

USAARL Report No. 2018-22

# Assessment of Middle Ear Function during the Acoustic Reflex Using Wideband Acoustic Reflectance

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**United States Army Aeromedical Research Laboratory**

**Warfighter Performance Group**

September 2018

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<b>1. REPORT DATE (DD-MM-YYYY)</b> 25-09-2018		<b>2. REPORT TYPE</b> Final		<b>3. DATES COVERED (From - To)</b>	
<b>4. TITLE AND SUBTITLE</b> Assessment of Middle Ear Function during the Acoustic Reflex Using Wideband Acoustic Reflectance				<b>5a. CONTRACT NUMBER</b>	
				<b>5b. GRANT NUMBER</b>	
				<b>5c. PROGRAM ELEMENT NUMBER</b>	
<b>6. AUTHOR(S)</b> Greene, Nathaniel T.; Jones, Heath G.; Ahroon, William A.; Deiters, Kristy K.; Tasko, Stephen M.; Flamme, Gregory A.				<b>5d. PROJECT NUMBER</b>	
				<b>5e. TASK NUMBER</b>	
				<b>5f. WORK UNIT NUMBER</b>	
<b>7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)</b> U.S. Army Aeromedical Research Laboratory P.O. Box 620577 Fort Rucker, AL 36362				<b>8. PERFORMING ORGANIZATION REPORT NUMBER</b> USAARL 2018-22	
<b>9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)</b> U.S. Army Medical Research and Materiel Command 810 Schreider Street Fort Detrick, MD 21702-5000				<b>10. SPONSOR/MONITOR'S ACRONYM(S)</b> USAMRMC	
				<b>11. SPONSOR/MONITOR'S REPORT NUMBER(S)</b> N/A	
<b>12. DISTRIBUTION/AVAILABILITY STATEMENT</b> Approved for public release; distribution unlimited.					
<b>13. SUPPLEMENTARY NOTES</b> The Geneva Foundation; Laulima Government Solutions, LLC; Western Michigan University; Stephenson & Stephenson Research and Consulting					
<b>14. ABSTRACT</b> The Auditory Hazard Assessment Algorithm for Humans (AHA AH) employs the response of the Middle Ear Muscle Contraction (MEMC) as a protective mechanism against high-level impulse noise. The "warned" option in the AHA AH model assumes that the MEMC is conditioned to activate prior to the noise when a person fires a weapon. This report details procedures used to evaluate MEMC response to both acoustic and non-acoustic stimuli, as well as three tasks for eliciting a conditioned MEMC in a group of normal hearing adults. The tasks varied on the sensory modality of the conditioning stimulus. Using the techniques described here, a conditioned response could be detected as an MEMC occurring prior to the unconditioned stimulus and when only the conditioned stimulus was presented. The techniques reported here will be used in future studies at USAARL, which will monitor MEMCs occurring during live fire of small arms at a rifle range on Fort Rucker.					
<b>15. SUBJECT TERMS</b> Wideband reflectance measures, middle ear muscle contraction (MEMC), acoustic reflex					
<b>16. SECURITY CLASSIFICATION OF:</b>			<b>17. LIMITATION OF ABSTRACT</b> SAR	<b>18. NUMBER OF PAGES</b> 38	<b>19a. NAME OF RESPONSIBLE PERSON</b> Loraine St. Onge, PhD
<b>a. REPORT</b> UNCLAS	<b>b. ABSTRACT</b> UNCLAS	<b>c. THIS PAGE</b> UNCLAS			<b>19b. TELEPHONE NUMBER (Include area code)</b> 334-255-6906

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## Summary

### Background

Middle ear muscle contractions (MEMCs) are involuntary activations of the stapedius and/or tensor tympani muscles of the middle ear. MEMCs can occur in response to a range of acoustic (i.e. acoustic reflexes) and non-acoustic stimuli. Responses are generally assumed to be bilateral and have the effect of increasing middle ear impedance for low frequencies (< 1k Hz). Increased middle ear impedance reduces the transfer of acoustic energy through the auditory system, and potentially serves as protection from intense sounds such as those observed during blasts. Damage-Risk Criteria (DRC), which guide decisions about the risk of hearing impairment from impulsive noise exposures, have included MEMC as a form of hearing protection, either as acoustic reflexes or in anticipation of imminent exposure. DRC inclusion assumes that MEMC are pervasive (defined as 95% confidence of 95% prevalence) within the population and are of sufficient strength and duration to serve as a protective mechanism. The overall goal of this project was to develop techniques to address these assumptions directly.

### Purpose

The overall goal of this project was to develop techniques to directly address assumptions about the state of MEMCs in response to various acoustic and non-acoustic stimuli. In addition, these techniques can be used to detect MEMC occurring prior to an elicitor stimulus and directly test the conditionality of the middle-ear muscles.

### Methods

In this study, MEMC were elicited using various stimuli and wideband acoustic reflectance was used to detect response. Briefly, click trains were played out of an ear insert phone and the click reflection of the ear drum was recorded by microphone placed in the same ear insert. Any difference in the root-mean-square (RMS) amplitude of clicks recorded after presentation of elicitor from the average of baseline click would indicate a MEMC occurred. In addition, while the MEMC elicitors were presented to subjects, electromyographic (EMG) activity of selected head and neck muscles were simultaneously recorded to identify any relationship between MEMC and other concomitant muscle activity.

### Conclusions

Using the techniques reported here, the appropriateness of including MEMCs as a protective mechanism in the Auditory Hazard Assessment Algorithm for Humans (AHHAH) model when calculating auditory injury risk for impulsive noise can be tested. Knowledge gained from this study will be used to inform updates to Damage-Risk Criteria that will improve predictions of impulse noise limits. Ultimately, the military wants to know how much hearing damage the Warfighter risks when firing particular weapons or when exposed to a particular impulse. Hearing risk assessment models based on scientific evidence increases the ability of the military to achieve this objective and helps better protect the hearing of Warfighters exposed to high-level acoustic impulses.

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## **Acknowledgments**

We would like to acknowledge the assistance of Ms. Lana Milam for help with executing the current study. We appreciate the feedback provided by Dr. Stephanie Karch on study protocols and preparation of this report. This work was funded by a DoD Injury Prevention, Physiological and Environmental Health Award (IPPEHA) grant from the Telemedicine and Advanced Technology Research Center (TATRC), an office of the US Army Medical Research and Materiel Command (USAMRMC), award number W81XWH-14-2-0140.

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## Table of Contents

	<b>Page</b>
Introduction.....	1
Setup .....	3
Equipment.....	3
Station 1: .....	3
Station 2: .....	6
Middle ear muscle activity .....	6
Acoustical stimulation .....	7
Electromyography.....	7
Trigger pull detection.....	8
MEMC Detection.....	10
Stimuli.....	10
Response summarization .....	11
Data collection timeline .....	12
Enrollment session.....	12
Data Collection Session.....	12
Data collection procedures and preliminary results.....	13
Questionnaires.....	13
Video Otoscopy .....	14
Audiometric assessment.....	14
Cranial nerve assessment.....	15
Tympanometry .....	15
MEMC probe frequency sweep .....	15
EMG control tasks .....	16
Reflexive MEMC.....	16
Acoustic Elicitors.....	16
Non-Acoustic Elicitors.....	18
Conditioned MEMC Tasks .....	20
Simulated Trigger .....	20
Shooter/Spotter .....	22
Conclusions.....	25
References.....	26

## List of Figures

1. The anatomy of the external, middle and inner ear. ....	1
2. The middle-ear cavity .....	2
3. Station 1 equipment. ....	3
4. Audiometric assessment.....	4
5. Cranial Nerve assessment .....	5
6. Tympanometric assessment .....	6
7. ER-10X probe system.....	7

**Table of Contents (continued)**

**List of Figures (continued)**

	<b>Page</b>
8. EMG sensor locations.....	8
9. Voltage divider circuitry.....	9
10. Trigger pull detection.....	10
11. MEMC detection method.....	11
12. Acoustically elicited MEMCs.....	17
13. Eye-close elicited MEMCs.....	18
14. Air puff elicited MEMCs.....	19
15. Simulated Trigger (ST) condition.....	21
16. Shooter (SH) condition.....	23
17. Spotter (SP) condition.....	24
18. Comparison of SH and SP conditions.....	25

**List of Tables**

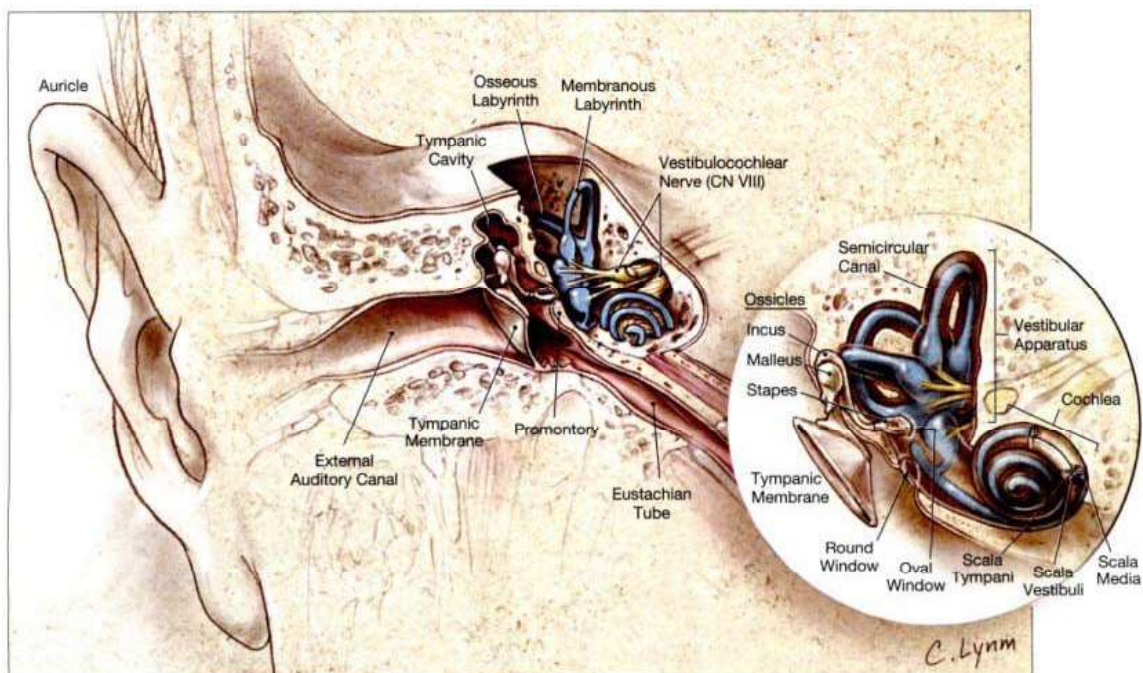
1. EMG sensor locations.....	16
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## Introduction

All air-borne sound passes through the three major divisions of the ear: the outer, middle and inner ear (Figure 1). The outer ear consists of the auricle or pinna, and funnels acoustic air-borne energy to the ear canal and the tympanic membrane (TM). The middle-ear is an air-filled cavity between the outer and inner ear and houses the TM, middle-ear muscles, and the ossicular chain (i.e., malleus, incus, and stapes). The TM converts acoustic energy in the ear canal into vibrations of the ossicular chain (comprised of the malleus, incus, and stapes), which transmits that energy to the fluid of the inner ear. These pressure waves within the cochlear fluid then stimulate the auditory sensory cells that sit on the basilar membrane within the cochlea. Electrical impulses are imparted from the cochlea via the eighth cranial nerve (vestibulocochlear nerve) to the central nervous system.

