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Middle Ear Muscle Contractions in Response to Non-Acoustic Stimuli: The Role of Voluntary Motor Activity

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14. ABSTRACT

Middle ear muscle contractions (MEMCs) are most commonly considered a response to high-level acoustic stimuli. However, MEMCs have also been observed in the absence of sound, either as a response to somatosensory stimulation or in concert with other motor activity. The relationship between MEMCs and non-acoustic sources is not well understood. This study examined the occurrence rate of MEMCs associated with different levels of voluntary eye closure effort while controlling for demographic and clinical factors in a large group of participants (N=190) with the presence of acoustic reflexes and excellent hearing sensitivity. MEMCs were detected via changes in total energy reflected in the ear using a filtered (0.2 to 8 kHz) click train. Participants were instructed to voluntarily close the eye ipsilateral to the MEMC detection probe at three levels of effort. Muscle activity associated with eye closure was measured using surface electromyography. Results revealed that MEMCs commonly occur in association with voluntary eye closure and that the likelihood of observation increases with eye closure effort level and associated orbicularis oculi muscle activity. MEMC occurrence rates for eye closure were higher than for high level brief acoustic stimuli in the same participant pool, suggesting that motor activity may be a more robust elicitor of MEMCs than acoustic stimuli.

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Middle ear muscle contraction (MEMC), Non-acoustic elicitor, Auditory hazard, Damage risk criteria

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Thus, motor activity can serve as a confounding factor for auditory exposure studies. Finally, these results can complicate the interpretation of impulsive noise damage risk criteria that assume MEMC's serve a consistent, uniform, protective function.

Summary

Background

Middle ear muscle contractions (MEMCs), which are activations of the stapedius and/or tensor tympani muscles, can be elicited by a range of acoustic and non-acoustic stimuli. MEMCs can increase the impedance of the middle ear system, reducing the efficiency of sound energy transmission to the cochlea. As a result, some damage risk criteria (DRC), which estimate hearing damage from noise exposure, have assumed a protective role for MEMCs. Inclusion in DRC implies that MEMCs are pervasive within the exposed population, exhibit magnitudes sufficient to provide protection, and presence and magnitude should not be substantially altered by other variables. Studies characterizing MEMCs have largely focused on responses to acoustic stimuli for auditory diagnosis (i.e. the acoustic reflex), while studies examining the role of non-acoustic elicitors, such as motor activity, are few. DRC are implemented in operational environments that place a range of situational and behavioral demands on the warfighter. These activities will engage motor systems that may influence the presence and magnitude of MEMCs, and therefore the warfighter's noise exposure. The overall goal of this project was to examine relations between voluntary facial muscle activity and MEMCs.

Purpose

The purpose of this study was to (1) examine the occurrence rates of MEMCs in response to voluntary eye closure; (2) assess if eye closure effort influences occurrence rates; and (3) assess the association between MEMC occurrence rates and demographic and clinical factors in a large participant group with excellent hearing sensitivity and normal middle ear function.

Methods

A group of 190 participants performed unilateral voluntary eye closure at different effort levels while the status of the ipsilateral ear was monitored using an otoacoustic emissions (OAE) probe. Eye closure was quantified using electromyographic (EMG) recordings of the orbicularis oculi muscle. A broad band click was delivered into the ear canal by the OAE probe at 50 ms intervals. Eye closure related changes in root-mean-square amplitude of the clicks relative to an average baseline click indicated the presence of middle ear muscle activity. A panel of judges independently identified MEMCs from a 25th percentile trace of the distribution of replicates for each participant at each eye closure effort level. Occurrence rates were estimated from these judgements and logistic regression was used to determine how experimental and participant factors influenced the likelihood of observing MEMCs.

Conclusions

MEMCs were commonly observed during EMG-verified eye closure gestures. Voluntary eye EMG activity was a potent elicitor of MEMCs. MEMC occurrence rates at even moderate levels of eye muscle activity are much greater than are observed for impulse-like acoustic stimuli and MEMCs were very common at maximum levels of eye muscle activity. In conclusion, eye-related motor activity is a powerful elicitor of MEMCs that can serve as a confounding factor for auditory exposure studies. These results also complicate the interpretation of impulsive noise DRC that assume MEMCs serve as a consistent, uniform protective factor.

