vertigo/disequilibrium? (Circle)	Yes	No
Do you have a history of any noise exposure? (Circle)	Yes	No
-If yes, did you wear hearing protection? (Circle)	Yes	No
Are you currently taking any medications? (Circle)	Yes	No
-If yes, please list: _____		
Have you had any musical training? (Circle)	Yes	No
-If yes, please see attached form		
Have you had instruction in languages other than English? (Circle)	Yes	No
-If yes, please list language(s) and number of years of instruction:		
_____		

Informed Consent Form

INFORMED CONSENT FORM

Project Title: Frequency Following Response (FFR): Examining the Variability of this Complex Response

PRINCIPAL INVESTIGATOR: _Laura Grinstead, B.S._____ PHONE: _302-299-6867_____

Purpose of the Study:
This study is designed to study neural representation of sounds in the brain and brainstem in adult listeners with normal hearing, with and without music training. The specific neural response being evaluated in this study is known as the Frequency Following Response (FFR).

Procedures:
Participants will complete a detailed case history form and questionnaire, with questions relating to hearing ability and musical training. All participants will undergo a comprehensive hearing test, which will include tests from the standard clinical audiology test battery. Brainstem neural responses in response to various auditory signals (known as the Frequency Following Response (FFR)) will be collected using sensors (electrodes) placed on the scalp. The investigator will place five electrodes at different spots on the head (two on the forehead, one behind each ear, and one on the nape of the neck). A soft foam insert earphone will be placed in the right ear. Participants will be asked to lay quietly or sleep in a comfortable recliner for the duration of the test session, while a tonal stimulus is delivered to the right ear. Testing will be completed in two test sessions (first session: 45 minutes; second session: 90 minutes).

Risks/Discomfort:
There are no known risks associated with participation in the study. Possible sources of minimal discomfort include removal of the medical grade adhesive tape used to secure the electrodes, and discomfort of insert earphone in the right ear. You may choose to discontinue participation at any time.

Benefits:
It is hoped that the results of this study will have beneficial effects to establish normative data information for the FFR, and to learn more about brainstem neural representation of auditory stimuli.

Alternatives to Participation:
Participation in this study is voluntary. You are free to withdraw or discontinue participation at any time. Refusal to participate in this study will in no way affect your class standing.

Cost Compensation:
Participation in this study will involve no costs or payments to you.

Confidentiality:
All information collected during the study period will be kept strictly confidential. You will be identified through identification numbers. No publications or reports from this project will include identifying information on any participant. If you agree to join this study, please sign your name below.