The most important function of the middle ear is to match the impedance of the normal sound transmission medium in the outer ear (air) to the inner ear (fluid). The middle-ear amplifies the acoustic energy impinging on the TM by a factor of approximately 18.6, due to the area ratio of the effective surface of the TM to the footplate of the stapes (Durrant & Lovrinic, 1984), and the lever action of the middle-ear ossicles.

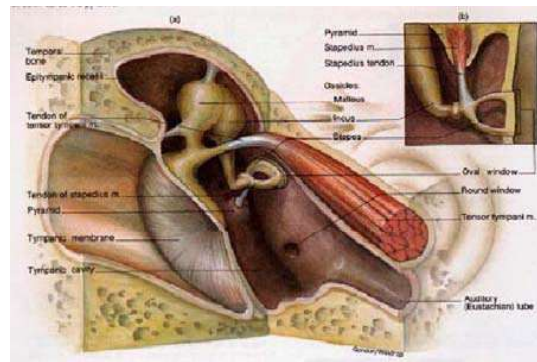
The middle-ear system also includes two muscles that can affect sound transmission to the inner ear (Figure 1). The first is the stapedius muscle, which attaches to the neck of the stapes from the apex of the pyramidal eminence and is innervated by a branch of the seventh cranial



*Figure 1.* The anatomy of the external, middle, and inner ear. From Yueh, Shapiro, MacLean and Shekelle (Yueh, Shapiro, MacLean, & Shekelle, 2003).



nerve (facial nerve). When contracted, it pulls the stapes away from the oval window of the cochlea. It is thought that the stapedius has the greatest effect on low frequency sounds, reducing transmission to the inner ear by up to 20 dB. The stapedius appears to be effective in reduction of sounds below about 1 kHz and apparently provides little effect to the higher frequencies (Feeny & Keefe, 1999). The second muscle, the tensor tympani (TT), inserts into the manubrium (handle) of the malleus, and is innervated by the fifth cranial nerve (trigeminal nerve), and pulls the malleus medially when contracted, thus tensing the TM. The TT motor neurons are located in the same area supplying the masseter and digastric muscles and course with the fifth cranial nerve (Mukerji, Windsor, & Lee, 2010).



*Figure 2.* The middle-ear cavity. The large red structure is the tensor tympani muscle. The tendon attaching the stapedius muscle to the stapes is located near the vertical center, slightly to the left of horizontal center, and depicted in the insert at the upper right).

In response to high-intensity acoustic stimuli, the acoustic reflex is elicited, simultaneously contracting the stapedius and TT muscles in both ears. The contraction of both muscles alters the acoustic energy entering the inner ear. As noted above, the TT muscle tenses the TM, thereby reducing the impedance match between the outer and inner ears. The stapedius muscle dampens the ability of the stapes footplate to move as a piston in the cochlea's oval window. During stapedius contraction, the stapes movement changes from a “pumping” action to more of a “rocking” action, thus reducing further the energy transfer into the cochlea. The contraction of the stapedius is thought to have the dominant effect on sound transmission in humans, but TT contractions could contribute and provide protection against acoustic trauma (Klockhoff & Anderson, 1960).

Middle-ear muscle contractions (MEMCs) can be elicited by non-acoustic stimulation (e.g., cutaneous stimulation, air blowing towards the face, disturbance of the eyelids) (Djupestrand, 1976; Mukerji et al., 2010) and voluntarily (Burns, Harrison, Bulen, & Keefe, 1993). The muscles also contract when swallowing, and it is possible that the TT and tensor veli palatini muscles are a functional unit involved in Eustachian tube function (Foss, Ison, Torre, & Wansack, 1989a; Grosse & Brown, 2003). In addition, there is a relationship between MEMC and the acoustic startle, which is a response to abrupt high-level stimuli that involves many muscles (Grosse & Brown, 2003), including the TT (Djupestrand, 1976) and has been shown to have a weak relationship with MEMC (Foss et al., 1989a; Foss, Ison, Torre, & Wansack, 1989b). The acoustic startle response is more closely tied to TT activity (Klockhoff & Anderson, 1960). It is unknown, however, how non-acoustic, startle-based, or voluntary MEMCs compare (in duration, strength, etc.) with reflexive MEMCs.

This project is designed to consider reflexive and classically conditioned activations of the middle-ear musculature in the presence of acoustic impulses. In particular, this report details a method, modified from a wide-band acoustic reflectance method described previously (Feeney & Keefe, 1999), developed to measure MEMCs in subject during both passive and active tasks. Here, we describe in detail the experimental setup, tasks, acoustic and non-acoustic stimuli, and preliminary analysis tools, developed to probe the association between MEMCs, the external stimuli, and independent muscle contractions of volunteer participants.

## Experimental Setup

The current study was conducted under the supervision of the U.S. Army Aeromedical Research Laboratory (USAARL) Regulatory Compliance Office and the U.S. Army Medical Research and Materiel Command Institutional Review Board. Study participants were recruited from in and around Fort Rucker, AL, and self-screened for age (19 [18 if Active Duty] years or older), good health with normal hearing, and regular occupational or recreational shooting experience. Subjects reviewed and signed an informed-consent prior to enrollment in the study, and were paid for their participation (if participating in an “off-duty” status).

### Equipment

The procedures described in this protocol are performed using two stations. The first includes equipment required to perform an enrollment eligibility screening, and the second includes the hardware necessary for measurements of middle ear muscle activity.

#### Station 1

Eligibility criteria for this protocol require normal hearing ( $< 10$  dB HL for frequencies 1 kHz and below, and  $< 20$  dB HL for higher frequencies), normal facial (CN VII) and trigeminal (CN V) nerve activity, and normal middle-ear function. The equipment is installed inside a small, double-walled, audiometric sound booth (ID: 4' x 5'), and interfaces with a PC (Dell



*Figure 3.* Station 1 equipment. Left: Control PC, NI PXI system, and ViAcoustics response box. Center: remote PC monitor, Titan system (upper left), video otoscope (upper right). Right: Sennheiser HD 200 headphones, Brüel & Kjær Nexus microphone power supply, and artificial ear.

Precision™ tower 5810) installed at a control station outside of the booth. Data collection is controlled by a custom MATLAB (MathWorks) script running on the control PC that interfaces with the experimental hardware and ensures consistent study procedures. Four sets of procedures are performed to assess eligibility for the study.

First, the subject's ears are inspected via video otoscopy. A custom MATLAB (MathWorks) script provides a real-time display from the otoscope camera to allow visualization of the ear canal and TM on each side of the head in order to ensure that there are no anatomical abnormalities that would prevent the use of the study ear probes, and to ensure there is no excess hair, impacted cerumen (ear wax), etc. Each video is saved to the PC and coded with the subject's identification number for offline review.

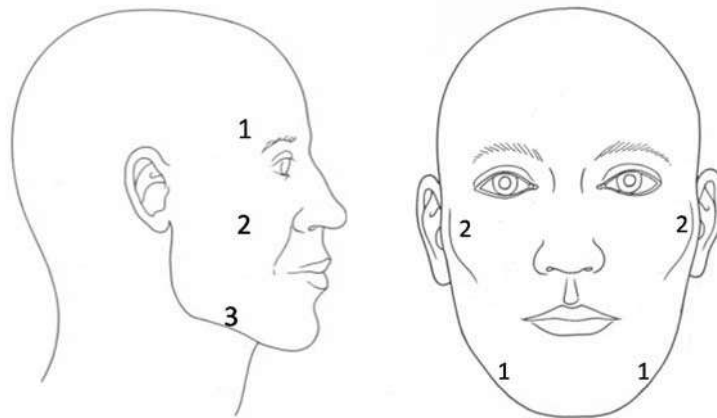
Second, a hearing test is performed using the ViAcoustic Nelson Audiometric Research Tool (ART) software. The hearing test is performed with Sennheiser HD 200 over-the-ear headphones, and can (if desired) use a radioear bone conduction transducer. ART was developed in the LabView (National Instruments) development environment, and was designed to run on National Instruments PXI-4461 or PXI-6220 series hardware. The ART software is a Hughston-Westlake based audiometer, utilizing an up-down staircase (adaptive-tracking) procedure. Briefly, the hearing test proceeds as such: The software begins the test with a specified frequency, presenting the stimulus at 30 dB HL in one ear. If the subject hears the stimulus and responds (via button press), the stimulus level is decreased by 10 dB. The stimulus level is then lowered repeatedly until the subject no longer responds to the stimulus, at which point the software reports a 'reversal'. The stimulus level is then *increased* in 5 dB increments until the subject again responds to the stimulus, at which point the software records another reversal. The stimulus level is increased and decreased, and the percentage of correct responses is tracked at each stimulus level, until the subject has responded to a particular stimulus level in at least 50%



Figure 4. Left: Welch-Allyn Digital MacroView Otoscope. Right: Nelson Audiometric Research Tool (ART) screenshot.