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Introduction

For more than half a century, middle ear muscle contractions (MEMCs) have been considered a protective mechanism in impulsive noise damage risk criteria (DRCs) in military populations (Price, 2007a, 2007b; Price & Kalb, 1991; Ward et al., 1968). DRCs estimate the level and probability of hearing damage associated with exposure to impulsive noise. The goal is to use such estimations to prevent injury in the intended population. If MEMCs are to be included in DRCs, they must be pervasive (i.e., present in 95% of people with 95% confidence) within the exposed population, their magnitude should be sufficient to provide protection, and their presence and magnitude should not be altered by other variables. Military DRCs are designed to be implemented in operational environments where warfighters are engaged in a wide range of behavioral activities including operating firearms, maintaining situational awareness, and communicating with team members. These activities will engage motor systems that may have significant influence on middle ear muscle activity. For example, in the field it might be common for a warfighter to tightly close their eyes when discharging a fixed weapon (e.g., mortar). On the other hand, a warfighter who is shooting a service rifle is explicitly instructed to maintain a relaxed body position with eyes open at the moment the weapon is discharged (U.S. Department of the Army, 2011, 2012). Both scenarios involve high-level noise, and it is unclear how the influence of non-acoustic factors, such as eye closure, would impact the warfighter's sound exposure.

The middle ear space contains two muscles: the stapedius and tensor tympani. Stapedius, the smaller of the two, originates from the pyramidal eminence, inserts onto the neck of the stapes and is innervated by the facial nerve (cranial nerve VII). Tensor tympani originates from the Eustachian tube and sphenoid bone, inserts onto the neck of the malleus and is innervated by the trigeminal nerve (cranial nerve V). Even though the innervation and lines of action of these two muscles are quite different, both have the potential to alter the transmission of sound energy through the middle ear. Specifically, MEMCs can alter middle ear impedance. The impedance changes lead to attenuation in the low frequencies (Pang & Peake, 1986), particularly below 1 kHz (Rabinowitz, 1977). Little to no reduction is observed for higher frequencies. The increased ossicular chain stiffness caused by MEMCs can result in a small increase in high-frequency energy transmission to the cochlea (Feeney & Keefe, 1999; Jones, Greene, & Ahroon, 2017). The biological purpose of MEMCs is not clear. Posited roles for MEMCs include enhancing ventilation of the middle ear space (Mukerji, Windsor, & Lee, 2010), reducing the spread of masking by low frequency sound produced during vocalization (Borg & Zakrisson, 1975) and serving as a protective mechanism against potentially damaging exogenous sound sources (Borg, Counter, & Rosler, 1984). However, given the substantial evolutionary remodeling of the middle ear and nearby structures, the middle ear muscle system may have provided selective advantages unrelated to hearing, such as mandibular stabilization, or the system may be an evolutionary vestigial that provides no clear selective advantage or biological role (Manley, 2010).

MEMCs can be elicited by a range of acoustic and non-acoustic stimuli (Djupesland, 1976). The best known and most studied is the acoustic reflex (AR), a middle ear muscle contraction to moderate to high intensity sound. Testing of the AR is a part of audiologic assessment, and an absent AR may be suggestive of damaged afferent or efferent limbs of the sensorimotor circuit. However, recent evidence suggests that the AR may not be a pervasive phenomenon, even in people with very good hearing sensitivity and no obvious auditory system dysfunction (Flamme, Deiters, Tasko, & Ahroon, 2017; McGregor et al., 2018). Further, AR findings obtained with standard clinical procedures do not necessarily generalize to other behaviorally relevant acoustic stimuli, such as impulsive sounds (Deiters et al., 2019). Various forms of tactile and electrical stimulation to the skin of the external ear and regions of the face are also known to be a reliable elicitor of MEMCs (Djupesland, 1976; Djupesland & Tvete, 1979; Klockhoff & Anderson, 1959, 1960), suggesting that multiple afferent pathways (e.g. cranial nerve V and X) converge onto middle ear motor neuron pools (Mukerji et al., 2010). As with the AR, the adaptive role of somatosensory-mediated responses is not clear.