_____ I have read and understood the information on this form.

_____ I have had the information on this form explained to me.

_____ _____
Subject's Signature Date

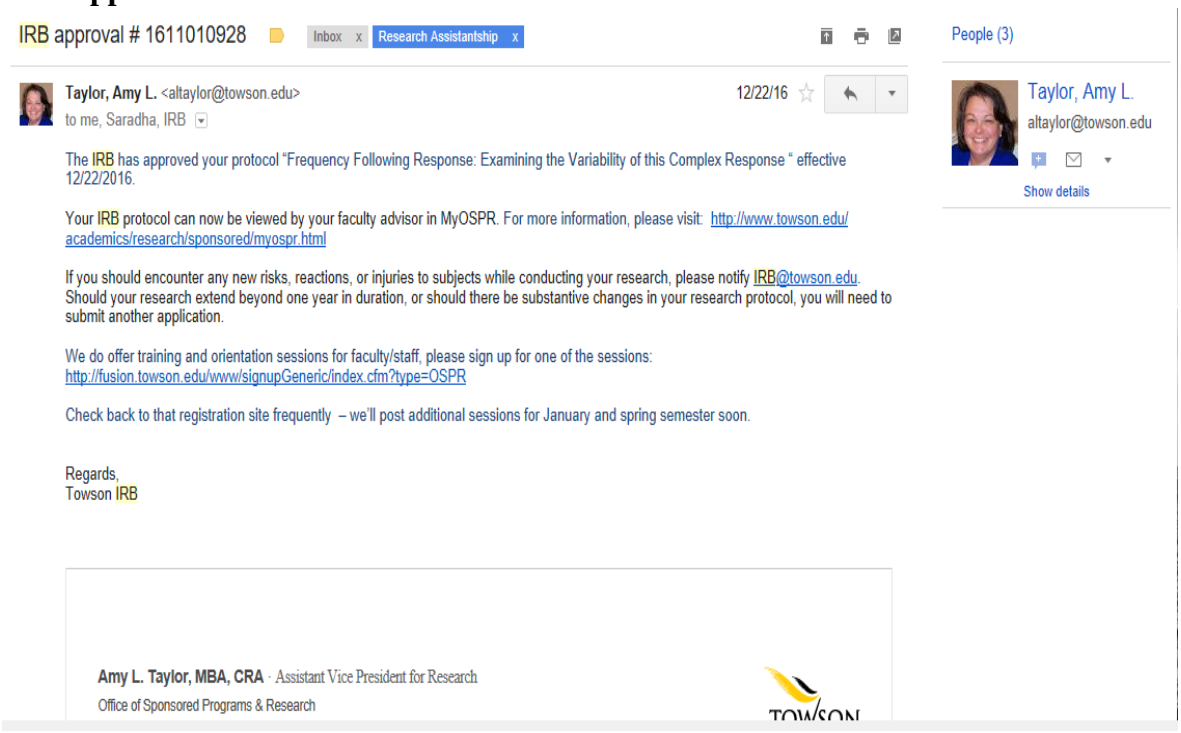
_____ _____
Principal Investigator Date

If you have any questions regarding this study please contact Dr. Saradha Ananthakrishnan, the faculty advisor, at 410-704-6369; or the Institutional Review Board Chairperson, Professor Elizabeth Katz, Office of University Research Services, 8000 York Road, Towson University, Towson, Maryland 21252; phone (410) 704-2236.

THIS PROJECT HAS BEEN REVIEWED BY THE INSTITUTIONAL REVIEW BOARD FOR THE PROTECTION OF HUMAN PARTICIPANTS AT TOWSON UNIVERSITY

Appendix B

IRB Approval



IRB approval # 1611010928 Inbox x Research Assistantship x

Taylor, Amy L. <altaylor@towson.edu> 12/22/16

to me, Saradha, IRB

The IRB has approved your protocol "Frequency Following Response: Examining the Variability of this Complex Response " effective 12/22/2016.

Your IRB protocol can now be viewed by your faculty advisor in MyOSPR. For more information, please visit: http://www.towson.edu/academics/research/sponsored/myospr.html

If you should encounter any new risks, reactions, or injuries to subjects while conducting your research, please notify IRB@towson.edu. Should your research extend beyond one year in duration, or should there be substantive changes in your research protocol, you will need to submit another application.

We do offer training and orientation sessions for faculty/staff, please sign up for one of the sessions: http://fusion.towson.edu/www/signupGeneric/index.cfm?type=OSPR

Check back to that registration site frequently – we'll post additional sessions for January and spring semester soon.

Regards,
Towson IRB

Amy L. Taylor, MBA, CRA · Assistant Vice President for Research
Office of Sponsored Programs & Research