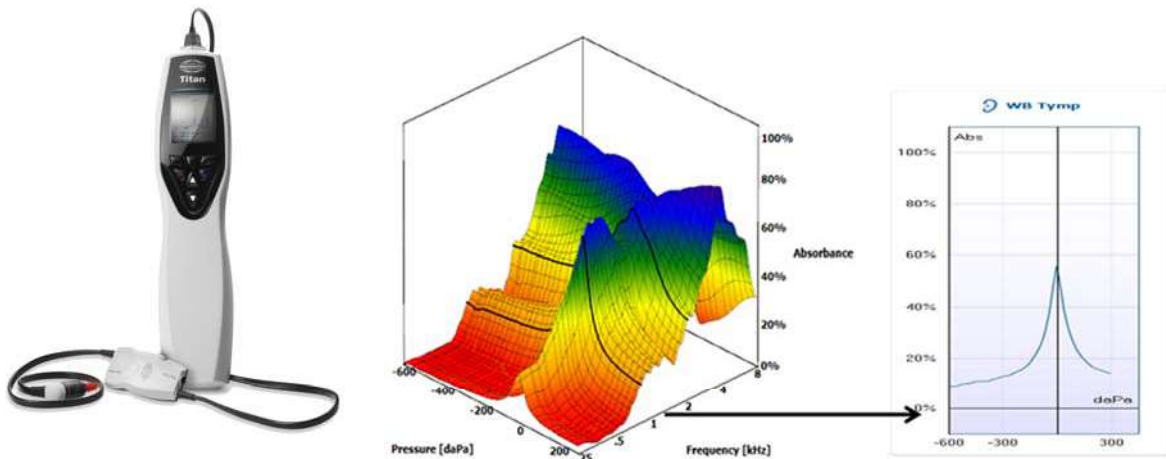
of three trials, and did not respond to a lower stimulus level in at least 50% of three trials. Threshold is then set at the higher of these two levels. Once threshold has been determined at one level, the software repeats this procedure for each of the remaining frequencies specified, and then repeats these procedures for each frequency in the opposite ear. The frequencies tested, the thresholds obtained, and the response history data for each determination are saved to an excel spreadsheet coded with the subject's identification number for later review.

Third, CN V and VII function is verified via an examination of the tactile and motor function of the face in the following order: (1) with the subject's eyes closed, the experimenter gently touches a cotton ball to the temple, cheek, and jaw on both sides of the face, (2) the muscles of the jaw and temple are palpated while the subject repeatedly clenches and unclenches his or her teeth, (3) the experimenter attempts to open and close the subject's mouth while the subject holds his or her mouth closed or open (resisting the experimenter), (4) with the subject looking at the experimenter's face, the subject raises his or her eyebrows, smiles, and pinches his or her nose while puffing his or her cheeks full of air, (5) with the subject's eyes closed, the experimenter gently attempts to pull open the subject's eyelids. Any failure to respond, weakness, or asymmetry in the subject's responses is recorded as a failure in the screening.



*Figure 5.* Locations on the face to palpate to test for tactile (Left), and motor (Right) function related to CN V & VII function.

Fourth, standard pure-tone tympanometry, as well as wide-band tympanometry and wide-band absorbance, and bilateral acoustic reflex thresholds, as well as reflex decay, are assessed with a Titan/IMP440 (Interacoustics A/S; Middlefart, Denmark) middle ear analyzer system. Tympanometry involves measuring acoustic emittance (via presentation and recording of a probe tone in the ear canal) while applying positive and negative pressure to the ear canal; the probe is coupled to the ear canal via a soft rubber eartip. Positive and negative pressure restrict tympanic membrane motion, thus altering ear canal emittance, resulting in a function with a distinct peak near zero pressure. Substantial deviations from zero, or a shallow peak, indicate tympanic membrane or middle ear dysfunction, thus represent a failure on this test. Wide-band tympanometry and admittance are comparable, but test middle ear function as a function of



*Figure 6.* Interacoustics Titan/IMP440 system. Left: the hand-help hardware. Center: Wideband tympanometry results showing absorbance as a function of frequency and pressure. Right: tympanometry results revealing absorbance as a function of pressure.

probe frequency. Acoustic reflex testing similarly involves presentation of a probe tone to the ear, coupled with presentation of a louder ‘elicitor’ tone to the same or opposite ear (via an E-A-RTone Gold 3A insert earphone, 3M Auditory Systems, Indianapolis, IN). Acoustic reflexes are assessed for a range of elicitor frequencies (0.5 – 4 kHz) and levels (80 – 100 dB) to determine reflex threshold and growth with stimulus level. A reflex decay function is recorded at a suprathreshold level to verify that the reflex does not decay for a 10 second stimulus.

## Station 2

Primary data collection occurs in a separate location, with different systems than are used in station 1. Station 2 is controlled by a PC (Dell Precision™ Tower 7910) running MATLAB (MathWorks), which controls experiment timing, stimulus playback, and data recording. Analog I/O is performed via a National Instruments PXI system. Sound stimuli are digitally generated in MATLAB, and output via an NI PXI-4461 card. Analog inputs are recorded via an NI PXI-4498 card. Digital I/O, as well as additional analog outputs, are controlled via a Tucker Davis Technologies (TDT) RP2.1 Enhanced Real-Time Processor. These I/O channels are necessary to control stimulus presentation and record the acoustical and physiological signals produced during the course of data collection.

## Middle ear muscle activity

The Etymotic ER-10X Extended bandwidth research probe system was designed to assess broadband distortion product otoacoustic emissions (dpOAEs), incorporating two independently driven drivers and a microphone into a package small enough to insert into the external ear. Here, we utilize one of these drivers to produce a broadband click, which we then record via the microphone, to detect changes in middle ear muscle activity via wide-band





Figure 7. Etymotic ER-10X Extended bandwidth probe system.

acoustic reflectance (Feeney & Keefe, 1999). The ER-10X probe utilizes disposable multi-lumen probe-tubes to allow the microphone and stimulus ports to align flush with the end of the eartip. The probe was designed to couple to the ear canal with a soft rubber ear tip; however, adapting a foam OAE eartip produces more stable placement in the ear canal, and more reliable results. The primary driver of the ER-10X is driven by one of the analog outputs of the NI PXI-4461, and the microphone channel is recorded by one of the inputs on the NI PXI-4498.

### Acoustical stimulation

Acoustic stimuli are presented to the ear contralateral to the ER-10X probe with an Etymotic ER-4PT insert earphone. The earphone is driven by the second NI PXI-4498 analog output (or by the TDT RP2.1 analog output), and is coupled to the ear with a foam ear tip. Ten separate acoustic stimuli are presented during the course of testing, and are presented at a level sufficient to elicit the acoustic reflex in most individuals. The white noise burst and pure tone pips are presented at 100 dBA, and the four gunshots presented at 110 dB Peak SPL. Probe tone clicks are presented at 90 dB SPL peak. Acoustic stimuli are calibrated at the beginning of each day in which a subject is scheduled, to ensure that each component of the stimulus presentation and recording systems are turned on, are functioning correctly, and that the stimulus levels are acceptable.

### Electromyography

Muscle activity on the face and arms is recorded by an 8 channel Bagnoli EMG system (Delsys Inc.), which includes small, lightweight, parallel bar electrodes that attach to the skin, a pre-amplifier, and amplifier/signal conditioner. The electrodes include two silver electrode contacts wired for differential operation. These electrodes do not require gel, and attach to the skin via double-sided adhesive medical tape. Electrodes are attached to the skin on the side of the body ipsilateral to the ear probe over the orbicularis oculi muscle underneath the eye, over the masseter muscle on the jaw, over the suprahyoid muscles under the chin, on the biceps brachii on the upper arm, and contralateral to the ear probe on the flexor digitorum superficialis muscle controlling the pointer finger. A reference electrode is placed on the elbow contralateral to the ear probe. The Bagnoli amplifier is comprised of analog outputs for each channel. All eight channels are routed to the analog inputs of the NI PXI-4461 card (the three unused channels serve as backups in the case of a channel failure).

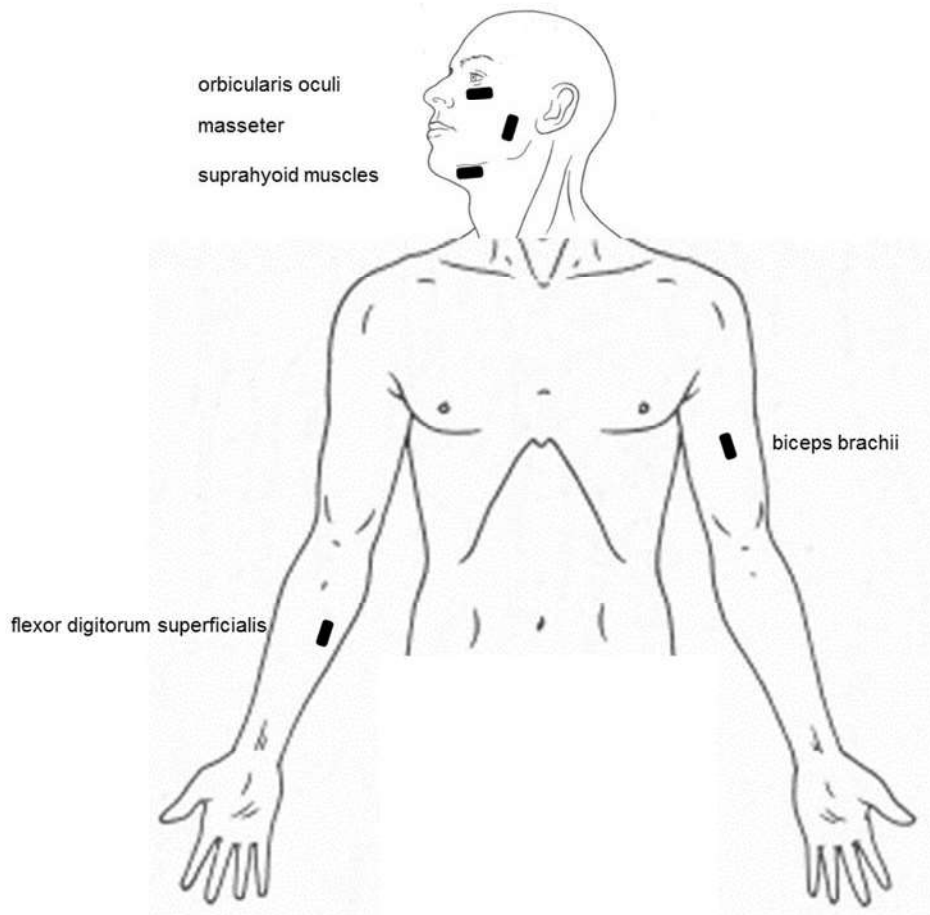


Figure 8. Locations of EMG sensors.

