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Waveform Characteristics of the Canine Click Evoked Brainstem Auditory Evoked Response Across Multiple Test Sessions

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UNIVERSITY OF NORTHERN COLORADO

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The Graduate School

WAVEFORM CHARACTERISTICS OF THE CANINE CLICK EVOKED BRAINSTEM AUDITORY EVOKED RESPONSE ACROSS MULTIPLE TEST SESSIONS

A Capstone Research Project Submitted in Partial Fulfillment of the Requirements for the Degree of Doctor of Audiology

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This Capstone Project by: Hayden H. Bruce

Entitled: Waveform Characteristics of the Canine Click Evoked Brainstem Auditory Evoked Response Across Multiple Test Sessions

has been approved as meeting the requirement for the Degree of Doctor of Audiology in the College of Natural and Health Sciences in the Department of Audiology and Speech-Language Sciences.

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ABSTRACT

Bruce, Hayden H. Waveform Characteristics of the Canine Click Evoked Brainstem Auditory Evoked Response Across Multiple Test Sessions. Unpublished Doctor of Audiology Capstone, University of Northern Colorado, 2019.

The purpose of this capstone research project was to investigate the consistency of waveform characteristics of the canine click evoked brainstem auditory evoked response (BAER) across multiple test sessions. Six canines were recruited to participate in the study. Brainstem auditory evoked response recordings were measured in each ear of the canines using a click stimulus at 102 dB peSPL, 82 dB peSPL and 62 dB peSPL. Canines included in the study were between the ages of one and seven to avoid both maturation and aging affects. Characteristics assessed throughout this study at each intensity level were absolute latencies of waves I, II, III and V, interpeak latencies of waves I-II, II-V, and I-V, amplitudes of waves I, II, III and V, wave V interaural differences at 102 dB peSPL, the lowest level wave V was observed, and the overall morphology of each BAER response. Results from this study indicated the BAER test was consistent across test sessions with respect to absolute and interpeak latencies, wave V interaural latency differences, and amplitudes. Overall waveform morphology was good for the highest stimulus intensity and varied from good to poor at lower intensity levels. The lowest level at which wave V was observed was consistent for the majority of canines in this study. Findings from this study suggested the canine click-evoked

BAER is a consistent assessment tool that both veterinarians and audiologists can be confident in when determining the integrity of the canine auditory system.

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CHAPTER I

STATEMENT OF THE PROBLEM

The auditory brainstem response is a waveform that represents responses of the auditory system through the level of the brainstem to acoustic stimulation (Jewett & Williston, 1971). Many synonymous terms have been used in place of the auditory brainstem response including brainstem auditory evoked response (BAER), which is often used in animal literature. Throughout this paper, BAER will be used when referring to testing in both humans and canines. The BAER is a popular diagnostic tool used by audiologists when estimating behavioral thresholds in infants and difficult-to-test patients. The BAER is also a commonly used neurodiagnostic tool.

In humans, the BAER waveform consists of several peaks represented by roman numerals with waves I, III, and V being the most robust peaks in normal hearing individuals (Arnold, 2007). Each of these peaks represents electrical activity originating from various structures within the auditory pathway.

Many stimulus and subject related parameters can affect the overall waveform morphology of the response. Stimulus-related parameters include intensity, presentation rate, polarity, and stimulus type. Subject-related factors include age, gender, and body temperature. However, when clinicians are able to appropriately set recording parameters and control for the factors related to the stimulus and subjects, the BAER has been proven to be a highly predictable response that is used clinically to assess the subcortical pathway (Jacobson, 1985).

In today's society, pets are considered part of the family and pet owners expect high quality health care when it comes to the care of their pets. This level of expectation from pet owners helps validate the need for diagnostic testing that can help prognosticate various disease processes (Albers, 2008; Coe, Adams, & Bonnett, 2007). The BAER is often utilized to verify that puppies do not have congenital hereditary deafness. It is also a useful tool for estimating auditory thresholds in canines and helps identify severity of hearing loss (Scheifele & Clark, 2012). The BAER might be utilized to assess hearing in working animals such as military or police canines to assure these canines do not have a hearing loss so they are able to complete their jobs to the best of their ability. Use of the BAER response is required by the Orthopedic Foundation for Animals (2018) database to assure that dogs who have hearing loss are not used for breeding.

Canine BAER testing might be done in a variety of settings but it is most often done in veterinary clinics. While there is a solid research base for the use of BAER in humans and its reliability, the body of research for the use of BAER in canines is much smaller. Normative data are available regarding BAER results in humans and what clinicians should expect but reports in the literature of canine BAER responses have been variable, making it difficult for veterinarians and audiologists to decipher what is considered "normal" and what is not.

Purpose of the Study

The purpose of this study was to examine canine BAER waveform characteristics including latency, amplitude, and morphology. Waveform characteristics were compared

for each canine across multiple test sessions to determine the consistency of evaluated waveform characteristics.

Research Question and Hypothesis

- Q1 Are waveform characteristics, using a click-evoked BAER consistent across multiple test sessions for three different presentation levels in canines?
- H1 Waveform characteristics will not vary significantly across sessions in canines absolute and interpeak latencies, interaural latency difference, amplitude, overall morphology, and lowest measured response when using a click-evoked BAER at three presentation levels.

CHAPTER II

REVIEW OF THE LITERATURE

Brainstem Auditory Evoked Response Testing in Humans

The Brainstem Auditory Evoked Response

The brainstem auditory evoked response (BAER) is a series of vertex-positive peaks that represent responses of the auditory nerve and the auditory brainstem structures to acoustic stimulation (Jewett & Williston, 1971). These peaks, or waves, occur within 10 ms after stimulation onset (Atcherson, 2012). The BAER is one of the most popular tools used clinically to estimate behavioral thresholds in infants and difficult-to-test patients. Each wave varies in latency and amplitude and is labeled by a roman numeral in order of latency. Waveform morphology is highly variable; often wave I is about half the size of wave V, wave II is relatively small, III is a larger and more distinguishable wave, while wave IV is usually a small component seen at the leading edge of wave V (Picton, Stapells, & Campbell, 1981). However, occasionally wave V will be a small component after wave IV but will still have a larger amplitude (peak to trough) than wave IV. Regardless of whether the peak of wave IV or V is larger, this is known as the IV/V complex. A recorded BAER of a normal hearing participant can be seen in Figure 1.

Each peak of the BAER represents electrical activity occurring along the auditory pathway, corresponding with areas from the vestibulocochlear cranial nerve (VIII) and

the brainstem (Knowles, Cash, & Blauch, 1988). Møller (2006) suggested that waves I and II originate from the distal and proximal portion of the auditory nerve, respectively, wave III from the cochlear nucleus, wave IV from brainstem structures including trapezoid bodies and the superior olivary complex, and wave V from the termination of the lateral lemniscus within the inferior colliculus on the contralateral side. However, it is important to note that this is an oversimplified model. Because the pathways within the central auditory nervous system are much more complex, there is no simple one-toone relationship between anatomical generators and peak components within the BAER waveform with the exception of wave I.



Figure 1. Brainstem auditory evoked response waveform of normal hearing human subject.

Waves I, III, and V are the most robust peaks in normal hearing individuals (Arnold, 2007). Waveform interpretation for neurodiagnostic purposes involves evaluation of waveform morphology and latencies of these peaks. Mean latencies are typically around 1.5, 3.5, and 5.5 ms for waves I, III, and V, while interpeak latencies are around 2 ms for I-III and III-V, and 4 ms for the latency difference between waves I and V (Atcherson, 2012). Amplitude information is considered to be the least useful during BAER interpretation because of how easily the amplitudes are affected by the signal-to-noise ratio (SNR) during testing (Atcherson, 2012). However, the wave V to wave I amplitude ratio is a diagnostically useful value. When comparing the wave V amplitude to the wave I amplitude, the amplitude of V is typically larger than that of wave I. Any wave V to wave I ratio below 0.75 is considered abnormal and could signify a retrocochlear pathology (Atcherson, 2012).

Several procedures and parameters related to testing would affect the measured BAER response: averaging, filtering, electrode placement, stimulus intensity, stimulus polarity, and stimulus type. Patient-related variables could also affect the test results such as age, gender, and body temperature (Atcherson & Stoody, 2012; Picton et al., 1981). These variables are discussed later in this chapter.

Maximizing Brainstem Auditory Evoked Response Visibility

The amplitude of the auditory brainstem response is not only very small but it is recorded in the midst of electrical and biological noise that would obscure the recording. The response is less than 50 microvolts and is recorded with other, much larger noise sources that include electroencephalogram and myogenic activity. Close proximity electrical devices are also a source of noise when recording the BAER. To accentuate the auditory brainstem response and minimize the amount of "noise" recorded, several procedures must be used to assure the SNR is improved enough so the BAER waveform is most visible (Oshrin & Terrio, 1990).

Differential amplifier. A differential amplifier serves to reduce the amount of background noise and then amplify the response so the response is maximally visible. The differential amplifier helps to sort out what is and is not pertinent to the recording. It is able to do this by using three electrodes (non-inverting, inverting, and ground). Before amplification takes place, any signal that is common to the inverting and non-inverting electrodes is eliminated while any signal that is different between these inputs is amplified. This is accomplished by the differential amplifier taking the response from the positive and negative electrodes and then inverting the response from the negative electrode. The responses are then summed and any similar response is eliminated from the response while differential amplifier helps to improve the SNR by only amplifying voltages that are part of the BAER instead of unwanted electrical noise from equipment or physiologic sources (Oshrin & Terrio, 1990), thus allowing for a more visible response.

Signal averaging. When recording the BAER, averaging must be used to improve SNR for visualization of the response. Signal averaging improves the visibility of the response by gradually canceling out noise when multiple samples of the response are collected. This is accomplished because the recorded response is time locked to the presented stimuli while noise is not; the time locked components will sum together while the random noise components will not (Atcherson & Stoody, 2012). Artifact rejection. Artifact rejection is another tool that is commonly used when recording the BAER to ensure elimination of as much artifact as possible. The artifact reject is a predetermined criterion and any activity above that criterion will be eliminated from the averaged response (Atcherson & Stoody, 2012). The typical background activity is around $\pm 5\mu V$ when a typical response is around $\pm 0.1\mu V$ (Picton et al., 1981). Larger background activity is generally caused by increased electrical activity and muscle activity including body movements such as head movement, swallowing, coughing, etc. If background activity steadily exceeds $\pm 5\mu V$, the number of samples to be averaged should be increased.

Filtering. Filtering is another strategy to decrease the contribution of unwanted background activity and noise because it allows for some frequencies to be passed and recorded while others are eliminated depending on the type of filter used. Filtering out specific frequencies allows for researchers to analyze only information pertinent to the spectral make-up of the BAER (Atcherson & Stoody, 2012; Picton et al., 1981). Types of filters include high-pass, low-pass, and band-pass filters. High- and low-pass filters pass and cut frequencies based on cutoff frequencies. For example, if the high-pass filter is set at 100 Hz, any frequencies above 100 Hz will be passed and frequencies below 100 Hz will be filtered out. On the other hand, if the low-pass filter is set at 1500 Hz, frequencies below 1500 Hz will be passed and frequencies above will be filtered out. Band-pass filters pass a selected range of frequencies and eliminate those outside of the specified range. When recording the BAER, it is up to the tester to determine the filter settings. An example of appropriate filter settings when recording the BAER in human

adults is 100 to 300 Hz for the high-pass filter and 1500 to 3000 Hz for the low-pass filter. A typical band-pass filter setting is 100 to 3000 Hz (Atcherson & Stoody, 2012).