In addition to responding to acoustic and somatosensory stimuli, the middle ear muscles contract in concert with other motor activity. Studies dating back to the middle of the last century have reported MEMCs concurrent with motor behaviors (Carmel & Starr, 1963, 1964; Starr & Salomon, 1965). These studies produced a number of relevant findings. First, MEMCs were reliably observed when cats engaged in movement of the head and body within the animal enclosure. Second, MEMCs were also observed when the cats spontaneously vocalized and the middle ear activity typically preceded the onset of vocalization by 75-500 ms. Third, MEMCs were observed during movement and vocalization even when the cat was deafened by eighth nerve ablation, demonstrating the neural pathways were unrelated to sound transmission. Fourth, the size of MEMCs during prolonged acoustic exposure were modulated by movement, indicating that middle ear activity is the consequence of different source interactions. The authors of these studies concluded MEMCs were not simply reflexive responses that rely solely on elicitor characteristics. Instead central processes dynamically control characteristics of the response.

The size and location of middle ear muscles complicate direct recording of muscle activity, at least in audiologically-normal humans performing behaviorally relevant tasks. As a result, studies that directly record middle ear muscle activity have relied on humans while under general or local anesthesia or with significant auditory pathologies such as a perforated tympanic membrane (Borg & Zakrisson, 1975; Salomon & Starr, 1963). Most human studies employ indirect measurements of middle ear muscle activity via impedance audiometry, in which middle ear impedance changes are assumed to be the result of middle ear muscle activity. The impedance-based approach is convenient but not without limitations. Measurement validity can be compromised by movement of the measurement probe or external sound sources contaminating the ear canal recordings. This is particularly relevant to studies of motor activity. Additionally, clearly disambiguating stapedius and tensor tympani activity is not possible. These limitations notwithstanding, the impedance-based approach has been an essential research tool for characterizing MEMCs among healthy and hearing disordered populations.

Human studies have reported MEMCs concurrent with motor behaviors such as swallowing (Klockhoff, 1961; Wersall, 1958), yawning (Klockhoff, 1961), vocalization (Borg & Zakrisson, 1975; Salomon & Starr, 1963; Simmons, 1964a, 1964b) and eye-related activity (Gruters et al., 2018; Salomon & Starr, 1963). Further, it appears that movement-related MEMCs also occur during sleep, indicating that conscious awareness is not a necessary condition (De Gennaro, Ferrara, Urbani, & Bertini, 2000; Slegel, Benson, Zarcone Jr, & Schubert, 1991). Of these behaviors, vocalization and eye-related motor activity have received the most attention.

Salomon and Starr (1963) and Borg and Zakrisson (1975) both examined the relation between vocalization and MEMCs in humans by performing direct recordings of stapedius and tensor tympani muscle activity in individuals with tympanic membrane perforations. In both studies, the authors found that MEMCs tended to precede vocalization, though the specific latencies varied by participant and muscle. Onset of tensor tympani activity preceded vocalization by as much as 300 ms (Salomon & Starr, 1963) while onset of stapedius activity occurred less than 75 ms before vocalization in more than 90% of cases (Borg & Zakrisson, 1975). Although a more recent study calls into question whether sound production is a necessary precursor to elicit vocalization-related MEMCs (Kawase, Ogura, Kakehata, & Takasaka, 2000), the observation of middle ear activity preceding sound production suggests a non-auditory neural pathway. Some have suggested that this non-auditory pathway is central in origin (Carmel & Starr, 1963, 1964), while others have provided evidence for a peripheral origin via a reflex pathway involving the laryngeal nerves (McCall & Rabuzzi, 1973). Regardless of the neural pathway, this observation has led to the hypothesis that vocalization-related MEMCs serve to reduce the degree of low frequency masking caused by vocalization to allow improved detection of external sound sources (Borg & Zakrisson, 1975).

Voluntary eye-related motor activity has also been shown to elicit MEMCs. Using direct muscle recording, Salomon and Starr (1963) reported that voluntary eye closure resulted in tensor tympani activity at, or slightly before, the onset of periorbital muscle activity. This coincident activity was highly reliable and not prone to decay. Additionally, the size of the tensor tympani response scaled with the degree of voluntary eye closure force. Reflexive eye closure, elicited by a brief puff of air to the orbit, resulted in tensor tympani activity that lagged eye muscle activity by 20-60 ms. Furthermore, reflexive eye closure was less effective in eliciting tensor tympani activity and was prone to habituation, with extinction typically occurring following the first few trials. In contrast, the stapedius muscle was not observed to regularly activate during either voluntary or reflexive eye closure.