### Trigger pull detection

Three of the experimental conditions require the subject to pull a trigger on a toy pistol or simulation rifle. This trigger pull is detected by a 5-mm diameter force-sensitive resistor (FSR); (Interlink Electronics FSR 400) mounted onto the surface of the trigger, and driven by a devoted power supply and voltage-divider circuit. These sensors attach to the power supply via custom cables, and the voltage across the voltage divider is measured by one channel of the NI PXI-4498 card. When the trigger is pulled, force applied to the FSR, the resistance of the FSR decreases, and the voltage measured across the measurement resistor increases, as described in Figure 7.

For the simulated trigger (ST) condition, using the toy pistol, the trigger pull is saved for offline analysis. In contrast, in the Shooter (SH) and Spotter (SP) conditions, the trigger pull must be detected in real time in order to initialize the playback of a recorded gunshot (an M4 recorded in an indoor firing range). The algorithm to detect the trigger pull is implemented on a



TDT RP2.1 Real Time Processor, and functions as shown in Figure 10. The trigger FSR records a stereotyped response composed of four events. First, the subject applies light pressure to the trigger, pre-loading the FSR. Second, the subject commits to the trigger pull, applying sufficient force to overcome the weight of the trigger. Third, the trigger mechanism is actuated, the trigger quickly moved towards the rear of the rifle briefly reducing the force on the FSR, and the rifle hammer falls. Fourth, the subject releases his or her finger from the trigger and unloads the FSR. The trigger pull is detected using two aspects of the FSR output ( $V_{OUT}$ ): the first requires sufficient force on the trigger using a simple threshold ( $V_{OUT} > 3V$ ), and the second requires a sufficient rate of change by taking the difference between the current value and the value obtained two samples ago ( $V_{OUT}(n) - V_{OUT}(n-2) > 0.05$ ). A trigger pull detection occurs when these two conditions are met simultaneously (i.e., the quick downwards “trough” is detected due to the trigger actuation), and the gunshot recording is then played to the subject’s ear contralateral to the probe. The gunshot recording includes a quiet period prior to the onset, thus the gunshot arrival at the ear occurs 62.2 ms after the trigger pull is detected. This delay is substantial compared to the travel time of an actual gunshot; however, this delay is not detectable by the subject, and allows time for rejection of the artifact resulting from the trigger/hammer “click” observed in the ear canal probe.

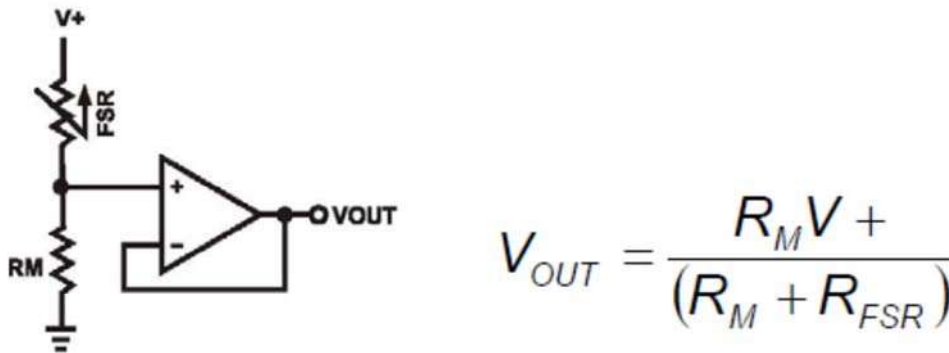


Figure 9. Voltage divider circuit (left) and equation specifying the voltage output (right). A source voltage ( $V_+$ ) is applied across the FSR ( $R_{FSR}$ ) and a measuring resistor ( $R_M$ ), and the voltage across  $R_M$  is applied to the positive input of an op-amp in a voltage follower (unity gain) configuration to obtain the output voltage ( $V_{OUT}$ ).

## MEMC Detection

### Stimuli

MEMC detection is based on the method outlined by Keefe, Fitzpatrick, et al. (2010). Briefly, a broadband click train is presented to the subject's ear via an insert earphone, and the reflected click is recorded with a probe-tube microphone integrated into the ear insert. Responses are band-pass filtered with a 4th order Butterworth filter, with cutoff frequencies corresponding to the bandwidth of the presented click. An idealized "baseline" click is estimated by taking the average of the clicks recorded for a quiet period of time preceding another stimulus, and each click during the analysis window is compared to that baseline click as the RMS amplitude of the difference between the test and baseline clicks as a function of time. Figure 11 demonstrates the method, here showing results for a 125-ms duration, 1-kHz tone pip. The elicitor tone (presented to the contralateral ear) envelope is shown at the top. The click train recorded from the microphone in the ear canal is shown in the middle, and the RMS amplitude of the difference between each of these clicks and the average baseline click (calculated in the frequency domain) is shown at the bottom. Each repetition is analyzed independently. This subject (USAARL-016) demonstrates a strong MEMC activation beginning ~50 ms (i.e., the second bin after the elicitor onset) in response to the 1-kHz tone.

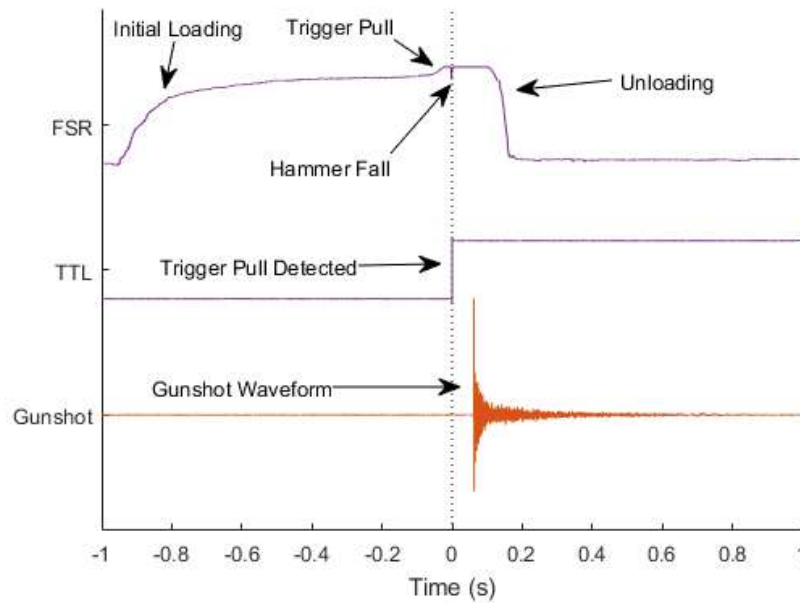


Figure 10. Method for detecting the trigger pull, and playback of a recorded gunshot.

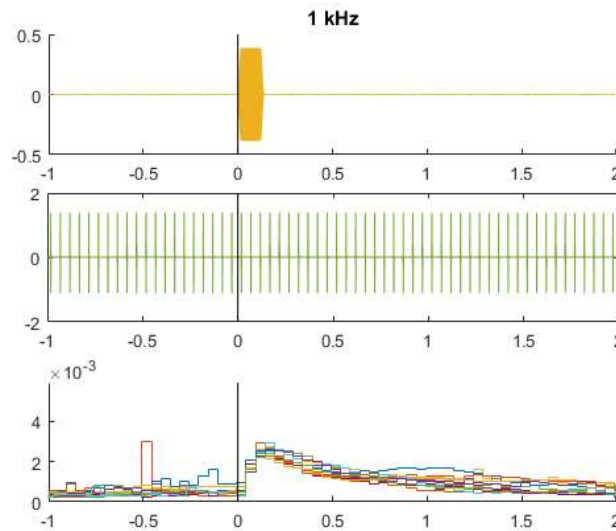
Responses to the reflexively activated stimuli (such as the 1-kHz tone shown in the example) are assessed through presenting clicks with a fixed timing relative to the elicitor onset, thus responses may be assessed at discrete times as shown in Figure 11. Other scenarios rely on

the subject's response to initiate the elicitor, thus the click timing may vary relative to the elicitor from one trial to the next. In these cases, click timing is generally uniformly distributed around the elicitor onset, thus providing a more continuous sampling of the MEMC across time.

### Response summarization

Responses may be summarized across stimulus presentations depending upon the timing with which the responses are collected. First, measures such as the mean, median, and 25th percentile response are easily assessed when the click timing is fixed relative to the elicitor onset. Second, continuously sampled responses present a greater challenge, but can be summarized in a couple ways. Continuously sampled responses may be binned into short time segments, thus converting the responses into a discrete-time form similar to the fixed-time datasets; however, this strategy necessarily discards finer-grained temporal information than the bin width, thus some subtlety in the data are lost. Another alternative is that the responses may be summarized by filtering or smoothing the response via a moving average, moving median, or moving percentile function. A moving average function involves the convolution of the responses with a weighting function (often a boxcar function), thus is equivalent to low-pass filtering the data and has the effect of smoothing the responses. The moving median (and percentile) function is comparable, but less susceptible to outliers, and was implemented to more effectively summarize data that is not normally distributed about the mean.

Both individual trial and summary data are then assessed for the presence of MEMC using various tools. Qualitatively, MEMC may be detected visually by multiple raters, who



*Figure 11.* Click-based MEMC detection method. Top: elicitor tone presented in the ear contralateral to the probe. Middle: click train presented to the ear via the probe in the ear canal. Bottom: RMS amplitude of the difference between each click and the average click recorded during the baseline period (-1.1 to -0.1 s), for each of 12 repetitions.

compare results, and the proportion of responses with a detectable MEMC counted. More quantitative means of MEMC detection include statistical measures, such as the mean exceeding a threshold based on the variance measured in the baseline segment, or similarity of the response to a prototypical reflex function. More complex analyses, such as multivariate regression, or variable reduction via tools such as principal component analysis, may prove useful as well.

### **Data collection timeline**

Data collection is separated into two recording sessions to minimize the risk of excess noise exposure, and to assess stability of each subject's hearing over time. The first visit involves an enrollment screening, and the second visit involves the primary data collection (though data is collected during both visits).

### **Enrollment visit**

The initial enrollment visit takes place in the laboratory, and obtains information about the subject's current and historical hearing health. Prior to any procedure, the subject completes an informed-consent process, including a detailed description of all procedures. The subject is then asked to provide basic contact information so that we may contact him or her during his or her participation in the study, complete a history questionnaire (documenting self-reported history of middle-ear issues, hearing loss, noise exposure, history of firearm use, or other health issues that may affect his or her participation in the study), and complete a daily questionnaire (assessment of his or her recent exposures and perceived hearing at today's visit).