Electrode placement. A minimum of three electrodes are needed to record the BAER. Electrode placement is driven by the desired response; which waves a tester wants to be the most evident will determine where electrodes are placed. Two typical electrode placements were discussed by Picton et al. (1981). The first configuration consisted of one electrode (non-inverting) on the vertex with the other electrode (inverting) on the earlobe or mastoid ipsilateral to the ear being stimulated. The other placement option was one electrode (non-inverting) on the vertex with the other electrode (inverting) on the earlobe or mastoid contralateral to the ear being stimulated. When ipsilateral and contralateral placements were compared, larger amplitudes and shorter latencies were reported for ipsilateral measurements. When comparing earlobe to mastoid electrode placement, Picton et al. (1981) reported no difference, leaving the decision to the preference of the tester.

An additional electrode placement includes the use of the nape of the neck. The non-inverting electrode is placed on the vertex and the inverting electrode is placed on the nape. While earlobe versus mastoid placement showed no significant difference in a study by Herrmann, Thornton, and Joseph (1995), a slight difference between BAER responses was seen when using mastoid versus nape electrode placement. The authors screened 451 neonates between the ages of 30 and 58 weeks using BAER over a 46-month period. Instead of using the vertex to mastoid electrode montage, the researchers chose to use a vertex to nape montage. This was chosen because they had observed wave V to be significantly larger when using the nape compared to the mastoid. This increased

amplitude of wave V with nape electrode placement was seen because of the electrode montage matching the direction of the dipole. This was created during depolarization when positive ions rushed into one region of a neuron and caused the extracellular space in that region to become negative. This initial inward flow of positive ions must eventually exit in another region of the neuron, causing the extracellular space in the adjacent region to become positive. As this current flow was generated, a separation of charges at different regions of the neuron was set up and produced the dipole. If a strong wave V was desired, the nape was used to match the vertical dipole. However, if a strong wave I was desired, then mastoid or earlobe placement should be used to match the horizontal direction of the dipole.

In a study by Atcherson, Lim, Moore, and Minaya (2012), the traditional electrode placement of earlobe or mastoid was compared to ear canal electrode placement. The study consisted of 20 adults between 18 and 50 years of age. When recording the BAER, the researchers hoped to see a larger wave I with the ear canal electrode placement compared to the earlobe or mastoid. Recordings yielded no statistical difference for peak latency or peak-to-trough measurements for waves I, III, or V between the various electrode placements.

Transducer. The BAER can be obtained with stimuli presented to the ear with either supra-aural or insert earphones. For better BAER results, it is best to use insert earphones because of increased interaural attenuation and the separation of the transducer box from the insert phone that is coupled to the patient's ear via a tube, which helps to reduce stimulus-related artifact (Atcherson, 2012).

Stimulus. Brainstem auditory evoked response waveforms are best recorded using transient (quick onset and offset times) stimuli such as clicks or tone bursts. The click stimulus is more commonly used to diagnose a lesion or screen hearing sensitivity and it is chosen because of its ability to generate synchronous firing of the auditory nerve fibers. Although a click stimulus contains spectral energy across a range of frequencies, the BAER represents activity primarily in the 1000-4000 Hz region of the cochlea because frequencies below 1000 Hz take longer to travel along the basilar membrane than higher frequencies and are therefore not included in the response (Atcherson, 2012).

While a click stimulus is more commonly used diagnostically, a tone burst stimulus might be used to obtain more frequency-specific estimates of hearing sensitivity because of its activation of a more specific section of the basilar membrane compared to the click stimulus (Arnold, 2007; Dagna, Canale, Lacilla, & Albera, 2014).

Stimulus-Related Parameters in Humans

When presenting the acoustic stimulus, several parameters affect the overall waveform morphology of the BAER response: stimulus intensity, rate, polarity, and type.

Stimulus intensity. The brainstem response can be obtained for a range of intensity levels but if the purpose is to assess neural integrity (neurodiagnosis), all components of the waveform are best displayed for a stimulus level at a moderately high intensity around 70 to 90 dB nHL. It is best to use these moderately high intensities because they provide BAER responses with the largest amplitudes (best morphology) and shortest latency responses and because normative databases reflect information collected with moderately high intensities. Picton et al. (1981) found changing the intensity of the click stimulus altered the latency, amplitude, and morphology of the BAER. They

reported that decreased intensity resulted in an increased peak latency for all components. Picton, Woods, Baribeau-Braun, and Healey (1976) found the peak latency for wave V changed from 5.6 ms at 80 dB nHL to 8.2 ms at 10 dB nHL. When comparing amplitude to intensity, "the amplitude decreased much more rapidly below 20 dB nHL and increased somewhat more slowly above 70 dB nHL" (Picton et al., 1981, p. 15).

Stimulus rate. Choice of appropriate stimulation rate for a given auditory evoked potential test depends upon the duration of the stimulus as well as the length of the recording time window. An example of this would be if a time window of 10 ms is being used, then a click stimulus could be presented 1 every 10 ms or 100 per second. The most commonly used stimulation rate for BAER testing varies between 10 and 40 stimuli per second (Atcherson, 2012). Stimulation rate can also affect the morphology of the response, specifically the latencies and amplitudes. Chiappa, Gladstone, and Young (1979) found that increasing the rate of the stimulus presentation increased the latencies and decreased the amplitudes. Picton et al. (1981) found that increasing the presentation rate from 10 to 80 clicks per second decreased wave V amplitude to only 90% from what it was at 10 per second but decreased waves I and III to 50% of their values at 10 per second. When comparing an increase in presentation rate and latencies, it was observed that from 10 to 80 clicks per second, latencies for waves I, III, and V increased by 0.14, 0.23, and 0.39 ms, respectively. Until the presentation rate of the stimulus was greater than 10 clicks per second, there were no significant changes in the latencies (Picton et al., 1981). It could also be said that decreasing the rate would decrease the latencies and increase the amplitudes. However, faster rates are often used when completing a threshold estimation because interpretation is more reliant on the presence of wave V

rather than morphology and latency values. A faster presentation rate would also allow for shorter test time, which could be very important with infants and difficult-to-test patients.

Stimulus polarity. When choosing a stimulus polarity, which refers to the starting phase of the acoustic stimulus waveform that affects the direction of the output waveform, there are three options: rarefaction, condensation, or alternating between rarefaction and condensation (Hall, 2007). Polarity does not significantly affect waveform results but a slight difference has inconsistently been reported in the literature. The biggest difference noted was wave I had a shorter latency and larger amplitude with a rarefaction click (Stockard, Stockard, Westmoreland, & Corfits, 1979). For wave IV, Stockard et al. (1979) found shorter latencies with a rarefaction click compared to a condensation click, which was thought to be due to rarefaction polarity causing depolarization first rather than hyperpolarization as was seen with condensation polarity stimulation. No apparent differences were noted related to polarity in waves III and V.

Stimulus type. As previously stated, BAER waveforms are best recorded using transient stimuli such as clicks or tone bursts. Clicks are generally used for neurodiagnostic testing and newborn hearing screening while tone bursts are used to obtain more frequency-specific information if a hearing loss is suspected following a screening test. However, Campbell and Brady (1995) found that a click stimulus and 1,000 Hz tone burst were similar in sensitivity and specificity when recording a neurodiagnostic BAER. The study consisted of 45 patients with sensorineural hearing loss and 13 patients with tumors affecting the eighth nerve. In the patients with tumors, waves I and III were generally absent for both the click stimulus and 1000 Hz tone and

wave V was present in response to both stimuli. One difference noted was when evaluating patients with sensorineural hearing loss, waves I and III appeared less frequently in response to the tone burst than they did with the click stimulus. The researchers concluded the click stimulus should be the primary stimulus in otoneurologic BAER evaluation due to the interaural latency difference of wave V with a click stimulus, yielding high sensitivity and specificity and more often allowing analysis of interpeak latencies of waves I through V than the 1000 Hz stimulus.

Subject-Related Parameters in Humans

Several subject factors need to be considered when performing the BAER. Researchers have studied these subject-related parameters and how they directly influenced the overall waveform of the BAER. Some of these parameters related to the participant included age, gender, and body temperature.

Age. The auditory system changes with age and these changes could influence the BAER (Boettcher, 2002). In a study by Hecox and Galambos (1974), BAER responses were compared for 35 infants ranging in age between three weeks and three years. Waveforms were also recorded from three adults between the ages of 24 and 25 to determine the reliability of the measures. The authors reported adult-like responses by the age of 12 to 18 months. Waveforms recorded from infants exhibited latency decreases with increasing age. Another important finding from Hecox and Galambos was age could be a predictor of latency for wave V. Rowe (1978) reported an increase in latency as age increased when comparing the waveform responses for 25 young (17- to 33-years-old) and 25 old (51- to 74-years-old) subjects. Konrad-Martin et al. (2012) reported age-related effects on the BAER. Participants in this study included 131 predominantly male veterans whose ages ranged from 26 to 71 years. The results showed that aging greatly reduced amplitudes of all peaks in the BAER response but the greatest decrease in amplitudes occurred for waves I and III. There was about a 1μ V difference for amplitudes of waves I and III when comparing the youngest age to the oldest age. Konrad-Martin et al. also found that with older age, peak latencies increased by about 0.5 ms when interpreting the recorded BAER response.

Gender. Sex of the participant was reported in the literature to influence the amplitude and peak latencies of the BAER response, resulting in some clinics developing gender-specific norms for their equipment. Adult females had significantly shorter latencies for waves III and V when compared to adult males (Picton et al., 1981). Females also had larger amplitudes of all components compared to males (Kjaer, 1979; Michalewski, Thompson, Patterson, Bowman, & Litzelman, 1980). Picton et al. (1981) also found the amplitudes of waves I, III, and V to be larger in females than in males. Waves I and V were measured to be about 30% larger while wave III was 23% larger in females than in males.

There are several theories for the differences between male and female BAER responses. McClelland and McCrea (1979) observed no differences in responses in participants between the ages of 9 and 13 but they noted differences for participants who were 14 or older. Hormonal changes were cited as a possible reason for these changes. O'Donovan, Beagley, and Sen (2009) found significant differences in the responses as early as eight-years-old and onwards that were credited to anatomical differences

between males and females. These anatomical differences included males having larger heads and thicker scalps than females, thereby creating increased latencies. Today, though, we know these anatomical differences are more related to basilar membrane length differences between males and females (McFadden, 1998). Since females tend to have shorter basilar membranes than males, the signal has a shorter pathway to travel, creating a shorter latency than typically seen in males.