Recently, Gruters et al. (2018) examined the relationship between movements of the tympanic membrane (measured with an otoacoustic emission probe) and saccadic eye movement in humans and rhesus monkeys. A number of intriguing findings emerged. First, ear canal pressure changes were observed approximately 10 ms prior to the onset of a saccadic eye movement. These pressure changes lasted throughout the eye movement and into the eye fixation period between successive saccades. Second, the amplitude of ear canal pressure change scaled with the amplitude of the saccadic eye movement. Finally, the amplitude and phase of the eardrum movements were associated with the direction and amplitude of the eye saccades. While the authors interpreted these findings within the perspective of multi-modal sensory integration, it suggests that MEMCs can be precisely graded to the parameters of coincident motor activity.

The middle ear muscle system is a site of complex sensorimotor integration. There is continued need to gain a better understanding of the relationship between MEMCs and non-middle ear motor activity, non-acoustic sensory elicitors and acoustic sensory elicitors. This knowledge would have both scientific and practical implications. Carefully designed studies can help elucidate the range of neural circuits involved in activation of middle ear muscles, including afferent pathways across different sensory modalities and sites, and efferent pathways to the middle ear muscles from central structures. Despite known complexities in the activation of middle ear muscles, non-acoustic MEMCs have received little attention in both clinical applications and by those who assume a protective role for MEMCs against noise exposure (Price, 2007a, 2007b; Price & Kalb, 1991).

The aim of the current study was to (1) examine the occurrence rates of MEMCs in response to voluntary closure of the eye; (2) assess the effect of varying levels of eye closure effort on the occurrences rate of MEMCs; and (3) assess the association between occurrence rates of eye closure related MEMCs and demographic and clinical factors in a large group of participants with excellent hearing sensitivity and no signs or symptoms of abnormal middle ear function. An impedance-based approach was used to infer MEMCs from ear canal pressure changes in response to a click probe. Due to the indirect approach used, it was not possible to disambiguate the contributions of tensor tympani and stapedius for a given MEMC. Eye closure was selected as a simple motor task because previous work indicated that voluntary periorbital muscle activity elicits MEMCs and eye closure is a simple voluntary motor gesture that might routinely occur in military operational environments.

Methods

Participants

Study participants included 190 adults ranging from 18 to 55 years of age at the time of enrollment. Most participants were female (71%), and all were noninstitutionalized individuals drawn from the general population of Kalamazoo, Michigan between 2015 and 2017. The large number of female participants was likely due to the recruitment location of participants, which was a health and human services college in which the fields of study are mostly female. This gender imbalance was established at the time of entry to the study (i.e., there was no gender difference in exclusion or attrition). Oversight of human research subject protection was provided via institutional review boards at Western Michigan University (Protocol #15-04-09) and the U.S. Army Medical Research and Materiel Command (HRPO # A-18436.2). Participants were reimbursed for their time at the end of each visit.

Inclusion criteria restricted participation to those at least 18 years of age, willing to commit 2-4 hours of time across two visits, with no current ear pain and no reported history of Bell's Palsy, concussion or unexplained dizziness. Participant exclusion criteria included anatomy preventing proper fit of the measurement equipment, evidence of excessive cerumen, irritation or ear infection, pure tone air conduction thresholds at any single test frequency poorer than 10 dB HL from 125 Hz to 1 kHz or poorer than 20 dB HL from 2 to 8 kHz (including 3 and 6 kHz), left-right ear threshold asymmetry greater than 20 dB at any test frequency, evidence of abnormal middle ear function based on clinical conventional and wideband acoustic immittance and wideband tympanometry, absent clinical acoustic reflexes (defined by 0.02 mmho immittance change and growth), or evidence of abnormal trigeminal or facial nerve function by screening. In short, participants exhibited excellent hearing and, from an audiological perspective, were unremarkable.

Description of Study Procedure

Enrollment and Candidacy Visit

Potential participants were recruited through presentations, handouts and email canvasses. Interested persons contacted the lab for more information and a brief summary of study procedures, risks, and inclusion/exclusion criteria was provided. Those with continued interest were scheduled for an initial "enrollment and candidacy" visit, which included a detailed study description, documentation of informed consent, and a determination of candidacy to participate in the main experiment. Determining candidacy was a multi-step procedure. First, participants completed a questionnaire about ear and hearing issues, trigeminal and facial nerve health as well as current and past noise exposure. Next, a video otoscope with USB connection to a PC (Welch-Allyn Digital Macroview®, Skaneateles, NY) was used to capture video images of both the external ear and tympanic membrane of each ear.