The subject is then asked to sit in a double-walled sound booth where the remainder of the initial enrollment visit takes place. Video otoscopy is performed in both ears, and the output is saved. Pure tone air conduction audiometry in the initial enrollment visit is performed with the Nelson ART audiometric testing software. Additionally, a middle-ear assessment, including tympanometry, wide-band absorbance, wide-band tympanometry, reflex threshold, and reflex latency, is conducted with the Interacoustics Titan System. Finally, all equipment is then removed from the subject, and he or she is screened for cranial nerve V (trigeminal) and VII (facial) function by assessing sensitivity and symmetry of facial touch and motor function.

Results from testing completed during the initial enrollment visit are examined prior to scheduling a follow-up visit to determine if the subject is eligible to continue participation in the study according to inclusion and exclusion criteria. Subjects meeting the necessary criteria are scheduled for the data collection visit(s). Subjects not meeting the necessary criteria are dismissed from the study.

### **Data Collection visit**

The data collection visit takes place as soon as possible following the enrollment visit, and consists of three sections: a hearing pre-test, the data collection consisting of several tasks, and a hearing post-test.

First, the subject is seated in the audiometric sound booth, his or her identity is verified, and he or she is asked to complete the daily questionnaire. Next, an abbreviated hearing test is administered. An otoscopic examination using a video otoscope is completed to confirm the ear canals are free from cerumen or other conditions that would affect the fit of the MEMC probe. Pure tone air conduction thresholds are determined with the Nelson ART software, and an abbreviated assessment of middle-ear function is completed with the Interacoustics Titan System.

Second, the subject is moved to the data collection site and seated in a barber's chair. EMG electrodes (Bagnoli 8-channel desktop EMG system) are attached to the skin over muscle groups on the face and upper extremities to measure muscle activity related to facial nerve, trigeminal nerve, and hand/arm activity. An acoustic probe (the Etymotic ER-10X probe system) is inserted into the ear (usually left) contralateral to the subject's dominant (shooting) hand to allow assessment of middle ear muscle activity. Experimental conditions include presentation of several stimuli aimed at eliciting both reflexive and conditioned MEMCs responses. Reflexive MEMCs are assessed in response to acoustic stimuli by presenting sounds to the ear contralateral to the acoustic probe (usually right) with an Etymotic ER-4PT insert earphone, to tactile stimuli by administering puffs of air to the face, and to muscle activity by having the subject close their eye ipsilateral to the acoustic probe in response to a video prompt. Conditioned MEMCs are assessed by having the subject (who is an experienced shooter) pull the trigger of a toy pistol (a bright orange cap gun with the hammer muffled with felt) when prompted by a video, as well as by performing a shooting task (where the subject initiates the playback of a recorded gunshot by pulling the trigger of a simulation rifle), and during a spotter task (where the experimenter initiates the playback of the gunshot by firing the simulation rifle).

Following these measurements, all study instruments are removed from the subject and a post-test of otoscopy, pure-tone thresholds, and the abbreviated middle-ear battery is completed. Finally, the subject is debriefed to determine whether any unexpected events occurred during the course of the study, reimbursement is arranged, and the subject is released from the study.

### **Data Collection Procedures and Preliminary Results**

The data collection procedures described above produce a large variety of datasets that must be analyzed and interpreted. Here, we describe examples of the initial review of each data type to demonstrate the types of results and conclusions, as well as limitations, able to be drawn from this dataset.

#### **Questionnaires**

Upon entry into the study, subjects answer a series of entry and history questions aimed at assessing basic demographic information, as well as hearing health history. Demographic information collected includes highest level of education, current employment, and whether the subject is currently or has ever been employed by the military or a law-enforcement agency.

Similarly, information is collected to determine whether the subject is either an occupational or recreational firearm user, as an experienced firearm user may be more likely to show a conditioned MEMC to the gunshot conditions tested in this study. Additionally, questions are asked to aim at assessing whether the subject has a substantial amount of experience with loud sounds and hearing protection. We assess the subject's self-perception of his or her hearing, asking questions assessing whether the subject has a history of ear infections and tympanostomy tube placement, whether the subject reports tinnitus, as well as a qualitative assessment of his or her own hearing ability. Similarly, questions also assess whether the subject has a history of concussion, dizziness, or facial paralysis (e.g., Bell's palsy), as the potentially affected nerve innervate both the face and middle ear muscles.

A second questionnaire, completed at the beginning of both visits, assesses more immediate exposures that might affect the subject's hearing. These questions assess qualitative changes in the subject's hearing or tinnitus, dizziness, or pain in the ears. Additional questions reveal recent exposures that might affect hearing status including exposure to loud environments, listening to music over headphones, tobacco use (e.g., (Cruickshanks et al., 1998)), and use of hearing protection.

### **Video Otoscopy**

Next, subjects' ears are inspected via video otoscopy to rule out any physical conditions that might prevent inclusion in the study. Conditions that might prevent the subject from participating in the study include ear canal size that is inappropriate for use with the measurement probes, excessive cerumen, birth defects, personal adornments, or prior ear surgery. Other conditions that may be revealed leading to a volunteer's exclusion from the study include impacted cerumen, fluid discharge, bony growths, inflammation or redness, perforated tympanic membrane, foreign objects, enlarged canal, or excessive hair growth. If any of these conditions are evident, the subject is referred to his or her healthcare provider for intervention. Output of the video otoscope was saved offline for review offline at a later date.

### **Audiometric Assessment**

Pure tone air conduction audiometry is performed with the Nelson Acoustics ART system. Only subjects with excellent hearing are enrolled in this study: subjects must have hearing thresholds of not more than 10 dB for 125, 250, 500, and 1000 Hz, and not more than 20 dB for 2, 4, and 8 kHz. These criteria are stricter than the H-1 hearing profile defined in the Army Standards of Medical Fitness (Army Regulation (AR) 40-501) as: "Audiometer average level for each ear not more than 25 dB at 500, 1000, 2000 Hz with no individual level greater than 30 dB. Not over 45 dB at 4000 Hz." The reason behind the strict criteria was that this selected subjects with the least possible amount of hearing loss, which could affect MEMC detection.

## **Cranial Nerve Assessment**

The middle ear muscles are innervated by the trigeminal (CN V) and facial (CN VII) cranial nerves, which also innervate areas of sensation and muscle movement in the face and neck. This assessment of touch and muscle activity is intended to assess weakness or asymmetry in these responses to identify nerve dysfunction, such as during Bell's palsy. Since the nerves that innervate the middle-ear muscles branch off the facial cranial nerve, any dysfunction detected during this assessment would indicate a potential problem with detecting a MEMC.

## **Tympanometry**

Mobility of the eardrum and middle ear, as well as presence of the acoustic reflex, is assessed with a clinical tympanometer (the Titan system). Five conditions are assessed in each ear of each subject: pure tone tympanometry, wide-band tympanometry, wide-band absorbance, ipsilateral and contralateral acoustic reflex thresholds, and ipsilateral acoustic reflex decay. Tympanograms are assessed for clear peak centered on ambient pressure. Subjects showing tympanograms with a low peak or a peak shifted to higher or lower pressures are either asked to return for a retest or are referred to their personal healthcare provider. Similarly, wide-band tympanometry and wide-band absorbance are assessed for abnormal responses. Such failures may result in dismissal from the study.

Acoustic reflexes are assessed with a 226 Hz probe tone, and elicitors presented at 500, 1000, 2000, and 4000 Hz, at 80, 85, 90, 95, and 100 dBA. Threshold is assessed for each elicitor frequency, and are identified by the Titan software if the acoustic impedance exceeds 0.2 mhos. Subjects are assessed for identifiable thresholds < 100 dBA both ipsilateral and contralateral, in both ears. Finally, acoustical reflex decay is measured for a 1 kHz tone elicitor presented 5 dB above the threshold identified by the Titan software (or at 95 dBA), and assessed for a consistent response throughout the 10-s duration of the elicitor presentation.

## **MEMC Probe Frequency Sweep**

The MEMC probe (Etymotic ER-10X) microphone is calibrated with a microphone calibrator, and the speaker driver calibrated with a calibrated 1/2-in. microphone, prior to data collection each day. The seal of the probe in the subject's ear, and the resulting frequency response of the probe and ear system, are measured several times throughout the course of the data collection session (generally before and after each measurement) to ensure a stable set of measurements. The probe check is performed using a frequency sweep. A smooth response at low frequencies indicates a good seal in the subject's ear, and a deviation from this smooth response indicates a failing/slipping seal. When a noisy low-frequency response is observed, the rubber or foam eartip and plastic probe tube are re-inserted and/or replaced.



## EMG Control Tasks

EMG electrodes are attached to five muscle groups on the face, neck, and upper extremities, in order to allow correlation between activity of these muscle groups and middle ear muscle activity. Each recording site is first cleaned with a disposable wipe soaked in rubbing alcohol, and the EMG electrode affixed with double-sided medical-grade adhesive tape. The signal quality of each EMG recording electrode is assessed during control tasks, during which subjects are asked (via a set of recorded instructions), to complete a task aimed at activating each muscle in isolation. Signals are recorded from all sensors simultaneously so that MEMC may be assessed during these tasks, and periods of the recording in which multiple muscles are activated may be excluded. Sensor activity was saved for further analysis.