Body temperature. Body temperature has also been found to affect the overall waveform morphology of the BAER response. A lower body temperature could increase the latencies of the components of the BAER (Kaga, Takiguchi, Myokai, & Shiode, 1979; Stockard, Sharbrough, & Tinker, 1978). Picton et al. (1981) suggested one reason for the effect temperature had on the BAER was that a higher temperature caused both action potentials and postsynaptic potentials to have quicker onsets and shorter durations. These changes in action and postsynaptic potentials caused an increase in both the neuronal conduction velocity and the speed of synaptic transmission. Wada (1981) studied the BAER response during sleep, which changed the body temperature. It was found that wave V showed on average a 0.2 ms increase in latency with a decrease in temperature of 1 degree Celsius.

Reliability of Brainstem Auditory Evoked Response Testing in Humans

The BAER has been shown to be a highly predictable response that is used clinically to assess the subcortical pathway (Jacobson, 1985). Munjal, Panda, and Pathak (2016) examined the long-term test-retest reliability of the BAER. The study consisted of 50 normal hearing participants who were retested at 3 months, 6 months, and 12 months beyond the initial test. Each participant was tested using a click stimulus at 70 dB nHL and 90 dB nHL with a bandpass filter of 100 Hz to 3000 Hz and a repetition rate of 19.3 clicks per second with a minimum of 1,024 clicks at each recording. Absolute latencies of waves I, III and V, and interpeak latencies of I-III, III-V and I-V were measured. Results showed no significant differences (p > 0.05) between test sessions for absolute latencies of waves III and V or interpeak latencies of waves I-III, III-V, and I-V, indicating good test-retest reliability for these measures. However, a statistical difference (p < 0.05) was reported for the absolute latency of wave I in the right ear, thereby not establishing reliability of wave I. Overall, the results indicated the BAER was an appropriate tool to monitor neurodiagnostic disorders as well as progressive disorders of the central nervous system.

Brainstem Auditory Evoked Response Testing in Animals

The BAER is a tool used by veterinarians and audiologists to aid in the detection of hereditary deafness in puppies and detection of hearing loss in canine pets, breeding stock, and working animals such as military or police dogs. Specifically, for canines, the BAER is used to certify that puppies do not suffer from congenital hereditary deafness (Orthopedic Foundation for Animals, 2018) but it is also a useful tool to estimate auditory thresholds in canines (Scheifele & Clark, 2012) or to assist medical professionals in locating brainstem lesions (Steiss, Cox, & Hathcock, 1994; Strain, 1996).

Scheifele and Clark (2012) described the BAER as an evoked response that could be measured through electrodes placed in the scalp and measured within the first 10 ms following presentation of an acoustic stimulus. This test measured neural responses from the vestibulocochlear nerve (CN VIII) and lower brainstem that consisted of up to seven waves represented by roman numerals with only five of them being clinically significant. The exact location of origin of each wave is largely unknown; however, Scheifele and Clark suggested wave I derived from the distal portion of CN VIII, wave II from the proximal portion of CN VIII, wave III from the cochlear nuclei, and waves IV and V from multiple areas including the inferior colliculus and medial geniculate body.

All of the same protocol parameters and variables involved in obtaining the human BAER must also be considered when recording the BAER in canines including differential amplifier, signal averaging, artifact rejection, filtering, electrode placement, transducers, and type of stimulus. The only differences in protocol for humans compared to canines is the electrode type and the electrode placement sites. Surface electrodes are used for humans while subdermal needle electrodes are used for testing canines. This difference is discussed later in this section. Table 1 shows typical protocol parameters used for the BAER in canines.

Table 1

Parameter	Typical Setting
Gain	100,000-150,000 μV
Signal Averaging	1000-2000 samples
Filter	High pass: 300 Hz
	Low pass: 1500 Hz
Electrode Placement	Subdermal (Cz, A ₁ , A ₂)
Transducer	ER-2 insert earphones
Stimulus	Click or tone burst
Stimulation Rate	33.3 clicks per second
Q_{1} Q_{2} Q_{2	

Recording Settings and Parameters Used When Recording the Brainstem Auditory Evoked Response in Canines

Source. Scheifele and Clark (2012).

Stimulus Intensity in Animals

Intensity is measured in dB nHL (normal hearing level) in humans but this is not the case for canines. When performing the BAER on canines, intensity is measured in dB peak equivalent sound pressure level (peSPL). Intensity must be measured in dB peSPL in canines rather than dB nHL because dB nHL are levels based on human behavioral responses and behavioral responses in canines are different from those of humans. Intensity levels that have been used diagnostically when using an air-conducted click stimulus are 70 dB peSPL, 80 dB peSPL, 90 dB peSPL, 102 dB peSPL, and 116 dB peSPL (Scheifele & Clark, 2012). Scheifele and Clark (2012) also recommended that at least two responses be collected from each ear at each intensity level to demonstrate reproducibility of the waveform. When presenting the stimulus, the transducer types used for the canine are similar to those used for humans. Transducers include insert earphones, supra-aural headphones, or a bone oscillator. Scheifele and Clark reported reduced stimulus artifact when insert earphones were used rather than the supra-aural headset.

Animal Preparation

The BAER can be administered with the animal either under sedation or awake with numbing topical cream (2.5% lidocaine/2.5% prilocaine cream) applied where the electrodes are placed (Scheifele & Clark, 2012). During testing, 13 mm subdermal bent needle electrodes are used and placed at three locations on the head. Scheifele and Clark (2012) discussed placement of electrodes during testing. The positive non-inverting electrode was placed on the vertex or midline of the subject's head (Cz). The negative electrode was placed in front of the tragus of the test ear (A_i) and the ground electrode was placed in front of the tragus of the non-test ear (A_c). Once the electrodes were placed on the canine, the impedance of each electrode was checked. Impedance refers to the amount of resistance present in an electrical circuit when voltage is applied (Scheifele & Clark, 2012). Scheifele and Clark recommended the impedance be equal between electrodes and no more than 5000 ohms to minimize electrical noise that could potentially affect the BAER response.

Factors Affecting the Brainstem Auditory Evoked Response in Animals

Similar to the BAER in humans, various stimulus- and subject-related factors can affect the BAER response in canines. Stimulus factors that might affect the BAER in canines are stimulation rate and intensity. Other factors specific to canines include age, breed, and head size.

Age. In a study by Shimada, Ebisu, Morita, Takeuchi, and Umemura (1997), 23 dogs ranging in age from 3 to 17 years were included to study the effects of age on the auditory structures in canines. What was found was histological and physiological changes in structures such as the organ of Corti, spiral ganglion, stria vascularis, and basilar membrane present with presbycusis in humans were very similar to those found in canines. The similarities in presbycusis for humans and canines suggested the BAER was affected by age for both groups. Ter Haar, Venker-van Haagen, van den Brom, van Sluijs, and Smoorenburg (2008) conducted a study to show the affects age had on the canine BAER. The study consisted of three groups of 10 dogs who were of similar weight but different ages. Group I consisted of dogs between the ages of 0.9- and 3.4-years-old, group II ranged from 3.5- to 7-years-old, and group III ranged from 11- to 14-

years-old. Brain auditory evoked response thresholds were estimated for each group at frequencies 1, 2, 4, 8, 12, 16, 24, and 32 kHz with tone burst stimuli delivered to the ear via a high frequency bandpass speaker connected to the ear via a transmission tube inserted deep in the ear canal starting at 80 dB peSPL and decreasing in 10 dB steps until threshold was reached. Researchers found thresholds were significantly higher in canines from group III compared to canines in groups I and II at all frequencies. They also found canines in group II showed higher thresholds than group I at 4000 Hz. In all dogs, as intensity decreased, latency increased until threshold was found. When looking at the BAER results from group II, the researchers were able to conclude that increasing thresholds associated with aging were most common around 8- to 10-years-old and affected the middle to high frequency range (8000-32,000 Hz) the most.

Breed and head size. Kemper, Scheifele, and Clark (2013) conducted a study to assess the effects of breed and head size on the BAER in canines. The study consisted of 43 dogs of 14 different breeds between the ages of 13 and 120 months. All canines included in the study were assumed to have normal hearing and underwent both a physical and otoscopic examination before participating in the study. Two measurements were included when recording head size for each canine. Measurements were taken with calipers from the temporal bone to the tragus and from the top of the head to the occipital bone. While preforming the BAER, researchers used a 90 dB peSPL click stimulus. Results of the study indicated neither head size nor breed had a significant impact on latencies, interpeak latencies, or waveform morphology of the BAER response.

Gaps in Brainstem Auditory Evoked Response Research

The BAER is a commonly used tool to estimate hearing thresholds in humans as well as help in locating site of lesion. As discussed previously, several procedural decisions are made when performing the BAER including averaging, filtering, and electrode placement. Stimulus- and participant-related factors could also affect the overall waveform morphology of the BAER response including stimulus intensity and presentation rate, stimulus polarity, age of participant, gender, and body temperature. However, even with all of these factors, the BAER has been shown to be a highly consistent tool in humans. Jacobson (1985) found the BAER to be a highly predictable response to assess the subcortical pathway. Munjal et al. (2016) found the long-term testretest reliability of the click-evoked BAER to be good.

The BAER waveform morphology in canines is affected by the same procedures and parameters as the BAER in humans with the addition of participant parameters including breed of the canine as well as head size. Since the BAER in humans has been shown to be consistent, one would assume the BAER in canines would also be a highly consistent tool. However, very little research existed on BAER testing in canines compared to the research base of BAER testing in humans. While there was some research pertaining to BAER testing in canines, a lack of normative data pertaining to latency, amplitude, and overall morphology made it difficult to decipher what was "normal." Scheifele and Clark (2012) discussed the importance of a universal protocol for canine BAER testing in order to obtain normative data and make animal audiology as effective as human audiology. Using a universal protocol and obtaining normative data would assist researchers in examining canine BAER waveform characteristics and their consistency across test sessions.

CHAPTER III

METHODOLOGY

Subjects

Following Institutional Animal Care and Use Committee approval (see Appendix A), six canines were included in this study who were volunteered by their owners. Canines of any breed were included between the ages of one and seven years. Dogs younger than one year were not included to avoid issues related to maturation and dogs eight years or older were not included to avoid canines with hearing loss related to aging. Subjects were available for testing three times during a three-month time period. Canines completed a wellness check before testing that included specific questions related to the hearing of the canine. Any canine with history of neurological or otic disease, previous use of ototoxic drugs, any known hearing loss, or exposure to hazardous sound levels within the testing time frame was excluded from the study.

Owners of canines were at least 18 years of age. Participants were recruited via word of mouth. At the time of testing, a written consent was obtained from the owner of each canine subject (see Appendix B).

Recording Equipment

All BAER recordings were done using the Intelligent Hearing Systems USB Box with SmartEP software, Version 5.10, installed on a Windows laptop computer.
Calibration of this system was current within one year. The stimulus was presented to the canine via standard ER-2 insert earphones.