TOWSON

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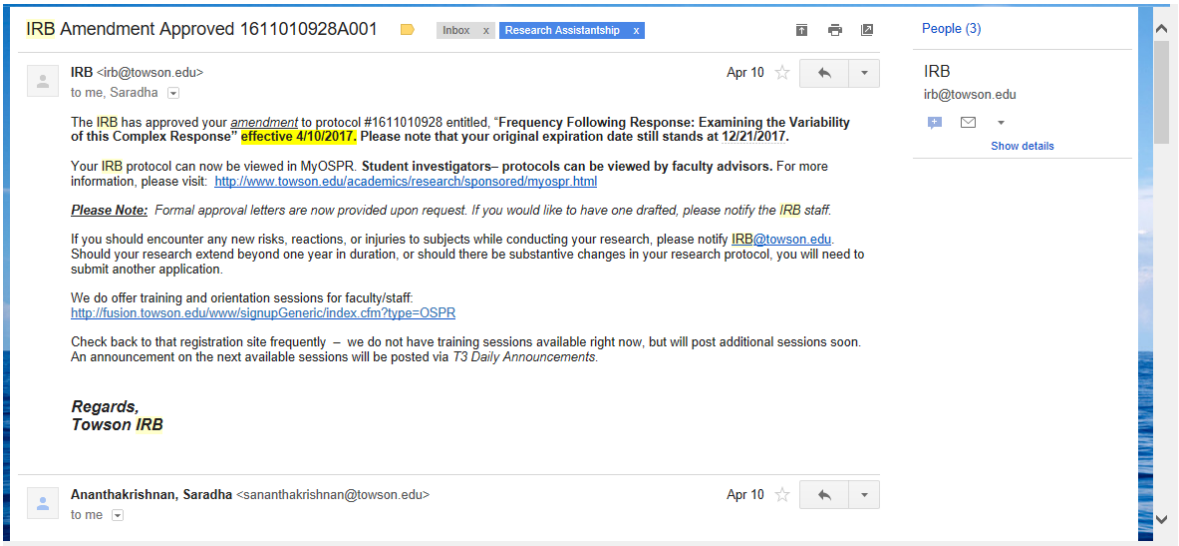
Taylor, Amy L.
altaylor@towson.edu

Show details

IRB Amendment Approval



IRB Amendment Approved 1611010928A001 Inbox x Research Assistantship x

IRB <irb@towson.edu> Apr 10
to me, Saradha

The IRB has approved your *amendment* to protocol #1611010928 entitled, "**Frequency Following Response: Examining the Variability of this Complex Response" effective 4/10/2017. Please note that your original expiration date still stands at 12/21/2017.**

Your IRB protocol can now be viewed in MyOSPR. **Student investigators– protocols can be viewed by faculty advisors.** For more information, please visit: http://www.towson.edu/academics/research/sponsored/myospr.html

Please Note: *Formal approval letters are now provided upon request. If you would like to have one drafted, please notify the IRB staff.*

If you should encounter any new risks, reactions, or injuries to subjects while conducting your research, please notify IRB@towson.edu. Should your research extend beyond one year in duration, or should there be substantive changes in your research protocol, you will need to submit another application.

We do offer training and orientation sessions for faculty/staff:
http://fusion.towson.edu/www/signupGeneric/index.cfm?type=OSPR

Check back to that registration site frequently – we do not have training sessions available right now, but will post additional sessions soon. An announcement on the next available sessions will be posted via *T3 Daily Announcements*.

Regards,
Towson IRB

Ananthakrishnan, Saradha <sananthakrishnan@towson.edu> Apr 10
to me

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References

Ahadi, M., Pourbakht, A., Jafari, A. H., Shirjian, Z., & Jafarpisheh, A. S. (2014). Gender disparity in subcortical encoding of binaurally presented speech stimuli: An auditory evoked potentials study. *Auris Nasus Larynx, 41*(3), 239-243. doi: 10.1016/j.anl.2013.10.010.

Aiken, S. J. & Picton, T. W. (2008). Envelope and spectral frequency-following responses to vowel sounds. *Hearing Research, 245*, 35-47. doi: 10.1016/j.heares.2008.08.004.

Ananthakrishnan, S., Krishnan, A., & Bartlett, E. (2016). Human frequency following response: Neural representation of envelope and temporal fine structure in listeners with normal hearing and sensorineural hearing loss. *Ear and Hearing, 37*(2), e91-e103. doi: 10.1097/AUD.0000000000000247.

Ballachanda, B.B., Rupert, A., & Moushegian, G. (1994). Asymmetric frequency-following responses. *Journal of the American Academy of Audiology, 5*(2), 133-137.

Bidelman, G. M. (2015). Multichannel recordings of the human brainstem frequency-following response: Scalp topography, source generators, and distinctions from the transient ABR. *Hearing Research, 323*, 68-80.

Bidelman, G. M., Gandour, J. T., & Krishnan, A. (2011). Cross-domain effects of music and language experience on the representation of pitch in the human auditory brainstem. *Journal of Cognitive Neuroscience, 23*(2), 425-434.