*Table 1. EMG Sensor Locations*

Muscle groups:	Orbicularis Oculi	Masseter	Suprahyoid muscle group	Biceps Brachii	Flexor Digitorum Profundus
Muscle purpose	Closes the eyelid	Mastication	Widening the esophagus & swallowing	Flex the upper arm	Flex the pointer finger
Location	Below the eye	On the jaw	Under the chin	Upper arm	Forearm
Task	Close the eye	Chew a mint	Press tongue to roof of the mouth	Flex arm against a fixed handle	Pull the trigger of a toy pistol

## Reflexive MEMC Recordings

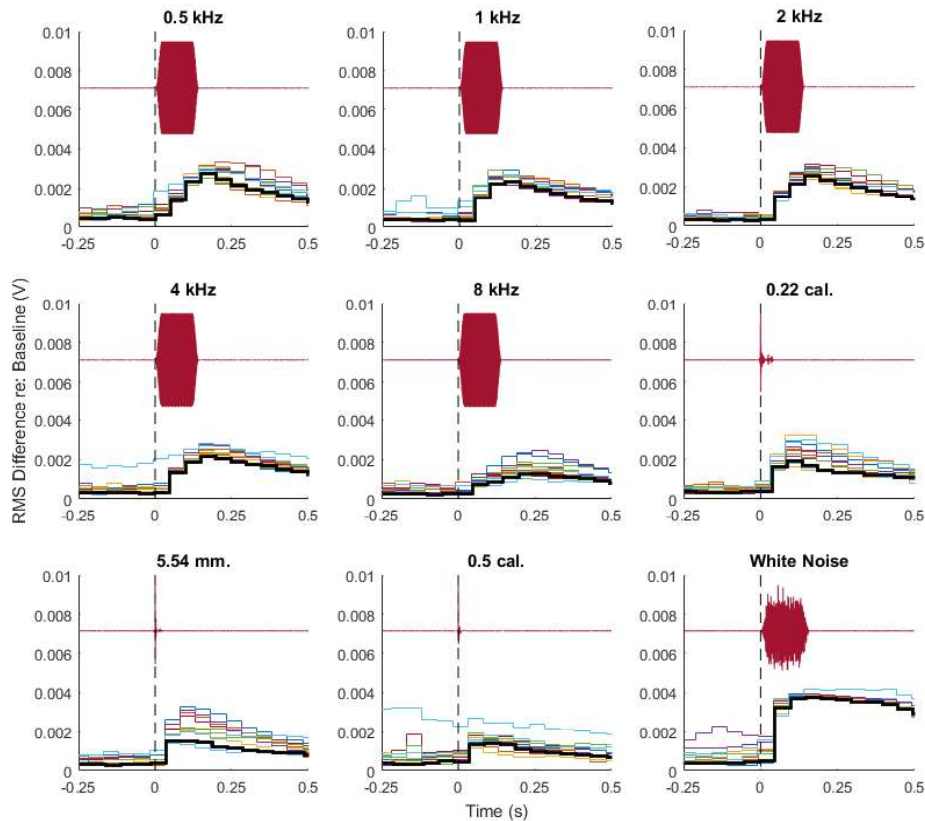
### Acoustic Elicitors

Contralateral acoustically elicited MEMCs are assessed to a number of sustained and impulsive sounds. Sustained sounds include short (500 ms) tone pips (500 Hz, 1 kHz, 2 kHz, 4 kHz, and 8 kHz) and a short (500 ms) white noise burst, whereas the impulse noises consisted of three recorded gunshots (0.22 cal, 5.56 mm, and 0.50 cal). Sounds are calibrated prior to data collection each day with a calibrated B&K microphone, such that the continuous sounds are 100 dBA, and the impulsive sounds are 110 dB Peak.

Responses were recorded for 12 repetitions of each stimulus. The probe clicks presented to the test ear were synchronized with the start of each elicitor presentation to assess reproducibility of the response over those 12 repetitions. The MEMC responses are thus quantified into 50 ms bins, and accordingly are plotted in Figure 12 as a staircase plot for one exceptional subject. Each subplot shows the change (in V RMS) relative to the average baseline click (calculated in the frequency domain) as a function of time relative to the start of the elicitor. The RMS difference for each elicitor presentation is represented by a colored line, the 25<sup>th</sup> percentile response

(i.e., 25% of responses are below this line) is represented by a heavy black line. The onset of the elicitor is indicated with a vertical dashed line, and the envelope of the elicitor is indicated at the top of each subplot (Note, the envelope of the sound is narrow, and thus difficult to discriminate from the vertical dashed line in some gunshot conditions).

One subject (USAARL-016) showed a robust MEMC engagement following presentation of all nine stimulus conditions (Fig. 12). In each condition, the deflection noted in the RMS difference following the elicitor, indicating an MEMC, is first observable in the *second* bin, suggesting the latency of the MEMC is between 50-100 ms. Similarly, the MEMC disengages shortly after the offset of the elicitor sound, beginning to decrease within 1-2 bins (50-150 ms) of the elicitor offset, and decaying exponentially back towards baseline. An exception to this trend is noted in the white noise burst, where the MEMC remains engaged for some hundreds of milliseconds after the elicitor offset; however, the MEMC decay has begun by the end of the window shown, and decays back to baseline by  $t = 1$  s thereafter (similar to the other stimuli; not shown). Note, the responses recorded from this subject are exceptionally clear and prevalent; most responses collected thus far show a strong response to a subset of these acoustic elicitors,



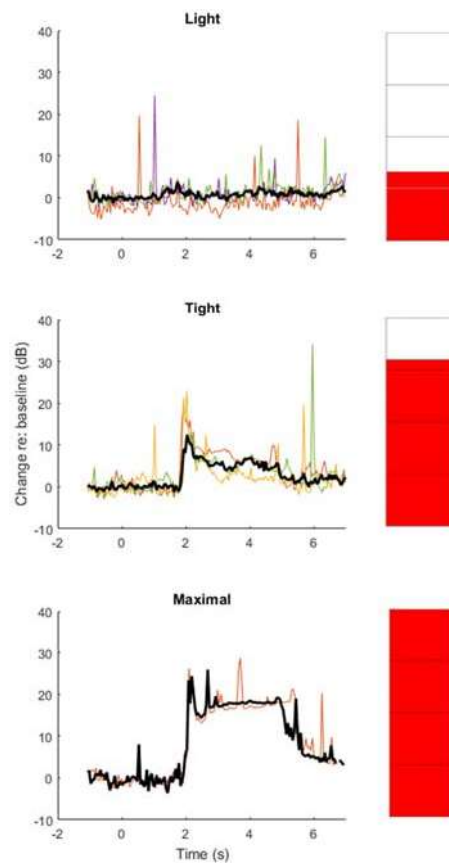
*Figure 12.* MEMCs elicited by the acoustic elicitors for one individual (USAARL-016). RMS difference relative to baseline is shown for twelve repetitions of each acoustic elicitor (five tones, three recorded gunshots, and a white noise burst), as well as the 25<sup>th</sup> percentile response (heavy black line). Time is shown relative to elicitor onset.

and some subjects do not show a clear MEMC in response to any of these elicitors (despite showing acoustic reflexes during the clinical reflex testing with the Titan system).

### Non-Acoustic Elicitor Recordings

#### Eye-close

The correlation between facial muscle activity and MEMC was assessed by having the subject close the eye ipsilateral to the ear probe in response to a visual cue. Eye close activity was assessed at three levels: light, tight, and maximal. The time, duration, and level of each eye close was indicated by presenting a video of an illustrated “thermometer” on an LCD display, where a red bar of varying height (which indicates the desired eye close strength) appears on screen for the requested eye close duration. The thermometer appeared on screen throughout the duration of the experiment, and each eye close request appeared on screen for three seconds at a



*Figure 13.* Example eye close results, along with example effort meters. Colored lines indicate individual eye closes, heavy black lines the 25th percentile response. Responses (RMS change relative to baseline, in dB re: baseline) as shown as a function of time re: effort request onset.

time. Each session began and ended with a maximal effort request, with lower effort requests in between.

Responses were recorded to multiple presentations of each requested effort level, and assessed relative to the baseline, no-effort, period preceding each eye close. A set of eye close measurements is shown in Figure 13 for one subject (USAARL-008), along with the corresponding effort meter to the right of each plot. Responses to the light, tight, and maximal eye closes are shown in the top, center, and bottom plots. Each colored line indicates the RMS change relative to baseline (in dB re: baseline) for one trial (i.e., one eye close), as a function of time relative to the start of the eye close request. The heavy black lines represents the 25th percentile response across all repetitions presented for each condition. In general, responses quickly rise, after some onset latency, to a peak, remain at a plateau level for the duration of the eye close, and return to baseline after the eye is opened. The peak and plateau levels scale with the requested eye close effort.

### Air-Puff

The correlation between tactile sensation on the face and MEMC was assessed by applying light puffs of air to the face around the nose and eyes. Puffs of compressed air (medical-grade nitrogen), presented via computer-controlled, electrically activated air valves, were applied bilaterally to the temples (posterior to each eye) and the alar junction, the most lateral aspect of the alar sidewall (i.e., the exterior of the nasium). Air was directed to each location with flexible PVC tubing, inserted through a modular hose system (Loc-Line Products

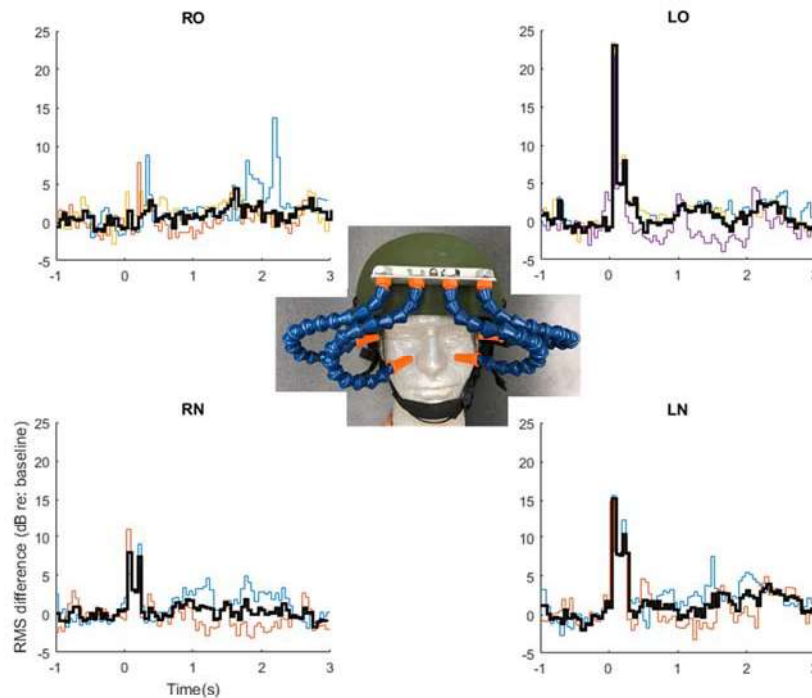


Figure 14. Air-puff locations and example responses from one subject.

Inc., Oswego, OR) that was rigidly attached to a large combat helmet. Air puffs were 200 ms in duration and applied to each location in a random order, approximately three times each during the 60-s recording period.