Pure tone air and bone conduction threshold testing was conducted in a double-walled sound booth with ambient noise levels permitting testing below -10 dB HL at all stimulus frequencies. Audiometric stimuli were controlled using the Nelson Acoustics Audiometric Research Tool (ART, VIacoustics, Austin, TX) automatic audiometry software utilizing National Instruments (NI) Hybrid PXI/PXIe-4461 modules (National Instruments, Austin, TX). Air conduction signals were delivered with Sennheiser HDA-200 circumaural earphones (Sennheiser, Wedemark, Germany) and bone conduction signals were delivered through a RadioEar B-71 bone oscillator (RadioEar, Middelfart, Denmark). Participant responses were tracked via a custom response switch box. Pure tone air conduction thresholds were obtained for each ear at octave frequencies from 0.125 to 8 kHz plus the inter-octave frequencies of 3 and 6 kHz. Bone conduction thresholds were obtained at 0.5, 1, 2, 4 kHz with the transducer placed on the forehead. Threshold was defined as the lowest presentation level producing a 50% or greater likelihood of response on at least three ascending trials, using a 5-dB step via the modified Hughson-Westlake procedure (Carhart & Jerger, 1959).

Following threshold testing, a brief trigeminal and facial nerve screening was conducted. Sensory function of the trigeminal nerve was screened using light tactile stimulation of each side of the upper, middle and lower face with a cotton ball and trigeminal nerve motor function was screened by observing range of motion of the mandible, symmetry of masseter and temporalis contraction during voluntary teeth clenching and participant’s voluntary resistance to the examiner’s attempt to elevate and depress the mandible. Facial nerve motor function screening involved observing for facial asymmetry at rest, during smiling and during bilateral eye closure and assessing the voluntary resistance to examiner’s attempt to open the participant’s tightly closed eyes and mouth. Next, a full middle ear evaluation was completed using an Interacoustics Titan® middle ear analyzer (Interacoustics, Middelfart, Denmark). The Titan® was used to conduct conventional 0.226 kHz and wideband tympanometry, wideband immittance, acoustic reflexes, and acoustic reflex decay. Wideband immittance and tympanometry were completed using a band-limited click stimulus. Tympanometry and wideband tympanometry were completed using a single pressure sweep from +200 to -300 daPa. Ipsilateral and contralateral acoustic reflex traces were obtained in each ear using pure-tone elicitors (700-800 ms) at 0.5, 1, 2 and 4 kHz and a conventional 0.226 kHz probe tone. Elicitors were presented in 5 dB increments beginning at 80 dB HL and ceasing when a repeatable immittance change of 0.05 mmho was measured, or at 100 dB HL, whichever occurred first. Although a 0.02 mmho immittance change was used to indicate the presence of an acoustic reflex, continuing elicitor presentation to a maximum of a 0.05 mmho immittance facilitated examination of reflex growth with presentation level. Participants who met the candidacy requirements were scheduled to participate in the main experiment while those who did not meet the candidacy requirements were dismissed from the study. The initial visit did not exceed two hours.

Main Experimental Visit

The main experimental visit typically occurred within two weeks of the enrollment and candidacy visit. This visit began and concluded with video otoscopy, pure tone air conduction thresholds, conventional and wideband tympanometry and wideband immittance to rule out changes since enrollment or during the visit. The main experiment included assessment of MEMCs during twelve different test conditions. For nine of the test conditions, participants were exposed to different types of brief acoustic elicitors (see Deiters et al. (2019) for results obtained from brief acoustic elicitors). Two conditions involved non-acoustic elicitors. One non-acoustic condition exposed participants to a pneumotactile stimulus that was presented to different locations on the face. A second non-acoustic condition, which is the focus of this study, elicited voluntary unilateral eye closure at different effort levels. The twelfth condition required the participant to engage in one of five possible test conditions designed to assess either the presence of, or potential for, associative learning involving the middle ear muscle system. In all conditions, middle ear status was monitored using a research-grade otoacoustic emissions (OAE) probe and five muscles of the head, neck and upper extremity were monitored using surface electromyography (EMG, see Figure 1).