Bidelman, G.M., Weiss, M.W., Moreno, S., & Alain, C. (2014). Coordinated plasticity in brainstem and auditory cortex contributes to enhanced categorical speech perception in musicians. *European Journal of Neuroscience, 40*, 2662-2673.

Borden, G. J., Harris, K. S., & Raphael, L. J. (1994). *Speech Science Primer: Physiology, Acoustics, and Perception of Speech.* Baltimore, MD: Lippincott Williams & Wilkins.

Burkard, R. & McNerney, K. (2009). Introduction to Auditory Evoked Potentials. In J. Katz, L. Medwetsky, R. Burkard, & L. Hood (Eds.), *Handbook of Clinical Audiology* (6th ed., pp. 222-241). Baltimore, MD: Lippincott Williams & Wilkins.

Campbell, K.B. & Bartoli, E.A. (1986). Human auditory evoked potentials during natural sleep: The early components. *Electroencephalography and Clinical Neurophysiology, 65*(2), 142-149.

Clinard, C.G. & Cotter, C.M. (2015). Neural representation of dynamic frequency is degraded in older adults. *Hearing Research, 323,* 91-98. doi: 10.1016/j.heares.2015.02.002.

Davis, H. & Hirsh, S. K. (1976). The audiometric utility of brain stem responses to low-frequency sounds. *Audiology, 15*(3), 181-195.

Deacon-Elliott, D., Bell, I., & Campbell, K.B. (1987). Estimation of auditory threshold during sleep using brainstem auditory-evoked potentials. *Audiology, 26*(6), 363-368.

Don, M. & Kwong, B. (2009). Auditory Brainstem Response: Differential Diagnosis. In J. Katz, L. Medwetsky, R. Burkard, & L. Hood (Eds.), *Handbook of Clinical Audiology* (6th ed., pp. 222-241). Baltimore, MD: Lippincott Williams & Wilkins.

Emanuel, D. C. & Letowski, T. (2009). *Hearing Science*. Baltimore, MD: Lippincott Williams & Wilkins.

Galbraith, G. C. (1994). Two-channel brain-stem frequency-following responses to pure tone and missing fundamental stimuli. *Electroencephalography and Clinical Neurophysiology, 92,* 321-330.

Gardi, J., Merzenich, M., & McKean, C. Origins of the scalp-recorded frequency-following response in the cat. *International Journal of Audiology, 18* (5), 353-380.

Henry, K. S. & Heinz, M. G. (2013). Effects of sensorineural hearing loss on temporal coding of narrowband and broadband signals in the auditory periphery. *Hearing Research, 303*, 39-47. doi: 10.1016/j.heares.2013.01.014.

Hood, L. J. (1998). *Clinical Applications of the Auditory Brainstem Response*. San Diego, CA: Singular Publishing Group.

Hoormann, J., Falkenstein, M., & Hohnsbein, J. (2000). Early attention effects in human auditory-evoked potentials. *Psychophysiology, 37*, 29-42.

Hornickel, J., Skoe, E., & Kraus, N. (2008). Subcortical laterality of speech encoding. *Audiology & Neurotology, 14*, 198-207. doi: 10.1159/000188533

Jewett, D. L. & Williston, J. S. (1971). Auditory-evoked far fields averaged from the scalp of humans. *Brain, 94*, 681-696.

Johnson, K. (2011). *Acoustic and Auditory Phonetics.* Malden, MA: Wiley-Blackwell.

Krishnan, A. (2002). Human frequency-following responses: Representation of steady-state synthetic vowels. *Hearing Research, 166*(1-2), 192-201.

Krishnan, A. & Gandour, J. T. (2009). The role of the auditory brainstem in processing linguistically-relevant pitch patterns. *Brain and Language, 110*(3), 135-148. doi: 10.1016/j.bandl.2009.03.005.

Krishnan, A., Gandour, J. T., & Suresh, C. H. (2016). Language-experience plasticity in neural representation of changes in pitch salience. *Brain Research, 1637*, 102-117.