Responses were recorded to multiple presentations of each air puff position, and assessed relative to the baseline period preceding each presentation. A set of measurements for one subject (USAARL-013) is shown in Figure 14, arranged according to the corresponding position to which the airpuff was applied. Each colored line indicates the RMS change relative to baseline (in dB re: baseline) for one trial (i.e., one air puff), as a function of time relative to the start of the puff. The heavy black line represents the 25th percentile response across all repetitions presented for each condition. In general, responses show a fast, short duration excursion relative to baseline, suggesting a fast activating, fast inactivating MEMC. Note, the air-puff elicits a substantial startle reaction, involving involuntary contractions of head and neck muscles, in many subjects, thus MEMC activity correlated with the air puff stimulus may be the result of correlated muscle activity, rather than correlation with the tactile stimulus itself. Further analysis is necessary to clarify the contributions of these effects.

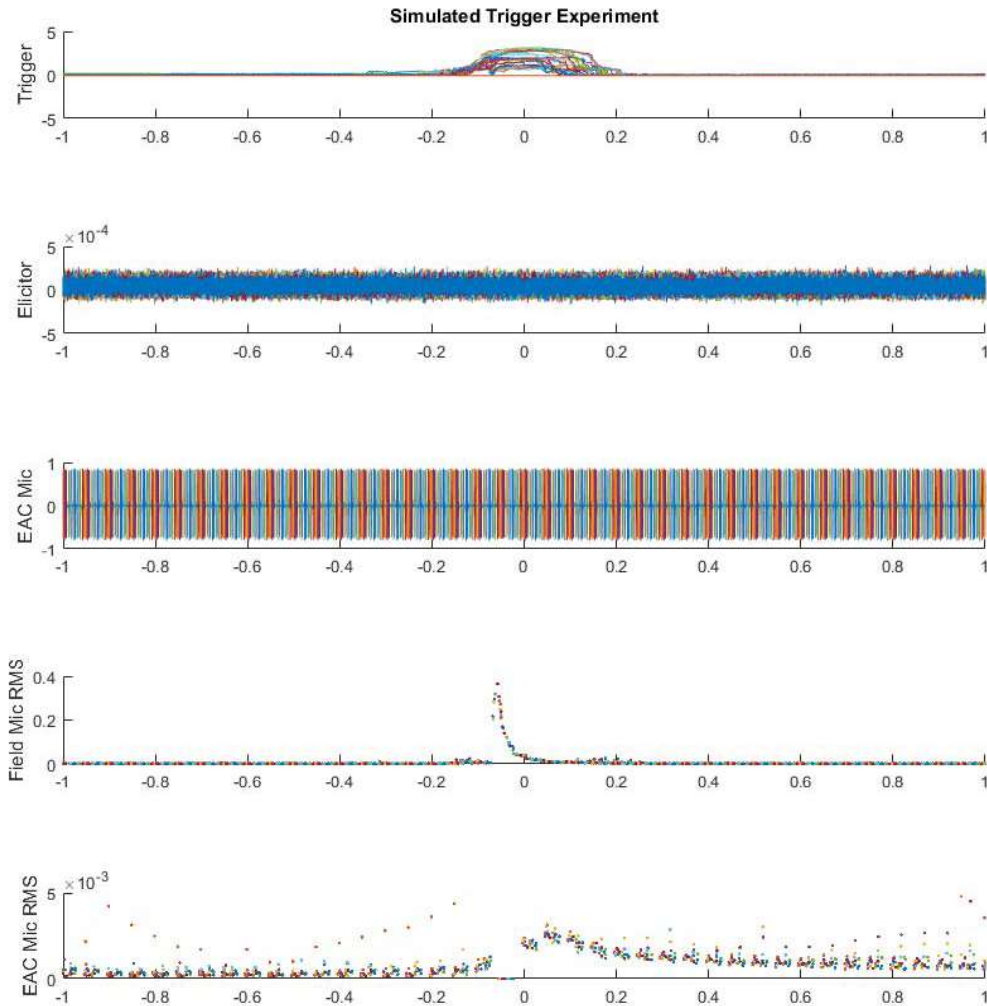
### **Conditioned MEMC Tasks**

Three conditions were presented to each subject, intended to isolate different components of the action of firing a weapon. First, MEMC activity correlated with flexing the pointer-finger was assessed while the subject pulled the trigger of a toy pistol in a Simulated Trigger (ST) task. Second, the reflexive activation of the MEMC in response to the playback of a recorded gunshot is measured in a Spotter (SP) task, where the subject observes the experimenter fire the rifle simulator. Third, anticipatory activation of the MEMC was assessed in a Shooter (SH) task, which is identical to the SP task, except the subject is pulling the trigger on the rifle simulator.

#### **Simulated Trigger**

The ST task assesses the relationship between the motor control and execution involved with pulling a trigger, and MEMC using a toy cap gun modified to accept a force sensitive resistor (FSR) on front face of the trigger. The timing and rate of the trigger pulls are controlled by engaging the subject in a distractor task: the subject is instructed to point the pistol at a colored target displayed on a video monitor, which moves around the screen in an unpredictable pattern, and pull the trigger when the target changes color from red to blue. The target changes color approximately every ten seconds ( $\pm 5$  seconds), cueing the subject to fire the pistol 20-30 times over the course of the 250-s recording. Subjects are instructed to support the pistol in a comfortable fashion, and to be consistent throughout the duration of the recording. Most subjects supported the pistol with both arms, as would be expected when firing a live pistol. The hammer of the toy pistol was padded with a small piece of felt, but nevertheless made an audible click when fired. The pistol uses a double-action firing mechanism, thus the pistol does not have to be re-cocked between each trigger pull.

Responses are segmented based upon two features visible in the recordings. First, the signal from the FSR shows a reliable pattern across subjects due to the mechanics of the trigger mechanism: the force on the trigger increases to approximately 2/3 the maximum as the subject prepares to fire; as the trigger pull is executed the force rises to a maximum; as the trigger



*Figure 15.* Example of an ST test condition in one subject. The x ordinate is Time (s). In each plot, color represents the time-course of a single trigger pull (i.e., a single elicitor presentation). The response of the FSR mounted on the rifle’s trigger is shown at the top, the acoustic elicitor (i.e., no signal) is shown in the second row, the click train presented to the probe ear is shown in the third row, and the RMS amplitude measured during each analysis window (around each click presentation), is shown for the field microphone, and the ear canal (EAC) microphone, in the fourth and fifth rows. Note, the amplitude in the EAC microphone has been scaled down using field microphone RMS amplitude to reduce the effects of the acoustic click artifact.

mechanism engages, the force briefly drops; once the trigger pull has completed, the force returns to zero as the subject removes his or her finger. The brief drop in the force as the trigger mechanism engages has proven to be a reliable indicator of the trigger pull that is easily identifiable by searching for a sharp decrease occurring during a period of high force. Second, although the hammer of the pistol is padded with felt, an audible click is generated, and this click is readily identifiable in the free-field microphone recording. These two techniques (i.e., the FSR signal and the free-field microphone signal) thus allow cross-validation of the trigger pull, and a backup mechanism should one fail.

Although the timing of the target is controlled, the trigger pull itself is controlled by the subject, thus the relationship between the trigger pull, and the probe click stimulus timing, varies during the duration of the recording. For this reason, the RMS change relative to baseline (i.e., the period preceding each trigger pull) are shown as points, rather than steps, to indicate the precise timing of each click presentation relative to the trigger pull. In general, responses reveal little or no MEMC activation in response to the trigger pull across the subjects tested thus far.

### **Shooter/Spotter Condition**

The SH and SP conditions are similar in setup to the ST condition and identical to one another except for the identity of the individual eliciting the stimulus. In these conditions the shooter pulls the trigger of a rifle simulator (an M4 Carbine modified for use in the EST 2000 rifle simulator system; note the EST 2000 system was *not* used during these measurements). The rifle is an actual M4 Carbine that has been modified such that it cannot accept, and will not fire ammunition, but the firing mechanism remains intact and functional otherwise. We outfit the trigger with an FSR in a similar manner as the toy pistol described previously to record the shooter's trigger pull. The hammer fall resulting from the trigger pull generates an audible click, which is once again recorded via the free-field microphone. Since the EST 2000 full simulator system is not used here, the rifle does not operate in a semi-automatic fashion and must be re-charged in between each trigger pull, which is done by the experimenter in both the SH and SP conditions.

Two major changes differentiate the SH and SP conditions from the ST condition, besides the firearm used. First, the rifle is supported on a rifle stand, on a table, and aims at paper targets mounted at the distant end of the room (these measurements take place in the air rifle range in the basement of the USAARL acoustics building), rather than aiming the rifle at the distractors on the video monitor. Second, in the SH and SP conditions, the trigger FSR is continuously monitored by custom software operating on a Tucker-Davis Technologies RP2.1 real-time processor, and a recorded gunshot played back to the subject's right ear (contralateral to the acoustic probe) when the trigger pull is detected. These conditions thus aim to test whether pairing the trigger pull (in as realistic a fashion as is possible in a laboratory setting) with an MEMC-eliciting acoustic stimulus, affects the timing of the elicited MEMC.