Brainstem Auditory Evoked Response Recording Procedure

Data collection took place at Full Circle Veterinary Care, located in Johnstown, Colorado where the attending veterinarian completed overall health inspections of the canines included in the study.

At the beginning of each test session, a checklist was filled out to assure complete preparation for testing as well as consistency across test sessions (see Appendix C). Prior to any testing, each canine was placed in a ThundershirtTM to help calm the dog. Once the Thundershirt was placed on the canine, the researcher applied a small application of topical anesthetic (2.5% lidocaine, 2.5% prilocaine) directly to the skin of the canine at the site of sub-dermal electrode placement. The anesthetic soaked into the skin of the canine for 15-30 minutes to numb areas of the skin where sub-dermal electrodes were placed. Rhythmlink disposable subdermal needles with a 13 mm length and 0.4 mm diameter were placed at Cz (top of the head), A_1 (just below the tragus of test ear), and A_2 (just below tragus of non-test ear). The impedance of each electrode was checked before BAER testing began and was not more than 3,000 ohms.

Restraint of canines was accomplished without the use of sedation. Instead, a member of the research team gently restrained the canines depending on their size and activity level.

Testing was done on both ears of the canine separately. The BAER recordings were obtained using a high intensity broadband click stimulus of 102 dB peSPL with a duration of 0.1 milliseconds in order to analyze the overall morphology of the BAER

response. Testing also included recordings at 82 dB peSPL and 62 dB peSPL using a 0.1 millisecond broadband click stimulus. At least two runs were obtained at each stimulus intensity to ensure reproducibility. Polarity of the stimulus was alternating. The stimulus was presented at a rate of 33.3 clicks per second with 1,000 sweeps. The high-pass filter was set at 100 Hz while the low-pass filter was set at 1500 Hz. An absolute gain of 100,000 was utilized with an artifact rejection rate of 35.1%.

Descriptive Analysis

Overall morphology of the BAER responses was compared across test sessions. When interpreting the BAER waveform response, several measurements were reported for each stimulus intensity: absolute latency of waves I, II, III, and V (assuming the canine waveform had clear definitive waveforms); amplitude of waves I, II, III, and V; wave V interaural difference; and overall morphology of each BAER response. Other measurements included interpeak latencies between waves I-II, II-V, and I-V at the intensity level of 102 dB peSPL and the lowest measured response of each session. The researcher and at least one experienced member of the research team discussed the absolute latency of waves I, II, III, and V at the time of testing and the waveforms were re-analyzed after the testing and latencies were agreed upon by the researcher and two experienced members of the research team. Absolute latency information was reported for each intensity level and averaged between runs. When looking at overall waveform morphology, descriptive ratings of good, fair, and absent were used. It was determined by the researcher and two experienced members of the research team that a good rating consisted of identifiable and repeatable waves I, III, and V, or I, II, and V. Fair consisted of some identifiable wave components, reduced amplitude, and poor repeatability. An absent rating for overall morphology consisted of no identifiable waveform response.

CHAPTER IV

RESULTS

Participants

Six healthy canines were recruited for participation in the study and were between the ages of two- and six-years-old to avoid any maturational or aging effects on the response. All six canines completed three sessions of testing scheduled once a month for three months. Table 2 provides subject demographic information.

Table 2

SUA
М
М
М
М
F
F
M M M F F

Age, Breed, and Sex of Each Test Subject

Descriptive Analysis

Absolute Latency

Absolute latency measurements were obtained based upon marked components for waves I, II, III, and V at each intensity level for each of the three sessions.

102 dB peSPL. Wave I mean absolute latencies across all three test sessions were calculated for each ear for all six subjects and ranged from 1.29 ms to 1.71 ms with a grand average of 1.56 ms. Wave II mean absolute latencies at 102 dB peSPL ranged from 2.24 ms to 2.48 ms with a grand average of 2.3 ms. Mean absolute latencies of wave III at 102 dB peSPL ranged from 2.45 ms to 3.07 ms with a grand average of 2.93 ms. Wave V mean absolute latency at 102 dB peSPL ranged from 3.55 ms to 4.16 ms with a grand average of 3.87 ms. Table 3 reports the means and standard deviations for waves I, II, III, and V of each ear at 102 dB peSPL.

Table 3

Subject/Ear	Wave I		W	ave II	Wa	ave III	Wave V		
	М	SD	М	SD	М	SD	М	SD	
1/Left	1.59	0.13	2.28	0.11	2.93	0.11	3.95	0.13	
1/Right	1.58	0.06	2.24	0.04	2.90	0.03	3.93	0.08	
2/Left	1.71	0.05	2.48	0.09	3.07	0.08	4.16	0.10	
2/Right	1.64	0.04	2.40	0.02	2.97	0.04	3.68	0.17	
3/Left	1.67	0.08	2.39	0.05	3.03	0.09	3.97	0.14	
3/Right	1.49	0.04	2.32	0.03	2.99	0.04	3.88	0.05	
4/Left	1.29	0.02	2.03	0.00	2.45		3.58	0.09	
4/Right	1.34	0.06	2.05	0.03			3.55	0.09	
5/Left	1.63	0.14	2.43	0.11	2.92	0.05	3.96	0.18	
5/Right	1.66	0.02	2.46	0.05	3.04	0.05	3.86	0.12	
6/Left	1.49	0.09	2.25	0.05	2.79	0.06	3.90	0.03	
6/Right	1.62	0.12	2.29	0.10	2.81	0.04	3 99	0.16	

Absolute Latencies (in Milliseconds) at 102 dB peSPL Across All Ears and Test Sessions

Note. "--" represents a mean or standard deviation unable to be calculated due to insufficient data points across the three sessions.

To better understand the variability of absolute latency values for the individual canines, the range of absolute latencies across ears and test sessions of each canine at 102 dB peSPL is summarized in Table 4.

Table 4

Subject/Ear	Wave	Ι	Wave	II	Wave	III	Wav	e V
	Range	SD	Range	SD	Range	SD	Range	SD
1/Left	1.45-1.70	0.13	2.15-2.35	0.11	2.80-3.00	0.11	3.83-4.08	0.13
1/Right	1.53-1.65	0.06	2.20-2.28	0.04	2.88-2.93	0.03	3.85-4.00	0.08
2/Left	1.65-1.75	0.05	2.38-2.53	0.09	3.00-3.15	0.08	4.10-4.28	0.10
2/Right	1.60-1.68	0.04	2.38-2.42	0.02	2.93-3.00	0.04	3.50-3.83	0.17
3/Left	1.60-1.75	0.08	2.33-2.42	0.05	2.93-3.08	0.09	3.83-4.10	0.14
3/Right	1.45-1.53	0.04	2.30-2.35	0.03	2.95-3.03	0.04	3.83-3.93	0.05
4/Left	1.27-1.30	0.02	2.03-2.03	0	2.45	—	3.53-3.68	0.09
4/Right	1.27-1.38	0.06	2.03-2.08	0.03		—	3.48-3.65	0.09
5/Left	1.50-1.78	0.14	2.30-2.50	0.11	2.88-2.95	0.05	3.78-4.13	0.18
5/Right	1.65-1.68	0.02	2.40-2.50	0.05	3.00-3.10	0.05	3.78-4.00	0.12
6/Left	1.38-1.53	0.09	2.20-2.30	0.05	2.73-2.85	0.06	3.88-3.93	0.03
6/Right	1.50-1.73	0.13	2.20-2.40	0.10	2.78-2.83	0.04	3.83-4.15	0.16

Range and Standard Deviation of Absolute Latencies (in Milliseconds) at 102 dB peSPL for Each Canine Across All Ears and Test Sessions

Note. "--" represents a mean or standard deviation unable to be calculated due to insufficient data points across the three sessions.

82 dB peSPL. As expected, mean absolute latencies of wave I for each canine occurred slightly later at 82 dB peSPL compared to 102 dB peSPL. Mean absolute

latencies for wave I at 82 dB peSPL ranged from 1.48 ms to 2.04 ms with a grand average of 1.79 ms. Wave II mean absolute latencies at 82 dB peSPL ranged from 2.41 ms to 2.80 ms with a grand average of 2.58 ms. At 82 dB peSPL, one canine was reported to have no wave III response in both ears in at least two of the three sessions. Wave III mean absolute latencies at 82 dB peSPL ranged from 3.16 ms to 3.47 ms with a grand average of 3.29 ms. Wave V mean absolute latency ranged from 3.79 ms to 4.46 ms at 82 dB peSPL with a grand average of 4.19 ms. Table 5 reports the means and standard deviations for waves I, II, III, and V of each ear at 82 dB peSPL.

Table 5

Absolute Latencies (in Milliseconds) at 82 dB peSPL Across All Ears and Test Sessions

Subject/Ear	Way	ve I	Wav	re II	Wav	e III	Wav	e V
·	M	SD	M	SD	M	SD	M	SD
1/Left	1.74	0.08	2.53	0.13	3.17	0.10	4.23	0.24
1/Right	1.76	0.03	2.46	0.08	3.35	0.40	4.20	0.13
2/Left	2.04	0.19	2.80	0.21	3.47	0.25	4.46	0.18
2/Right	1.93	0.08	2.61	0.06	3.28	0.07	4.24	0.13
3/Left	1.83	0.07	2.63	0.07	3.36	0.03	4.33	0.06
3/Right	1.82	0.09	2.68	0.07	3.32	0.10	4.20	0.26
4/Left	1.48	0.04	2.41	0.11			3.79	0.01
4/Right	1.48		2.59	0.30		—	3.95	0.11
5/Left	1.79	0.09	2.59	0.12	3.24	0.18	4.26	0.14
5/Right	1.82	0.12	2.68	0.09	3.36	0.10	4.20	0.12
6/Left	1.73	0.03	2.49	0.04	3.20	0.08	4.24	0.10
6/Right	1.76	0.09	2.50	0.09	3.16	0.14	4.22	0.19

Note. "--" represents a mean or standard deviation unable to be calculated due to insufficient data points across the three sessions.

62 dB peSPL. All canines had an identifiable wave I in each ear at both 102 and 82 dB peSPL; however, when testing at 62 dB peSPL, one canine was reported to have no wave I response in at least two of three sessions. Wave I mean absolute latencies at 62 dB peSPL ranged from 1.93 ms to 2.34 ms with a grand average of 2.09 ms. Similar to wave I responses at 62 dB peSPL, three canines had no response in at least one ear for at least two of the three sessions at 62 dB peSPL. Wave II mean absolute latency differences at 62 dB peSPL ranged from 3.69 ms to 3.01 ms with a grand average of 2.80 ms. Two canines had no wave III response in at least one ear for at least two of the three sessions at 62 dB peSPL. Mean absolute latencies of wave III ranged from 3.33 ms to 3.65 ms at 62 dB peSPL. Wave V mean absolute latencies ranged from 4.42 ms to 5.05 ms at 62 dB peSPL with a grand average of 4.64 ms. Table 6 includes the means and standard deviations of waves I, II, III, and V of each ear at 62 dB peSPL. Grand averaged absolute latency values across all canines and sessions for the three intensity levels are summarized in Table 7. The latency-intensity function showed linear growth in absolute latency for all waveform components as the intensity was decreased (see Figure 2).