Krishnan, A., Xu, Y., Gandour, J., & Cariani, P. (2005). Encoding of pitch in the human brainstem is sensitive to language experience. *Cognitive Brain Research, 25*(1), 161-168. doi: 10.1016/j.cogbrainres.2005.05.004.

Krizman, J., Skoe, E., & Kraus, N. (2012). Sex differences in auditory subcortical function. *Clinical Neurophysiology, 123*(3), 590-597. doi: 10.1016/j.clinph.2011.07.037.

Kujawa, S. G., & Liberman, M. C. (2009). Adding insult to injury: Cochlear nerve degeneration after "temporary" noise-induced hearing loss. *Journal of Neuroscience, 29*(45), 14077-14085. doi: https://doi.org/10.1523/JNEUROSCI.2845-09.2009.

Landa, L., Krpoun, Z., Kolarova, M., & Kasparek, T. (2014). Event-related potentials and their applications. *Activitas Nervosa Superior, 56*(1-2), 17-23.

Lee, K. M., Skoe, E., Kraus, N., & Ashley, R. (2009). Selective subcortical enhancement of musical intervals in musicians. *The Journal of Neuroscience, 29*(18), 5832-5840. doi: 10.1523/JNEUROSCI.6133-08.2009.

Marsh, J. T., Brown, W. S., & Smith, J. C. (1974). Differential brainstem pathways for the conduction of auditory frequency-following responses. *Electroencephalography and Clinical Neurophysiology, 36*, 415-424.

Marsh, J.T. & Worden, F.G. (1968). Sound evoked frequency-following responses in the central auditory pathway. *The Laryngoscope, 78*(7), 1149-1163.

Moore, B. C. J. (2008). The role of temporal fine structure processing in pitch perception, masking, and speech perception for normal-hearing and hearing-impaired people. *Journal of the Association for Research in Otolaryngology, 9*, 399-406. doi: 10.1007/s10162-008-0143-x

Moore, B. C. J. & Glasberg, B. R. (1989). Mechanisms underlying the frequency discrimination of pulsed tones and the detection of frequency modulation. *Journal of the Acoustical Society of America, 86*(5), 1722-1732.

Moore, B. C. J. & Sęk, A. (2009). Sensitivity of the human auditory system to temporal fine structure at high frequencies. *Journal of the Acoustical Society of America, 125*(5), 3186– 3193.

Moushegian, G., Rupert, A.L., & Stillman, R. D. (1973). Scalp-recorded early responses in man to frequencies in the speech range. *Electroencephalography and Clinical Neurophysiology, 35,* 665-667.

Müllensiefen, D., Gingras, B., Musil, J., & Stewart L. (2014). The Musicality of Non-Musicians: An Index for Assessing Musical Sophistication in the General Population. *PLoS ONE, 9*(2): e89642. doi: 10.1371/journal.pone.0089642.

Nuwer, M. R. (1998). Fundamentals of evoked potentials and common clinical applications today. *Electroencephalography and Clinical Neurophysiology, 106*(2), 142-148. doi: 10.1016/S0013-4694(97)00117-X.

Parbery-Clark, A., Skoe, E., & Kraus, N. (2009). Musical experience limits the degradative effects of background noise on the neural processing of sound. *The Journal of Neuroscience, 29*(45), 14100-14107. doi: 10.1523/JNEUROSCI.3256-09.2009.

Parthasarathy, A., Lai, J., & Bartlett, E. L. (2016). Age-related changes in processing simultaneous amplitude modulated sounds assessed using envelope following responses. *Journal of the Association for Research in Otolaryngology, 17*, 119-132. doi: 10.1007/s10162-016-0554-z.

Paulraj, M. P., Subramaniam, K., Yaccob, S. B., Bin Adom, A. H., & Hema, C. R. (2015). Auditory evoked potential response and hearing loss: A review. *The Open Biomedical Engineering Journal, 9*, 17-24. doi: 10.2174/1874120701509010017.

Picton, T. (2010). *Human Auditory Evoked Potentials*. San Diego, CA: Plural Publishing.