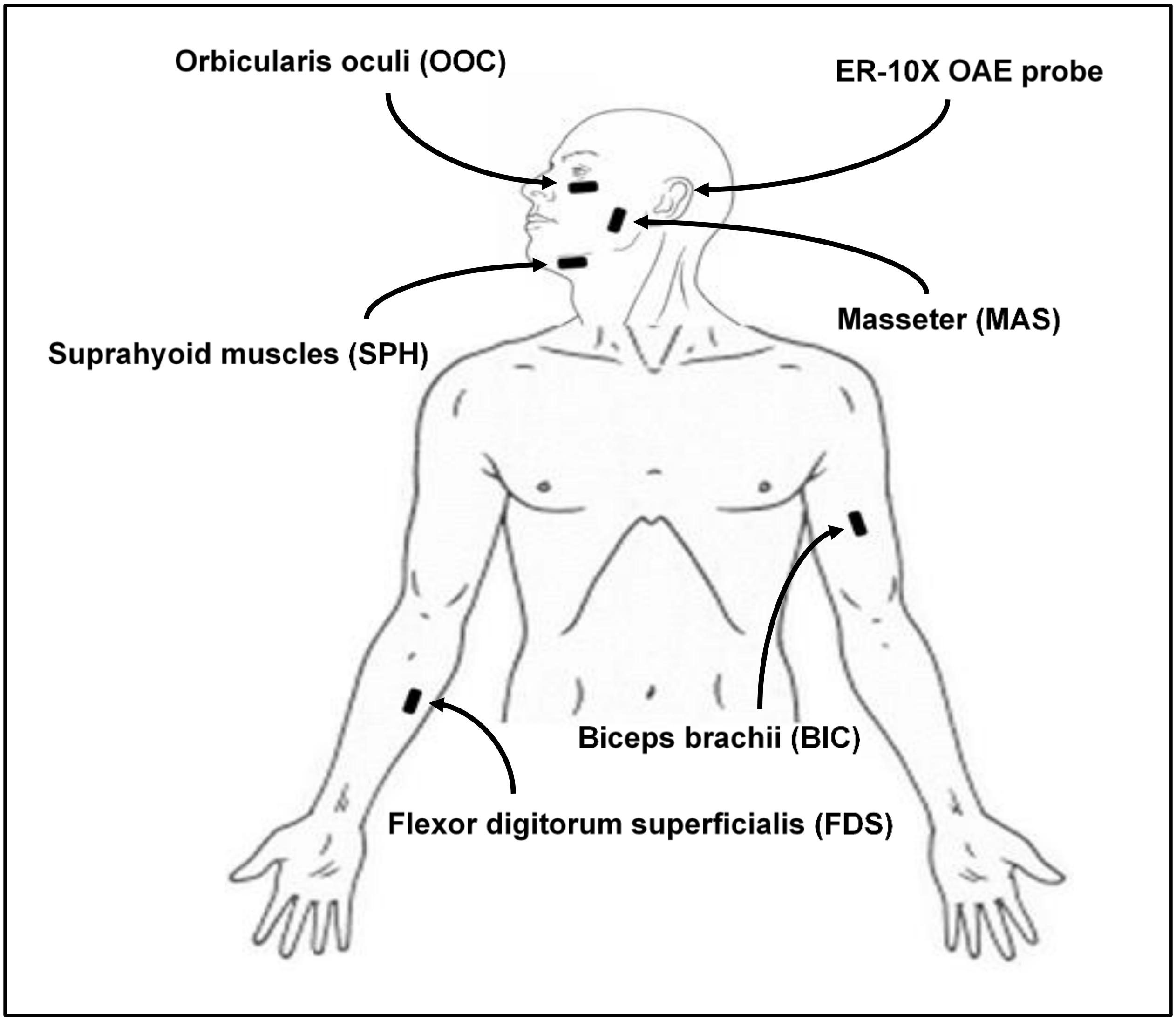

Figure 1. Location of EMG electrode array and OAE probe. Note that all electrodes except the FDS were positioned ipsilateral to the OAE probe.

The voluntary unilateral eye closure task was chosen as a simplification of the situation wherein a person closing their non-dominant eye while aiming a shoulder-mounted rifle. OOC muscle activation for different reasons (e.g., voluntary, spontaneous, reflexive) can lead to different EMG morphology (Kaneko & Sakamoto, 1999), which could indicate different motor control mechanisms. Different control mechanisms could imply different relationships between OOC and MEMC.