Table 6

Subject/Ear	Wa	ive I	Wa	ive II	Wa	ve III	Wa	ve V
, c	M	SD	M	SD	M	SD	M	SD
1/Left	2.09	0.06	2.74	0.09	3.33	0.18	4.60	0.18
1/Right	2.06	0.12	2.75	0.07	3.33		4.48	0.17
2/Left	2.34	0.01			3.65		4.84	0.21
2/Right	2.10	0.10	2.85		3.55		4.66	0.10
3/Left	2.07	0.05	2.92	0.05	3.62	0.12	4.71	0.30
3/Right	2.16	0.18	3.01	0.11	3.58		4.71	0.35
4/Left							4.63	
4/Right							5.05	
5/Left	1.93		2.80		3.45		4.64	0.23
5/Right	2.28		_	_		_	4.73	0.28
6/Left	1.95	0.05	2.69	0.08	3.41	0.11	4.54	0.01
6/Right	1.99	0.01	2.70	0.04	3.38	0.04	4.42	0.02

Absolute Latencies (in Milliseconds) at 62 dB peSPL Across All Ears and Test Sessions

Note. "--" represents a mean or standard deviation unable to be calculated due to insufficient data points across the three sessions.

Table 7

Grand Averaged Absolute Latencies (in Milliseconds) Across All Ears and Test Sessions

	Wave I			Wave II			Wave III			Wave V		
	N	М	SD	N	М	SD	N	М	SD	N	М	SD
102 dB peSPL	36	1.56	0.14	36	2.30	0.15	31	2.93	0.14	36	3.87	0.20
82 dB peSPL	31	1.79	0.14	33	2.58	0.14	30	3.29	0.18	36	4.19	0.21
62 dB peSPL	21	2.09	0.13	16	2.80	0.12	13	3.46	0.14	29	4.65	0.22



Figure 2. Latency-intensity function showing the grand averaged absolute latency values at each intensity level for waves I, II, III, and V.

Interpeak Latency

Interpeak latencies among waves I-II, II-V, and I-V were calculated at 102 dB peSPL only because the wave components at a high intensity were more identifiable compared to those at the lower intensities of 82 dB peSPL and 62 dB peSPL. Included interpeak waveform components were chosen based upon the wave components that occurred the most. Table 8 displays the means, standard deviations, and grand averages of all interpeak latency components for each ear at 102 dB peSPL. Table 9 displays the grand averaged interpeak latency of all data points from all sessions. Waves I-II mean interpeak latencies ranged from 0.67 ms to 0.82 ms with a grand average of 0.74 ms.

Waves II-V mean interpeak latencies ranged from 1.28 ms to 1.69 ms with a grand average of 1.57 ms. Waves I-V mean interpeak latencies ranged from 2.03 ms to 2.45 ms with a grand average of 2.31ms.

Table 8

Interpeak Latencies (in Milliseconds) at 102 dB peSPL Across All Ears and Test Sessions

Subject/Ear	Way	ves I-II	Wave	es II-V	Wav	es I-V
-	М	SD	М	SD	M	SD
1/Left	0.68	0.03	1.67	0.07	2.35	0.05
1/Right	0.67	0.04	1.69	0.08	2.36	0.06
2/Left	0.77	0.04	1.68	0.10	2.45	0.08
2/Right	0.76	0.02	1.28	0.19	2.03	0.21
3/Left	0.72	0.13	1.58	0.18	2.30	0.06
3/Right	0.82	0.05	1.56	0.03	2.39	0.04
4/Left	0.74	0.02	1.55	0.09	2.29	0.08
4/Right	0.71	0.05	1.50	0.07	2.21	0.10
5/Left	0.80	0.16	1.54	0.08	2.34	0.15
5/Right	0.80	0.05	1.40	0.12	2.20	0.10
6/Left	0.76	0.06	1.65	0.03	2.42	0.08
6/Right	0.67	0.11	1.69	0.06	2.37	0.15
Grand Avg	0.74	0.06	1.57	0.09	2.31	0.10

Table 9

Subject #	102	dB peSPL	82 dB	peSPL	62 dB	peSPL
-	M	SD	M	SD	M	SD
1	0.10	0.03	0.10	0.02	0.06	0.02
2	0.48	0.27	0.25	0.26	0.18	0.10
3	0.12	0.13	0.19	0.16	0.03	0.03
4	0.11	0.05	0.16	0.12	0.42	
5	0.12	0.09	0.06	0.06	0.06	
6	0.15	0.09	0.07	0.01	0.13	0.01
Grand Avg	0.18	0.11	0.14	0.10	0.15	0.04

Wave V Interaural Latency Differences for All Canines at Each Intensity Level Across Three Test Sessions

Note. "--" represents a mean or standard deviation unable to be calculated due to insufficient data points across the three sessions.

Wave V Interaural Latency Difference

Wave V interaural latency difference was calculated for each canine at each intensity level. Table 10 shows the means, standard deviations, and grand averages of wave V interaural latency differences for all canines at each intensity level. Mean wave V interaural latency differences ranged from 0.10 ms to 0.48 ms with a grand average of 0.18 ms at 102 dB peSPL, 0.06 ms to 0.25 ms with a grand average of 0.14 ms at 82 dB peSPL, and from 0.03 ms to 0.42 ms with a grand average of 0.15 ms at 62 dB peSPL.

Table 10

Subject/Ear	Wa	ve I	Wa	ve II	Way	/e III	Way	ve V
-	М	SD	M	SD	М	SD	M	SD
1/Left	0.47	0.20	0.46	0.11	0.61	0.11	1.51	0.13
1/Right	0.47	0.08	0.79	0.04	0.78	0.03	1.50	0.08
2/Left	0.34	0.09	0.23	0.09	0.19	0.08	0.35	0.10
2/Right	0.51	0.17	0.40	0.02	0.28	0.04	0.74	0.17
3/Left	0.27	0.09	0.32	0.05	0.12	0.09	0.91	0.14
3/Right	0.33	0.16	0.60	0.03	0.33	0.04	1.04	0.05
4/Left	1.16	0.06	0.25	0.00	0.58	_	1.68	0.09
4/Right	0.51	0.33	0.39	0.03	_	_	1.55	0.09
5/Left	0.30	0.08	0.57	0.11	0.17	0.05	0.93	0.18
5/Right	0.36	0.07	0.23	0.05	0.23	0.05	1.19	0.12
6/Left	0.53	0.33	0.51	0.05	0.62	0.06	1.67	0.03
6/Right	0.30	0.14	0.25	0.10	0.59	0.04	1.25	0.16
Grand Avg	0.46	0.15	0.42	0.05	0.40	0.06	1.19	0.11

Peak Amplitudes (in Microvolts) at 102 dB peSPL Across All Ears and Test Sessions

Note. "--" represents a mean or standard deviation unable to be calculated due to insufficient data points across the three sessions

Amplitude

Amplitude data were obtained for waves I, II, III, and V for all three sessions at each intensity level. Amplitudes between left and right ear of each canine were consistent at all three intensities with the largest mean amplitudes present at 102 dB peSPL and smallest mean amplitudes at 62 dB peSPL. Mean amplitude of wave V was the largest for all canines in both ears at both 102 and 82 dB peSPL; however, the largest mean amplitude waveform component across canines was much more variable at 62 dB peSPL. Tables 11, 12, and 13 show the amplitude means, standard deviations, and grand averages of waves I, II, III, and V of each ear at each intensity level for all canines. Table 14 shows the grand averaged amplitudes of all ears tested across all three test sessions.

Table 11

Peak Amplitudes (in Microvolts) at 82 dB peSPL Across All Ears and Test Sessions

Subject/Ear	Wa	ive I	Wa	ive II	Way	ve III	Wa	ve V
C C	M	SD	M	SD	M	SD	M	SD
1/Left	0.46	0.15	0.20	0.17	0.59	0.13	1.04	0.24
1/Right	0.46	0.13	0.58	0.08	0.72	0.19	0.99	0.13
2/Left	0.20	0.21	0.14	0.12	0.18	0.05	0.54	0.18
2/Right	0.60	0.28	0.21	0.06	0.16	0.08	0.79	0.13
3/Left	0.42	0.15	0.32	0.06	0.18	0.09	0.68	0.06
3/Right	0.36	0.10	0.24	0.12	0.19	0.22	0.67	0.26
4/Left	0.11	0.11	0.49	0.16			1.22	0.01
4/Right			0.22	0.06			0.84	0.11
5/Left	0.42	0.16	0.23	0.04	0.23	0.10	0.88	0.14
5/Right	0.22	0.20	0.12	0.05	0.20	0.06	0.93	0.12
6/Left	0.64	0.14	0.38	0.14	0.51	0.42	1.33	0.10
6/Right	0.51	0.04	0.42	0.09	0.37	0.20	1.15	0.19
Grand Avg	0.42	0.15	0.29	0.09	0.33	0.15	0.92	0.14

Note. "--" represents a mean or standard deviation unable to be calculated due to insufficient data points across the three sessions.

Table 12

Subject/Ear	Wa	ve I	Way	ve II	Way	ve III	Way	ve V
-	M	SD	M	SD	M	SD	M	SD
1/Left	0.11	0.04	0.16	0.12	0.59	0.15	0.29	0.14
1/Right	0.16	0.10	0.14	0.07	0.29		0.59	0.25
2/Left	0.10	0.07		—	0.25		0.22	0.06
2/Right	0.32	0.11	0.13		0.14		0.46	0.30
3/Left	0.22	0.13	0.17	0.18	0.13	0.03	0.38	0.29
3/Right	0.17	0.11	0.15	0.01	0.09		0.38	0.18
4/Left			—	—	—	—	0.54	
4/Right			—	—	—	—	0.51	
5/Left	0.28		0.15	—	0.12	—	0.40	0.20
5/Right	0.01		_	_	_	_	0.29	0.17
6/Left	0.18	0.14	0.15	0.07	0.44	0.14	0.62	0.35
6/Right	0.16	0.08	0.15	0.11	0.37	0.21	0.83	0.28
Grand Avg	0.17	0.10	0.15	0.09	0.30	0.13	0.45	0.22

Peak Amplitudes (in Microvolts) at 62 dB peSPL Across All Ears and Test Sessions

Note. "--" represents a mean or standard deviation unable to be calculated due to insufficient data points across the three sessions.

Table 13

Grand Averaged Peak Amplitudes (in Microvolts) Across All Ears and Test Sessions

		Wave	Ι	Wave II			Wave III			Wave V		
	Ν	М	SD	Ν	М	SD	Ν	М	SD	Ν	М	SD
102 dB peSPL	36	0.46	0.15	36	0.42	0.05	29	0.40	0.06	36	1.19	0.11
82 dB peSPL	31	0.42	0.15	33	0.29	0.09	30	0.33	0.15	36	0.92	0.14
62 dB peSPL	21	0.17	0.10	16	0.15	0.09	14	0.30	0.13	29	0.45	0.22

Overall Morphology

Overall morphology was rated for both ears of all canines across all sessions at each intensity level. Descriptive ratings of good, fair, and absent were used. A good rating consisted of identifiable and repeatable waves I, III, and V or I, II, and V. A waveform rated fair consisted of some identifiable wave components, reduced amplitude, and/or poor repeatability. An absent rating for overall morphology occurred when there was no identifiable waveform response. Figure 3 illustrates good, fair, and absent waveform morphologies.