Picton, T. W., Hillyard, S. A., Krausz, H. I., & Galambos, R. (1974). Human auditory evoked potentials I: Evolution of components. *Electroencephalography and Clinical Neurophysiology, 36*, 179-190.

Picton, T. W., Woods, D. L., Baribeau-Braun, J., & Healey, T. M. G. (1977). Evoked potential audiometry. *Journal of Otolaryngology, 6*(2), 90-119.

Plyler, P. N. & Ananthanarayan, A. K. (2001). Human frequency-following responses: Representation of second formant transitions in normal-hearing and hearing-impaired listeners. *Journal of the American Academy of Audiology, 12*, (523-533).

Reichenbach, T. & Hudspeth, A. J. (2012). Discrimination of low-frequency tones employs temporal fine structure. *PLoS ONE, 7*(9), 1-5. doi: 10.1371/journal.pone.0045579.

Robinson, R. (2015). Evoked Potential Studies. In J. L. Longe (Ed.), *The Gale Encyclopedia of Medicine* (5th ed., Vol. 3, pp. 1867-1869). Farmington Hills, MI: Gale.

Rossing, T. (2014). *Springer Handbook of Acoustics.* New York: Springer-Verlag. doi: 10.1007/978-1-4939-0755-7.

Ruggles, D., Bharadwaj, H., Shinn-Cunningham, B. G. (2012). Why middle-aged listeners have trouble hearing in everyday settings. *Current Biology, 22*, 19, 1417-1422.

Sanju, H. K. & Kumar, P. (2016). Enhanced auditory evoked potentials in musicians: A review of recent findings. *Journal of Otology, 11*, 63-72.

Sayles, M. & Winter, I. M. (2008). Reverberation challenges the temporal representation of the pitch of complex sounds. *Neuron, 58*, 789-801. doi: 10.1016/j.neuron.2008.03.029.

Skoe, E. & Kraus, N. (2010). Auditory brainstem response to complex sounds: A tutorial. *Ear & Hearing, 31*(3), 302-324. doi: 10.1097/AUD.0b013e3181cdb272.

Smith, J.C., Marsh, J. T., & Brown, W. S. (1975). Far-field recorded frequency following responses: Evidence for the locus of brainstem sources. *Electroencephalography and Clinical Neurophysiology, 39*, 465-472.

Snyder, R.L., & Schreiner, C.E. (1984). The auditory neurophonic: Basic properties. *Hearing Research, 15*, 261-280.

Varghese, L., Bharadwaj, H. M., & Shinn-Cunningham, B. G. (2015). Evidence against attentional state modulating scalp-recorded auditory brainstem steady-state responses. *Brain Research, 1626*, 146-154. doi: 10.1016/j.brainres.2015.06.038.

Walker, K. M. M., Bizley, J. K., King, A. J., & Schnupp, J. W. H. (2011). Cortical encoding of pitch: Recent results and open questions. *Hearing Research, 271* (1-2), 74-87. doi: 10.1016/j.heares.2010.04.015.

Wever, E.G. & Bray, C.W. (1930a). Action currents in the auditory nerve in response to acoustical stimulation. *Proceedings of the National Academy of Sciences of the United States of America, 16*(5), 344-350.

Wever, E.G. & Bray, C.W. (1930b). The nature of acoustic response: The relation between sound frequency and frequency of impulses in the auditory nerve. *Journal of Experimental Psychology, 13*(5), 373-387.

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NAME: Laura Grinstead

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DEGREE AND DATE TO BE CONFERRED: Doctor of Audiology; May 2018

Secondary education:
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Towson University	Fall 2010-May 2014	Bachelor of Science	May 2014
Towson University	Fall 2014-May 2018	Doctor of Audiology	May 2018

Major: Audiology

Professional publications:
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  York, PA 17404
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  Baltimore, MD 21287
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  104 Sleepy Hollow Dr. Suite 202
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- ENTAA Care: *Audiology Student Clinician*
  203 Hospital Dr.
  Glen Burnie, MD 21061
- Towson University Hearing and Balance Center: *Graduate Student Clinician*
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