While the associative learning condition was always presented last, the presentation order of the acoustic and non-acoustic elicitor conditions was randomized. The first level of randomization was between acoustic and non-acoustic elicitation. Then, tasks within each of these classes were permuted randomly for each participant. As a result, the eye closure condition could only occur in one of four positions in the overall presentation order: first, second, tenth or eleventh.

Surface Electromyography (EMG) Recording

Surface EMG recordings were made using a Delsys Bagnoli® 8-channel Desktop system and Delsys 3.1® double differential electrode sensors (Delsys, Natick, MA). Electrode sensors were reusable, active electrodes that contained three parallel silver bars, 10 mm in length, 1 mm in width, and spaced 10 mm apart. The system applied a double differential procedure to improve spatial localization of the EMG recordings and to minimize crosstalk with other recording sites. EMG activity was pre-amplified by a factor of 10 at the electrode site, thus improving the signal-to-noise ratio prior to transmission to the main amplifier. Additional gain was provided by the main amplifier resulting in a total gain multiplier of 1000 (i.e., 60 dB). The signal was also band-pass filtered (20-450 Hz) prior to analog-to-digital conversion. The surface electrodes were placed at five locations on the head, neck and upper extremity (Figure 1). Standard procedures were used to identify the location to place each electrode sensor (Criswell & Cram, 2011) and the recording sensors were positioned so that each electrode bar was perpendicular to, and the trio of electrodes ran parallel with, the underlying muscle fibers.

The muscles included orbicularis oculi (OOC), masseter (MAS), the suprahyoid muscle complex (SPH), biceps brachii (BIC), and flexor digitorum superficialis (FDS). Four of the five EMG recordings were ipsilateral to the ear with the OAE probe, described below. The FDS sensor was placed contralateral to the OAE probe and was used to monitor finger activity during test conditions unrelated to the current study. Although all five EMG sites were monitored during the main experiment, the OOC activity is of primary interest to the current study. The OOC sensor was positioned with a horizontal electrode orientation just inferior to one lower eyelid. The sensor was placed as close as possible to the lower eyelid while minimizing any mechanical interference by the sensor during tight eye closure. The monitored eye was contralateral to the finger nominated by the participant as the one they would imagine using on the trigger of a gun (e.g., the left eye of a right-handed shooter). The MAS sensor was placed with a vertical electrode orientation in the region between the zygomatic arch and the angle of the mandible. The SPH sensor was placed over the submental/submandibular space with the electrode orientation parallel to the inferior margin of the mandibular body. The BIC sensor was placed over the belly of the muscle while the participant flexed the elbow at a right angle with the forearm in a supinated position. The FDS sensor was placed on the inner forearm, located on a line two thirds the distance from the medial epicondyle of the humerus to the most distal point of palmaris longis tendon. A ground electrode was placed over the olecranon of the ulna at the elbow joint.

Prior to electrode placement, a disposable alcohol pad was used to gently scrub the skin surface, followed by a repeated "tacking" of the skin surface with biomedical tape to reduce electrode-skin impedance. The electrodes were then secured to the skin using disposable electrode collars. Following electrode placement, a series of motor tasks were completed to confirm the validity of EMG recordings. These tasks also served as reference data for subsequent normalization procedures to account for between-subject differences in EMG levels. Tasks included tightly closing the eye (OOC), chewing a small mint (MAS), forcefully pressing the tongue against the roof of the mouth (SPH), isometric flexion at the elbow (BIC) and repeatedly pulling the trigger of a toy cap gun (FDS). Each task was repeated at least three times. If task-related EMG activity was not observed, the electrode sensor was repositioned. If activity was not observed after repositioning, that EMG site was not used in subsequent analysis. In 28 cases, the presence of facial hair prevented placement of the MAS and/or SPH electrode sensor.

Middle ear Recording

An Etymotic Research ER-10X® OAE probe (Etymotic Research, Elk Grove Village, IL) was used to present probe stimuli and transduce the signal in the ear canal. The OAE probe was fitted to the ear contralateral to the participant's preferred trigger finger. The probe fit was continually monitored throughout the experiment via regular visual inspection of the probe in the ear and by assessment of the ear recordings of a swept sine wave (0.1-16 kHz) stimulus that was presented through the probe before and after each test condition.