Figure 3. Good, fair, and absent waveform morphologies from left to right.

The highest number of waveforms judged to be of good morphology occurred at 102 dB peSPL (100%). At 82 dB peSPL, waveforms were rated as either good or fair. At 62 dB peSPL, waveforms of all three ratings were obtained. Figure 4 shows the overall morphology ratings across sessions at each intensity level (see Appendix D for individual ratings for each canine).



Figure 4. Overall morphology ratings across sessions at each intensity level.

Consistency of waveform morphology across test sessions at 102 dB peSPL is depicted in Figure 5 for a single canine.



Figure 5. Repeatability of waveform morphology across test sessions at 102 dB peSPL for a single canine where the bottom two waveforms in each panel represent repeated waveforms, and the top waveform represents the averaged waveform.

Lowest Level Wave V Observed

Each canine was tested at three intensity levels including 102, 82, and 62 dB peSPL across three sessions. The lowest level a repeatable wave V was observed for each ear of all canines at each intensity level was recorded. The majority of canines had responses at 62 dB peSPL; however, several instances in which the lowest level wave V was identified occurred at 82 dB peSPL. Figure 6 is a representation of the lowest level wave V observed across all canines at each intensity level across test sessions.



Figure 6. Number of waveforms present at each intensity level that represent the lowest level wave V was observed across test sessions.

Statistical Analysis

Nonparametric statistics were used due to data not fitting a normal distribution. The Kruskal-Wallis test was performed to examine differences between absolute peak latencies, interpeak latencies, interaural differences, and peak amplitudes across sessions at each intensity level (see Appendix D). None of the analyses resulted in any statistically significant findings (p > 0.05).

In addition, statistical analyses were performed to investigate whether or not there were any ear effects for absolute peak latencies, interpeak latencies, and peak amplitudes (with the exception of interaural difference values since those incorporated information across both ears for each animal; see Appendix D). These analyses did not show any statistically significant differences, which supported our use of each ear as a separate value when calculating N for the various descriptive statistics reported. These non-

statistically significant findings suggested consistent waveform characteristics across ears and sessions at each intensity level.

CHAPTER V

DISCUSSION

The purpose of this study was to examine the consistency of waveform characteristics of the canine click-evoked BAER by comparing various latency and amplitude measures across multiple test sessions. Waveform characteristics analyzed throughout the study included absolute latencies of waves I, II, III, and V; interpeak latencies of waves I-II, II-V, and I-V; wave V interaural latency difference, and amplitudes of waves I, II, III, and V. In addition, overall morphology judgments and the lowest level a wave V was observed for three predetermined levels (102, 82, and 62 dB peSPL) were also compared across sessions. It was hypothesized that waveform characteristics would not vary significantly across sessions in canines when looking at absolute and interpeak latencies, interaural latency difference, amplitude, overall morphology, and lowest measured response when using a click-evoked BAER at different presentation levels.

Summary of Results

The results of this study showed the canine click-evoked BAER resulted in consistent waveform characteristics across multiple test sessions at different intensity levels. No previously published research has assessed the consistency of the canine click-evoked BAER; however, research has shown good test-retest reliability of waveform characteristics for human BAER testing (Jacobson, 1985; Munjal et al., 2016; Song, Nicol, & Kraus, 2010; Towers, Pisa, Froelich, & Krumm, 2005). All latency and amplitude values analyzed were not statistically significant (p > 0.05), suggesting high reliability of latency and amplitude values across test sessions. It was also noted that when looking at the standard deviations of values measured across test sessions, the variability tended to be small.

Latency

The canine clicked evoked BAER resulted in an increased latency as the intensity decreased as depicted in Figure 2. This finding was similar to published data for the BAER for normal hearing humans (Picton et al., 1976).

Plonek, Nicpoń, Kubiak, and Wrzosek (2017) reported mean absolute latencies for waves I, II, III, and V of normal hearing canines at a high intensity as 1.80 ± 0.03 ms, 2.59 ± 0.11 ms, 3.44 ± 0.23 ms, and 4.28 ± 0.26 ms, respectively, with an interpeak latency of wave I-V as 2.49 ± 0.26 ms. Kemper et al. (2012) reported canine mean latencies for waves I, II, III, IV, and V as follows: 1.46 ± 0.49 ms, 2.52 ± 0.54 ms, $3.45 \pm$ 0.41 ms, 4.53 ± 0.83 ms and 5.53 ± 0.43 ms, respectively, with a mean wave I–V interpeak latency of 3.69 ms. In the current study, 102 dB peSPL averaged mean latencies were reported for waves I, II, III, and V and all fell within the ranges reported by Kemper et al. except for wave III, which was slightly early by 0.11 ms, and wave V, which was much earlier by 1.23 ms. Table 15 shows absolute latency values reported by Kemper et al., Plonek et al., and data from the current study. Due to early wave V components in the current study, Figure 7 illustrates wave V latency distribution across all canines at 102 dB peSPL.

Table 14

	Study Parameters	Wave I	Wave II	Wave III	Wave V
Kemper et al. (2012)	Clicks 90 dB peSPL 33 clicks/s ≥ 2000 sweeps 300-1500 Hz filter	0.97-1.95	1.98-3.06	3.04-3.86	5.10-5.96
Plonek et al. (2017)	Clicks 105 dB nHL Click rate (NA) 300 Sweeps 150-3000 Hz filter	1.77-1.83	2.48-2.70	3.21-3.67	4.02-4.54
Current Study	Clicks 102 dB peSPL 33.3 clicks/s ≥ 1000 sweeps 300-1500 Hz filter	1.29-1.71	2.03-2.48	2.79-3.07	3.55-4.16

Comparison of Absolute Latency Values (+/- 1SD)



Figure 7. Distribution of wave V latency across all ears and test sessions at 102 dB peSPL. Yellow dots represent +2SD, green dots represent the mean, and red dots represent -2SD.

It is important to note that differences in latencies across all three studies compared in Table 15 could be due to varying test parameters across studies. Scheifele and Clark (2012) discussed the importance of a universal test protocol when using the BAER in canines to keep animal audiology in line with human audiology. Table 15 reinforced the need for a universal test protocol when using the BAER in canines so future researchers can tightly control for differences in test protocol and, as a result, be able to compare findings across studies and develop normative data.

While different testing parameters can result in varying BAER responses, in the current study, additional variables could have created differences across test sessions. One of these variables would be the state of the canine. Each canine had varying levels of panting and body movements throughout the testing timeframe that could have increased the amount of artifact. When looking at the vital signs recorded across all canines and test sessions, pulse rate was fairly consistent, ranging from 140 to 160 beats/minute. Respiration compared across all canines and test sessions also revealed consistent results ranging from 30-45 breaths/minute. Two canines were unable to have respiration measured due to increased panting; one canine could not have respiration measured at any of the three testing sessions while the other canine was unable to have respiration tested at one of the three testing sessions. However, it was interesting to note that while two of the canines had this increased panting, when looking at artifact amounts from these particular canines and testing sessions compared to the other canines, these particular canines did not have a larger number of artifacts compared to the other canines. Artifact ranges varied greatly from canine to canine across testing sessions: 6-255, 8-225,

1-145, 8-233, 4-199, 5-248 for canines 1, 2, 3, 4, 5, and 6, respectively (minimum of 1000 sweeps collected).

While interpeak latency components typically consist of I-III, III-V, and I-V for human responses, for this study, interpeak waveform components consisted of I-II, II-V, and I-V due to more wave II than wave III components present in the canine responses. As previously reported for both humans and canines, I-V interpeak latency is around 4ms (Atcherson, 2012; Kemper et al., 2012); however, the current study resulted in shorter interpeak latencies, although still consistent across test sessions. This discrepancy in wave V absolute latencies and waves I-V interpeak latencies could be due to differences in labeling strategies. It is possible that what was being labeled as wave V was actually wave IV. In humans, it is common for a wave IV-V complex to be present where either wave IV is prominent and wave V appears as a small shoulder or vice versa. This wave IV-V complex could have been present in the canines as well, causing wave IV to be incorrectly labeled as wave V. Throughout this study, the labeling strategy identified the most prominent peaks and used those as the labeled waveform components, taking into consideration the timeframe of those labeled waveform components. Wave IV-V complexes might have been missed in the current study because labeling on the shoulders of peaks was not a labeling strategy. Because the purpose of the present study was to determine repeatability of responses across test sessions and as long as the same strategy was used for each waveform, this difference in identifying peaks should not have altered the final conclusions. A possible wave IV-V complex with wave V labeled as what really might be wave IV is shown in Figure 8.



Figure 8. Possible wave IV-V complex causing the incorrect labeling of wave IV as wave V. The arrow indicates where wave V would have been marked if this is actually a wave IV-V complex.

Amplitude

The most robust waves in the human BAER are I, III, and V (Arnold, 2007). While this was mostly true for canine waveforms, at times wave II was much more prominent in the canine waveform compared to humans. Throughout this study, reported wave II and wave III amplitude measurements were very similar; however, there were more identifiable wave II components than wave III components. It is known that amplitude measurements are the least clinically useful during human BAER interpretation because of how easily the amplitudes are affected by the signal to noise ratio. However, Atcherson (2012) reported on the significance of the wave V to wave I amplitude ratio. When comparing the wave V amplitude to the wave I amplitude in humans, the amplitude of V is typically larger than that of wave I, which was also consistent in canine BAER waveforms throughout this study. Plonek et al. (2017) reported amplitudes of waves I, II, III, and V and also found that for all three intensities tested, wave V always had a larger amplitude compared to wave I in both the normal hearing canine group as well as the unilateral deaf canine group.

Overall Waveform Morphology

Picton et al. (1981) reported worsening overall morphology as the stimulus intensity decreased. When looking at overall waveform morphology throughout this study, it was clear that as intensity decreased, morphology became less clear. For the high intensity level of 102 dB peSPL, morphology appeared to be consistent and good across all test sessions with slightly more variability at 82 dB peSPL and no clear consistency at 62 dB peSPL. This decrease in morphology at 62 dB peSPL could have been due to testers being close to or below a canine's actual threshold. When converting the intensity used to a dB nHL value, the equipment used listed a correction factor of 32 dB, which would yield a 30 dB nHL value for 62 dB peSPL. It is very likely that when using 62 dB peSPL, researchers were close to threshold in some canines, resulting in decreased morphology that included a higher number of waveforms judged as fair as well as absent responses when compared to 102 dB peSPL. It could be concluded that good morphology occurred at high intensity levels but that could not be said for lower intensities especially if close to threshold. In a study by Strain, Tedford, and Jackson (1991), better morphology was also reported in canine BAER waveforms as intensity increased. Strain et al. reported increased amplitude and decreased latency with



Figure 9. Overall morphology comparison of a single canine brainstem auditory evoked response waveform at 102 dB peSPL and 62 dB peSPL.