The acoustic stimulus played back after each trigger pull is a recording of an M4 Carbine fired in an indoor range, thus the gunshot is reasonably similar to the sound one might predict



from firing the rifle in the test room (i.e., the USAARL air rifle range). The gunshot is played back at 110 dB SPL peak and is sufficiently loud enough to elicit an MEMC in most individuals tested. The onset peak of the gunshot arrives ~62 ms after the trigger pull, fast enough that no delay is detectable by the subject. In the SH condition, the subject is instructed to fire the rifle repeatedly, allowing a short pause (2-3 s) after the experimenter recharges the rifle. The experimenter allows a short pause after the trigger pull, thus the period from one trigger pull to

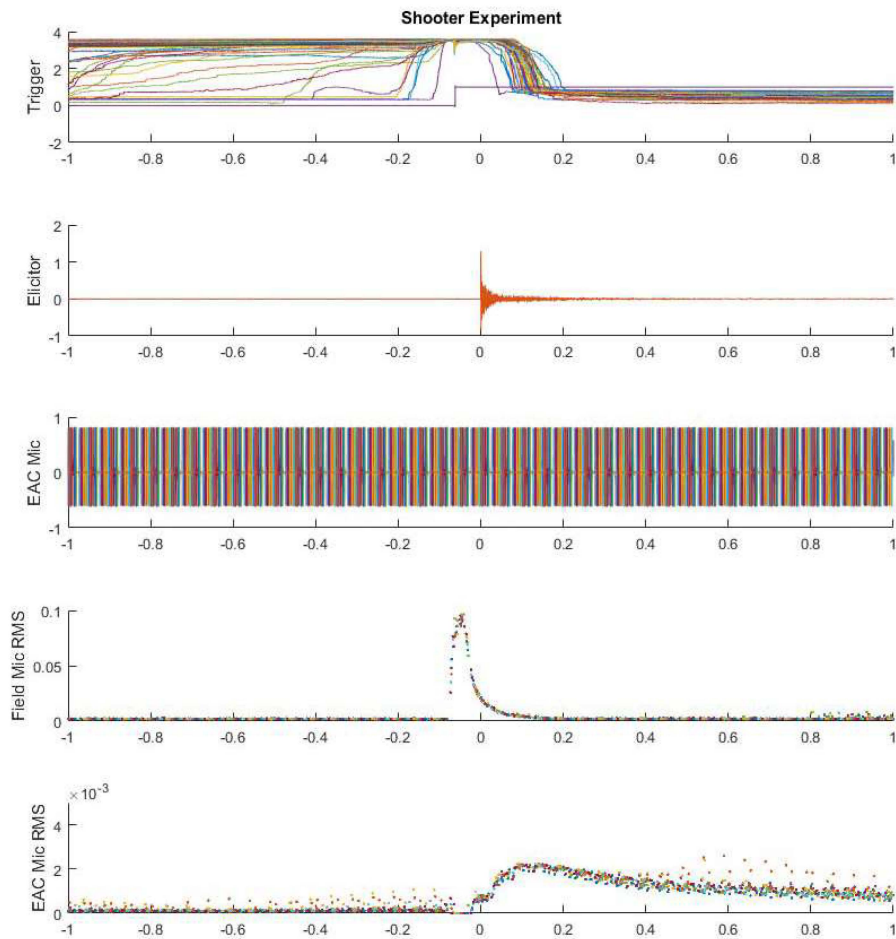
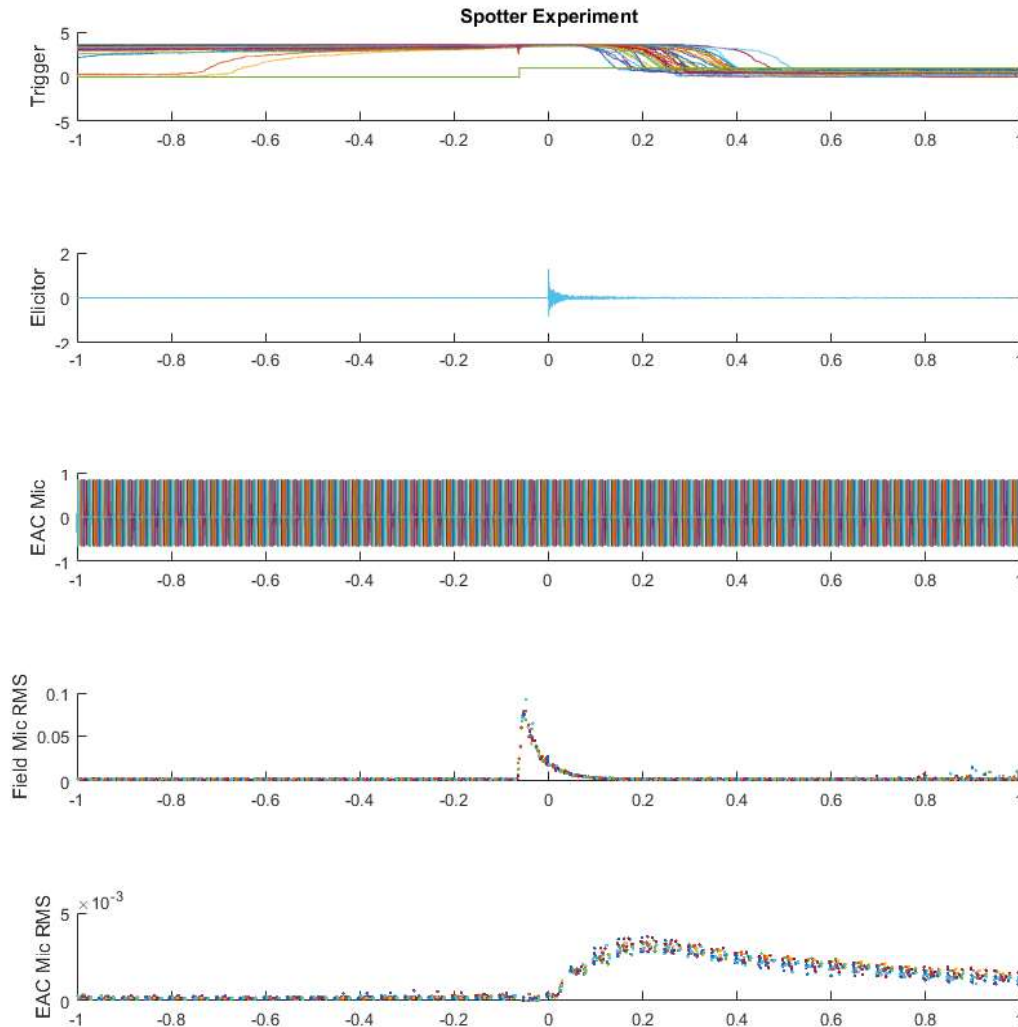


Figure 16. Example of an SH test condition in one subject. The x ordinate is Time (s). Results are shown in the same manner as the ST condition in Figure 15. In each plot, color represents the time-course of a single trigger pull (i.e., a single elicitor presentation). The response of the FSR mounted on the rifle’s trigger is shown at the top, the acoustic elicitor (i.e., the gunshot playback) is shown in the second row, the click train presented to the probe ear is shown in the third row, and the RMS amplitude measured during each analysis window (around each click presentation), is shown for the field microphone, and the ear canal (EAC) microphone, in the fourth and fifth rows. Note, the amplitude in the EAC microphone has been scaled down using field microphone RMS amplitude to reduce the effects of the acoustic click artifact.

the next varied between approximately 6-12 s. Similarly, in the SP condition, the experimenter fired and recharged the rifle, allowing a short delay between each action. Approximately 20-30 shots were fired in each condition, similar to the ST condition.



*Figure 17.* Example of an SP test condition in one subject. The x ordinate is Time (s). Results are shown in the same manner as the ST condition in Figure 15. In each plot, color represents the time-course of a single trigger pull (i.e., a single elicitor presentation). The response of the FSR mounted on the rifle’s trigger is shown at the top, the acoustic elicitor (i.e., the gunshot playback) is shown in the second row, the click train presented to the probe ear is shown in the third row, and the RMS amplitude measured during each analysis window (around each click presentation), is shown for the field microphone, and the ear canal (EAC) microphone, in the fourth and fifth rows. Note, the amplitude in the EAC microphone has been scaled down using field microphone RMS amplitude to reduce the effects of the acoustic click artifact.

Finally, responses are compared between the SH and SP conditions by calculating the moving average of the EAC RMS amplitude across time with a 40-point moving average filter and plotting the resulting smoothed functions together as a function of time. These responses are shown as RMS amplitude change relative to baseline and normalized to the maximum, in Figure 18. Assessment of these responses in this way will allow comparisons between both the timing (e.g., onset time, rise time, time to peak, etc.) and magnitude of the MEMCs in each condition. In particular, responses will be scrutinized for evidence of MEMC prior to the acoustic elicitor ( $t = 0$  s), and prior to the onset of the ‘click’ artifact from the trigger mechanism ( $t = -0.062$  s).

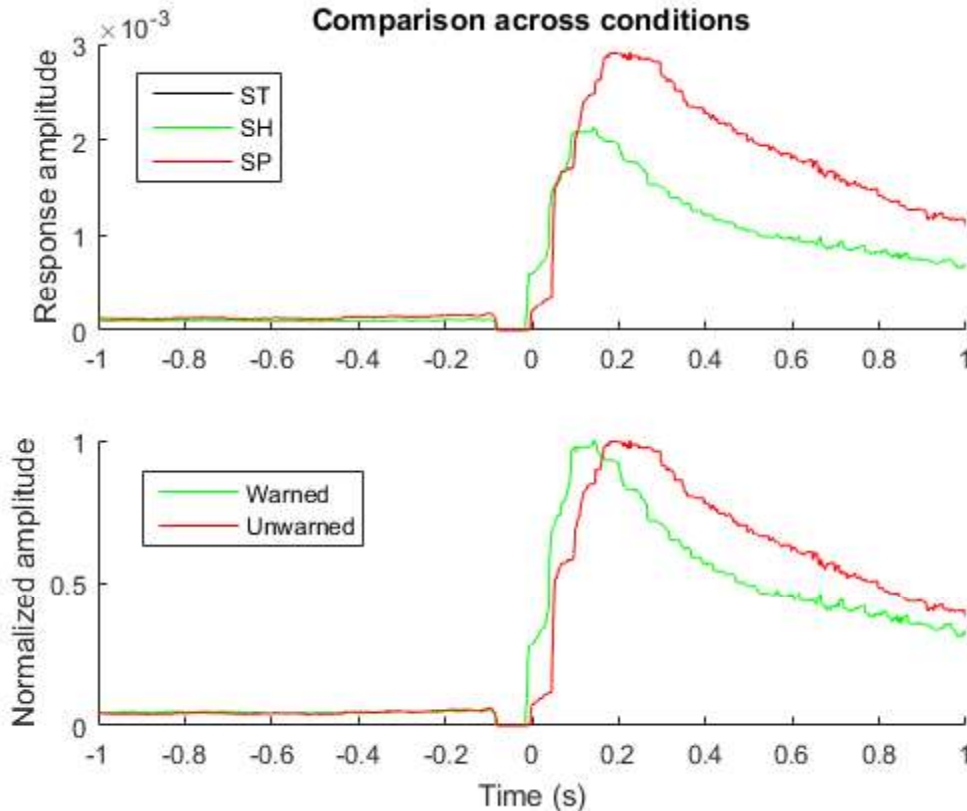


Figure 18. Comparison across SH and SP conditions.

## Conclusions

The goal of the current technical report is to describe a method using wideband acoustic reflectance developed to measure MEMCs elicited by both acoustic and non-acoustic stimuli. Here we detail the procedures and resulting data collected demonstrating the detection of MEMCs for various stimuli. These techniques reported here will be used in future studies at USAARL, which will monitor MEMCs occurring during live fire of small arms at a rifle range on Fort Rucker.

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