Lowest Level Wave V Observed

For the majority of test sessions, wave V was observed at all three presentation levels. All 36 test runs (100%) had a repeatable wave V at 102 dB peSPL and 82 dB peSPL, while 29 (80%) had a repeatable wave V at 62 dB peSPL. Of the six canines tested, three of them exhibited different minimum levels at which wave V was recorded across sessions. Two of the canines had different outcomes with respect to presence/absence of wave V in both ears, while one canine only had a different outcome in the right ear. This study did not aim to obtain a true threshold estimation so in order to determine a canine's actual BAER threshold, smaller decreasing increments of intensity should be used to better estimate the lowest measured response from session to session. Instead of decreasing stimulus intensity in 20 dB increments, 5-10 dB increments are recommended, especially when testing at intensity levels below 82 dB peSPL.

Strengths and Limitations of Study

While this study showed consistency for all analyzed waveform characteristics across sessions and between ears, the sample size was relatively small. Testing was found to be more difficult in canines with more fur around their neck and ears compared to those with less hair. This was due to the ease with which subdermal electrodes were able to be placed in canines with little hair surrounding the electrode sites compared to those with much more hair at the electrode sites; however, all canines were cooperative throughout this study. In addition to cooperative test subjects, experienced research members and testing of both ears rather than just one were also strengths of this study.

An additional difficulty encountered was equipment malfunction during two testing sessions; however, the software was able to be restarted at the time of testing to obtain recordings at the scheduled time. Malfunction during the time of testing was due to the transducers not presenting a click through the insert earphones, resulting in false negative responses. Inconsistencies in waveforms were noticed and the equipment was checked. This malfunction was resolved by unplugging the transducers from the Intelligent Hearing System USB Box and restarting the equipment. Once the equipment was restarted, it was confirmed by the principal investigator and a member of the research committee that the transducers were presenting an appropriate click stimulus.

Intrinsic and extrinsic factors might have affected test outcomes including different test protocols, the picking of waves/peaks between testers, physical state of each canine, and differences in depth of insertion of foam insert earphones. During this study, foam insert earphones were not placed by the same person for each dog across sessions; however, results from this study showed that while insertion depth might have varied across testers, it did not significantly affect the results.

Implications and Future Directions

While no current research reported the consistency of the BAER in canines, there were studies reporting expected latency values (Kemper et al., 2012; Plonek et al., 2017). However, this current research also highlighted the need for a universally accepted protocol when testing canines in order to be able to obtain normative data and compare research findings across studies. A universal protocol and additional normative data would allow for further investigation of the consistency of the click-evoked canine BAER waveform across test sessions.

This study provided evidence that the canine click-evoked BAER across multiple test sessions had good consistency. Additional studies with more participants would also aid in better understanding waveform components including mean latencies of both absolute latencies (especially for waves IV and V) and interpeak latencies. Future research should also include exploring waveform measures for tone burst stimuli to gain more frequency specific information about the canine BAER. It would also be interesting to assess the estimated threshold of the canine click-evoked BAER across test sessions. Due to a consistent prominent wave II throughout this study, future studies should investigate the use of wave II-V as an interpeak measure rather than III-V as is typical for human responses.

Conclusions

In summary, the canine click-evoked BAER across multiple test sessions at different intensity levels was consistent for measurements of absolute and interpeak latencies, wave V interaural latency differences, and amplitudes. With respect to morphology, one could expect good morphology at higher intensity levels compared to lower intensity levels. The lowest level at which a repeatable wave V was observed was similar across all three test sessions for the majority of canines in this study. However, smaller, decreasing increments of stimulus intensity should be used to better determine this waveform characteristic. To better understand absolute latency of wave V and interpeak latencies, close examination of waveform morphology across subjects is needed to further evaluate prominence of peak II compared to peaks I and III, and to evaluate the prevalence of wave IV-V complexes.

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APPENDIX A

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE APPROVAL

NORTHERN COLORADO

IACUC Memorandum

To: Dr. Tina Stoody From: Laura Martin, Director of Compliance and Operations CC: Dr. Katie Bright and Hayden Bruce Date: May 11, 2017 Re: IACUC Protocol 1705C-TS-D-20 Approval

The UNC IACUC has completed a final review of your protocol "Test-Retest Reliability of the Canine Click Evoked BAER". The protocol review was based on the requirements of Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research, and Training; the Public Health Policy on Humane Care and Use of Laboratory Animals; and the USDA Animal Welfare Act and Regulations. Based on the review, the IACUC has determined that all review criteria have been adequately addressed. The PI/PD is approved to perform the experiments or procedures as described in the identified protocol as submitted to the Committee. This protocol has been assigned the following number 1705C- TS-D-20.

Please note: Since this is not for clinical purposes in normal dogs, the IACUC will need to inspect the clinic and will add this site to our semiannual inspection list for the duration of the project. Please contact me to set up an inspection date prior to initiation of the study.

The next annual review will be due before May 11, 2018. Sincerely,

Laura Martin, Director of Compliance and Operations

APPENDIX B

OWNER OF PARTICIPANT CONSENT FORM



Audiology and Speech Language Sciences School of Human Sciences College of Natural and Health Sciences University of Northern Colorado Gunter Hall, Room 1400, Campus Box 140 501 20th Street Greeley, Colorado 80639

Project Title: Test-Retest Reliability of the Canine Click Evoked Brainstem Auditory Evoked Response (BAER) Across Multiple Test Sessions

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You have been asked to allow your dog to receive a hearing test as part of a study being conducted at the University of Northern Colorado (UNC). Your dog will receive a Brainstem Auditory Evoked Response (BAER) test.

We will be using very tiny, subdermal needles placed in three (3) different locations on the dog. Lidocaine/Prilocaine (2.5%/2.5%) will be applied to these locations, before placing the electrodes, for numbing of the area. Foam insert earphones will be inserted into the ear canal of the ear being tested, and a click stimulus will be presented. This protocol may take up to 30 minutes for each ear, but typically lasts no longer than 5 minutes per ear.

The on-site veterinarian will provide a brief wellness examination of your dog before the ABR test, that will look at your dog's temperature, weight, respiration, heart rate, etc. Members of the research team will gently restrain your dog since the test requires the dog to stay relatively still. All hearing screenings/assessments will be analyzed and confirmed by two (2) audiologists and a veterinarian.

By signing below, you indicate that you understand that your dog's participation is voluntary and that you may withdraw your dog from the test at any time. You also understand that you remain responsible for your dog's health and behavior, and that UNC will not be responsible for injuries to your dog or injuries or property damage caused by your dog.

Dog Owner	Principal Investigator
Signed:	Signed:
Name:	Name:
Phone or Email:	Date:

APPENDIX C

PRE-RECORDING CHECKLIST

Checklist for Completion During Testing

- ____ Consent Form signed
- ____ Health check
- ____ Thundershirt™ placement
- ____ Lidocaine/Prilocaine placement
- ____ Subdermal needle placement
- ____ Impedance Check
- ____ Alternating polarity
- ____ 33.3 clicks/s
- _____1,000 sweeps
- ____ Filter: 100-1500 Hz
- ____ Absolute gain of 100,000
- ____ Artifact rejection rate of 35.1%
- ____ 102 dB peSPL x2
- ____ 82 dB peSPL x2
- ____ 62 dB peSPL x2
- ____ Save

APPENDIX D

KRUSKAL WALLIS RESULTS

Absolute Latency									
Intensity	Wave I	Wave II	Wave III	Wave V					
102 dB SPL	0.815 (36)	0.486 (36)	0.193 (29)	0.354 (36)					
82 dB SPL	0.161 (31)	0.541 (33)	0.1 (30)	0.539 (36)					
62 dB SPL	0.748 (21)	0.619 (16)	0.617 (13)	0.188 (28)					

Data reported as p-value (number of ears) across sessions

Amplitude								
Intensity	Wave I	Wave II	Wave III	Wave V				
102 dB SPL	0.786 (36)	0.566 (36)	0.394 (29)	0.912 (36)				
82 dB SPL	0.252 (31)	0.566 (33)	0.631 (30)	0.755 (36)				
62 dB SPL	0.145 (21)	0.566 (16)	0.543 (13)	0.315 (28)				

Data reported as p-value (number of ears) across sessions

Interpeak Latency								
Intensity	Waves I-II	Waves II-V	Waves I-V					
102 dB SPL	0.147 (36)	0.642 (36)	0.68 (36)					

Data reported as p-value (number of ears) across sessions

Absolute Latency								
Intensity	Wave I	Wave II	Wave III	Wave V				
102 dB SPL	0.739 (36)	0.68 (36)	0.793 (29)	0.113 (36)				
82 dB SPL	0.536 (31)	0.664 (33)	0.901 (30)	0.366 (36)				
62 dB SPL	0.777 (21)	0.664 (16)	0.714 (13)	0.662 (28)				

Data reported as p-value (number of ears) across ears

Amplitude									
Intensity	Wave I	Wave II	Wave III	Wave V					
102 dB SPL	0.937 (36)	0.42 (36)	0.541 (29)	0.912 (36)					
82 dB SPL	0.592 (31)	0.986 (33)	0.803 (30)	0.548 (36)					
62 dB SPL	0.972 (21)	0.874 (16)	0.245 (13)	0.28 (28)					

Data reported as p-value (number of ears) across ears

Interpeak Latency							
Intensity Waves I-II Waves II-V Waves I-V							
102 dB SPL	0.949 (36)	0.128 (36)	0.066 (36)				

Data reported as p-value (number of ears) across ears

APPENDIX E

RAW DATA

		Wa	ave I Ab	solute L	atency (ms)				
Subject and Ear	102	2 dB peS	SPL	82	dB peS	PL	62	62 dB peSPL		
1L	1.45	1.70	1.63	1.65	1.80	1.78	2.03	2.10	2.15	
1R	1.53	1.65	1.55	1.75	1.80	1.75	2.00	2.20	1.98	
2L	1.65	1.73	1.75	1.88	2.25	2.00	2.35	2.33		
2R	1.60	1.65	1.68	1.75	1.78	1.90	2.17	2.03		
3L	1.75	1.65	1.60	1.75	1.85	1.88	2.03		2.1	
3R	1.45	1.50	1.53	1.73	1.90	1.78	2.03		2.28	
4L	1.30	1.27	1.30	1.50	1.45		—			
4R	1.38	1.38	1.27		—		—			
5L	1.60	1.78	1.50	1.70	1.80	1.88	1.93			
5R	1.65	1.68	1.65	1.78	1.95	NR	2.28			
6L	1.55	1.38	1.53	1.75	1.73	1.70	1.90	2.00	1.95	
6R	1.73	1.50	1.63	1.90	1.73	1.75		2.00	1.98	
		Way	ve II Ab	solute La	atency (1	ns)				
Subject and	102	2 dB peS	SPL	82	dB peS	PL	62	dB peS	PL	
Ear	0.1.5			2.40	0.65		0.60	0.70	• • • •	
IL	2.15	2.35	2.33	2.40	2.65	2.55	2.63	2.78	2.80	
IR	2.20	2.28	2.25	2.42	2.55	2.42	2.70	2.83	2.73	
2L	2.38	2.53	2.53	2.63	3.03	2.73				
2R	2.38	2.40	2.42	2.55	2.60	2.67	2.85			
3L	2.33	2.42	2.42	2.55	2.67	2.67	2.88		2.95	
3R	2.30	2.35	2.30	2.60	2.70	2.73	2.93		3.08	
4L	2.03	2.03	2.03	2.48	2.33					
4R	2.05	2.08	2.03	2.80		2.38				
5L	2.30	2.50	2.48	2.50	2.67		2.80			
5R	2.40	2.48	2.50	2.58	2.73	2.73				
6L	2.25	2.20	2.30	2.53	2.45	2.48		2.63	2.75	
6R	2.40	2.28	2.20	2.60	2.48	2.42		2.73	2.67	

		Wav	e III Abs	solute La	atency (1	ns)				
Subject and	102	2 dB peS	SPL	82	82 dB peSPL			62 dB peSPL		
Ear										
1L	2.80	2.98	3.0	3.05	3.25	3.20	3.20	3.45		
1R	2.88	2.93	2.90	3.05	3.20	3.80	3.33			
2L	3.00	3.05	3.15	3.30	3.75	3.35	3.65			
2R	2.93	3.00	2.98	3.20	3.33	3.30		3.55		
3L	2.93	3.08	3.08	3.33	3.38	3.38	3.53		3.70	
3R	2.98	3.03	2.95	3.23	3.30	3.43	3.58			
4L	2.45									
4R										
5L	2.88		2.95	3.10	3.45	3.18	3.45			
5R	3.00	3.10	3.03	3.25	3.38	3.45				
6L	2.85	2.73	2.80	3.13	3.18	3.28		3.33	3.48	
6R		2.83	2.78	3.33	3.08	3.08		3.35	3.40	
		Wav	e V Abs	olute La	itency (r	ns)				
Subject and	102	2 dB peS	SPL	82	dB peS	PL	62	dB peS	PL	
Ear										
1L	3.83	4.08	3.93	4.00	4.47	4.22	4.43	4.60	4.78	
1R	3.95	4.00	3.85	4.10	4.35	4.15	4.50	4.63	4.30	
2L	4.10	4.10	4.28	4.30	4.65	4.43	4.70	4.75	5.08	
2R	3.83	3.70	3.50	4.35	4.10	4.28	4.60	4.60	4.78	
3L	4.10	3.98	3.83	4.28	4.40	4.30	4.40	5.00	4.72	
3R	3.83	3.93	3.88	3.90	4.30	4.40	4.35	5.05	4.72	
4L	3.53	3.53	3.68	3.80	3.78	3.80	4.63			
4R	3.48	3.65	3.53	4.05	3.98	3.83	5.05			
5L	3.78	4.13	3.98	4.10	4.30	4.38	4.47		4.80	
5R	3.80	4.00	3.78	4.10	4.18	4.33	4.53		4.93	
6L	3.90	3.88	3.93	4.35	4.22	4.15	4.53	4.53	4.55	
6R	4 1 5	3 98	3 83	4 4 3	4 1 5	4 08		4 40	4 4 3	

	Interpeak Latencies (ms) at 102 dB peSPL								
Subject and	V	Wave I-I	Ι	V	Vave II-	V	I	Wave I-V	V
Ear									
1L	0.70	0.65	0.70	1.68	1.73	1.60	2.38	2.38	2.30
1 R	0.67	0.63	0.70	1.75	1.72	1.60	2.42	2.35	2.30
2L	0.73	0.80	0.78	1.72	1.57	1.75	2.45	2.37	2.53
2R	0.78	0.75	0.74	1.45	1.30	1.08	2.23	2.05	1.82
3L	0.58	0.77	0.82	1.77	1.56	1.41	2.35	2.33	2.23
3R	0.85	0.85	0.77	1.53	1.58	1.58	2.38	2.43	2.35
4L	0.73	0.76	0.73	1.50	1.50	1.65	2.23	2.26	2.38
4R	0.67	0.70	0.76	1.43	1.57	1.50	2.10	2.27	2.26
5L	0.70	0.72	0.98	1.48	1.63	1.50	2.18	2.35	2.48
5R	0.75	0.80	0.85	1.40	1.52	1.28	2.15	2.32	2.13
6L	0.70	0.82	0.77	1.65	1.68	1.63	2.35	2.50	2.40
6R	0.67	0.78	0.57	1.75	1.70	1.63	2.42	2.48	2.20

Wave V Interaural Latency Difference (ms)										
Subject	102	102 dB peSPL			82 dB peSPL			62 dB peSPL		
1	0.12	0.08	0.08	0.10	0.12	0.07	0.07	0.03	0.48	
2	0.27	0.40	0.78	0.05	0.55	0.15	0.10	0.15	0.30	
3	0.27	0.05	0.05	0.38	0.10	0.10	0.05	0.05	0.00	
4	0.05	0.12	0.15	0.25	0.20	0.03	0.42			
5	0.02	0.13	0.20	0.00	0.12	0.05	0.06			
6	0.25	0.10	0.10	0.08	0.07	0.07		0.13	0.12	

	Wave I Amplitude (µV)								
Subject and	102	2 dB peS	SPL	82	dB peS	PL	62 dB peSPL		
Ear									
1L	0.48	0.27	0.67	0.41	0.35	0.63	0.15	0.11	0.08
1R	0.51	0.38	0.51	0.61	0.38	0.40	0.27	0.11	0.09
2L	0.26	0.43	0.34	0.45	0.07	0.09	0.05	0.15	
2R	0.52	0.67	0.34	0.77	0.76	0.28	0.24	0.39	
3L	0.16	0.32	0.32	0.57	0.43	0.27	0.31		0.12
3R	0.37	0.46	0.15	0.27	0.33	0.47	0.24		0.09
4L	1.21	1.18	1.10	0.19	0.03				
4R	0.50	0.18	0.84						
5L	0.22	0.30	0.38	0.59	0.41	0.27	0.28		
5R	0.34	0.44	0.31	0.36	0.08		0.01		
6L	0.70	0.75	0.15	0.79	0.61	0.51	0.33	0.14	0.06
6R	0.29	0.44	0.17	0.52	0.55	0.47		0.10	0.22

Wave II Amplitude (µV)									
Subject and Ear	102 dB peSPL			82 dB peSPL			62 dB peSPL		
1L	0.42	0.53	0.42	0.39	0.15	0.05	0.14	0.06	0.29
1R	0.86	0.75	0.75	0.66	0.51	0.57	0.20	0.14	0.07
2L	0.39	0.10	0.19	0.27	0.03	0.13			
2R	0.24	0.64	0.32	0.27	0.15	0.21	0.13		
3L	0.19	0.46	0.30	0.27	0.38	0.32	0.30		0.04
3R	0.64	0.55	0.61	0.22	0.37	0.13	0.14		0.16
4L	0.22	0.21	0.33	0.38	0.60				
4R	0.37	0.25	0.54	0.26		0.18			
5L	0.28	0.92	0.50	0.26	0.20		0.15		
5R	0.28	0.30	0.12	0.17	0.11	0.08			
6L	0.66	0.67	0.20	0.34	0.53	0.26		0.20	0.10
6R	0.45	0.22	0.07	0.49	0.44	0.32		0.07	0.22
		W	Vave III	Amplitu	de (µV)				
Subject and	102 dB peSPL			82	dB peS	PL	62	dB peS	PL
Ear		-			-			-	
1L	0.61	0.77	0.44	0.72	0.59	0.47	0.69	0.48	
1R	0.88	0.94	0.53	0.93	0.58	0.65	0.29		
2L	0.36	0.12	0.10	0.18	0.23	0.13	0.25		
2R	0.29	0.31	0.23	0.24	0.09	0.16		0.14	
3L	0.05	0.24	0.07	0.08	0.24	0.23	0.15		0.11
3R	0.23	0.25	0.52	0.11	0.44	0.02	0.09		
4L	0.58								
4R									
5L	0.32		0.01	0.29	0.11	0.29	0.12		
5R	0.10	0.30	0.30	0.23	0.25	0.13			
6L	0.24	1.10	0.53	0.95	0.45	0.12		0.54	0.34
6R		0.75	0 4 3	0 14	0.53	0.44		0 22	0.52

Wave V Amplitude (μV)									
Subject and	102 dB peSPL			82 dB peSPL			62 dB peSPL		
Ear		-			-			_	
1L	1.40	1.60	1.52	1.04	0.75	1.32	0.33	0.41	0.14
1R	1.29	1.52	1.68	0.90	1.10	0.97	0.47	0.43	0.88
2L	0.20	0.72	0.14	0.71	0.47	0.43	0.28	0.19	0.18
2R	0.65	0.88	0.68	0.69	1.10	0.57	0.27	0.31	0.81
3L	0.63	1.18	0.92	0.66	0.74	0.64	0.66	0.09	0.40
3R	1.24	0.86	1.03	0.93	0.51	0.56	0.58	0.24	0.33
4L	1.87	1.70	1.47	1.51	0.99	1.16	0.54		
4R	1.65	1.19	1.81	0.57	0.73	1.21	0.51		
5L	1.46	0.63	0.71	1.34	0.84	0.47	0.54		0.26
5R	1.53	1.00	1.05	1.18	1.25	0.36	0.41		0.17
6L	1.59	1.84	1.58	1.43	1.14	1.42	1.03	0.43	0.41
6R	1.27	1.05	1.44	0.68	0.91	1.77		1.03	0.63

Wave V Lowest Measured Response (dB peSPL)								
Subject and Ear	Session 1	Session 2	Session 3					
1L	62	62	62					
1R	62	62	62					
2L	62	62	62					
2R	62	62	62					
3L	62	62	62					
3R	62	62	62					
4L	62	82	82					
4R	62	82	82					
5L	62	82	62					
5R	62	82	62					
6L	62	62	62					
6R	82	62	62					

Overall Morphology Rating									
Subject and	102 dB peSPL			82 dB peSPL			62 dB peSPL		
Ear									
1L	good	good	good	good	good	good	good	good	fair
1R	good	good	good	good	good	good	good	good	good
2L	good	good	good	good	good	good	good	good	good
2R	good	good	good	good	good	good	good	fair	fair
3L	good	good	good	good	good	good	good	good	fair
3R	good	good	good	good	good	good	good	fair	good
4L	good	good	good	good	good	good	good	fair	good
4R	good	good	good	good	good	fair	fair	poor	poor
5L	good	good	good	fair	fair	fair	fair	poor	poor
5R	good	good	good	good	good	fair	fair	poor	fair
6L	good	good	good	good	good	good	fair	good	good
6R	good	good	good	good	good	good	poor	good	good

Note. All tables with three rows under stimulus intensity levels represents sessions one, two, and three, respectively.