Impedance change was measured using a method described by Keefe, Fitzpatrick, Liu, Sanford, and Gorga (2010) and modified for the present study. Briefly, the probe stimulus was a train of clicks presented at a 20 Hz repetition rate. Each click was the impulse response of a digital filter with a passband from 0.2 – 8 kHz. Probe clicks were digitally sampled at 44.1 kHz and presented at 93 dB peak SPL as measured in an IEC-60318-4 occluded ear simulator. The advantage of using a band-limited click over a pure tone probe is that it allows for assessment of middle ear responses across a wider range of frequencies.

Data Acquisition System

The instrumentation used during the main experiment was controlled with custom-written MATLAB (The MathWorks, Inc., Natick, MA) scripts using a Windows-based (Windows, Redmond, WA) PC workstation (Dell model 7910, Dell, Austin, TX) connected to a National Instruments (NI, Austin, TX) PXIe-1082 chassis. NI PXI/PXIe (hybrid) 4461 dynamic signal analyzer and PXIe-4499 modules were used to produce the probe click and synchronously sample all input channels during recording. All input channels were sampled at 44.1 kHz (24 bit).

Experimental Task

As noted earlier, the experimental task for this study was randomly embedded within a battery of other test conditions. Participants were seated in front of a large computer display with the OAE probe and the EMG electrodes in place. Additionally, an Etymotic Research ER-4PT® high-output commercial insert earphone (Etymotic Research, Elk Grove Village, IL) was placed in the ear contralateral to the OAE probe. The earphone was used to deliver instructions to the participant. The experimental task required participants to close one eye using light, tight, or maximum muscular effort level.

A visual stimulus was used to help participants duplicate and grade muscular effort. A still image of an investigator's face with only the monitored eye closed was displayed. A thermometer-type visual analog image was presented on the computer monitor, alternating between being empty and being filled to varying degrees (Figure 2). An empty thermometer indicated an unclosed eye (no closure effort). The participants were instructed to watch the screen and, when the thermometer level changed, close the eye with an effort level they believed matched the thermometer level. There were three different effort levels. The thermometer could be 3/10 full, indicating a low level of muscular effort (light closure effort), 8/10 full indicating a greater level of muscular effort (tight closure effort), or completely full, indicating maximum muscular effort (maximum closure effort). Each magnitude setting lasted three seconds followed by a period in which the magnitude reset to zero. For each participant, a total of 14 trials were presented. The interstimulus (ISI) interval varied randomly around a mean ISI of five seconds. The experimental run lasted 114 seconds. The first and last stimuli were always full magnitude, prompting maximum closure effort. The twelve intermediate stimuli include six light closure effort trials and six tight closure effort trials, presented in random order.

Data Processing and Analysis

Cursory data monitoring took place during acquisition in order to identify gross errors and equipment failure. Prior to analyses, a systematic data review process was implemented. Part of the review process included a visual inspection of the ear canal acoustic channel and five EMG channels for each of the 14 trials across the 190 participants. This review was completed to verify the participant was producing OOC EMG patterns that correspond to the eye close instruction, flag unusual/atypical ear canal and EMG recordings and identify concomitant EMG activity in the non-OOC recording sites.

EMG Processing

All EMG recordings were notch filtered at 60 Hz to eliminate possible artifacts from power line noise, and a root-mean-square (RMS) trace of each EMG channel was generated using a 50 ms moving window. The identification of the onset and offset of OOC EMG activity associated with each of the eye close gestures was necessary to assess for corresponding changes in middle ear muscle activity. Eye closure related onset and offset identification was a two-step procedure. The first step employed a MATLAB-based automated detection algorithm. The detection algorithm employed a spectral subtraction procedure (Zavarehei, 2019) to remove background noise spectrum from the OOC EMG signal and facilitate onset/offset identification. The identification of related eye closure onset and eye closure offset was a two-step process. First, for each trial, the OOC RMS trace was extracted over a time window beginning one second prior to the onset of the eye closure instruction and ending three seconds following the offset of the eye close instruction. Eye closure onset was operationally defined as the point when the RMS trace first exceeded the 95th percentile of the distribution of RMS levels for the time window. Eye closure offset was identified as the point where the RMS trace dropped below the 95th percentile of the distribution of RMS levels for the time window. Results of the automated detection method were then visually inspected and the eye close onset and offset times were modified as necessary. Peak and mean EMG levels were calculated for each eye close gesture and expressed as a percentage of the maximal effort task described earlier. An example of the raw (notch-filtered) and RMS traces for the OOC recording site during a series of voluntary eye closures is shown in Figure 2.

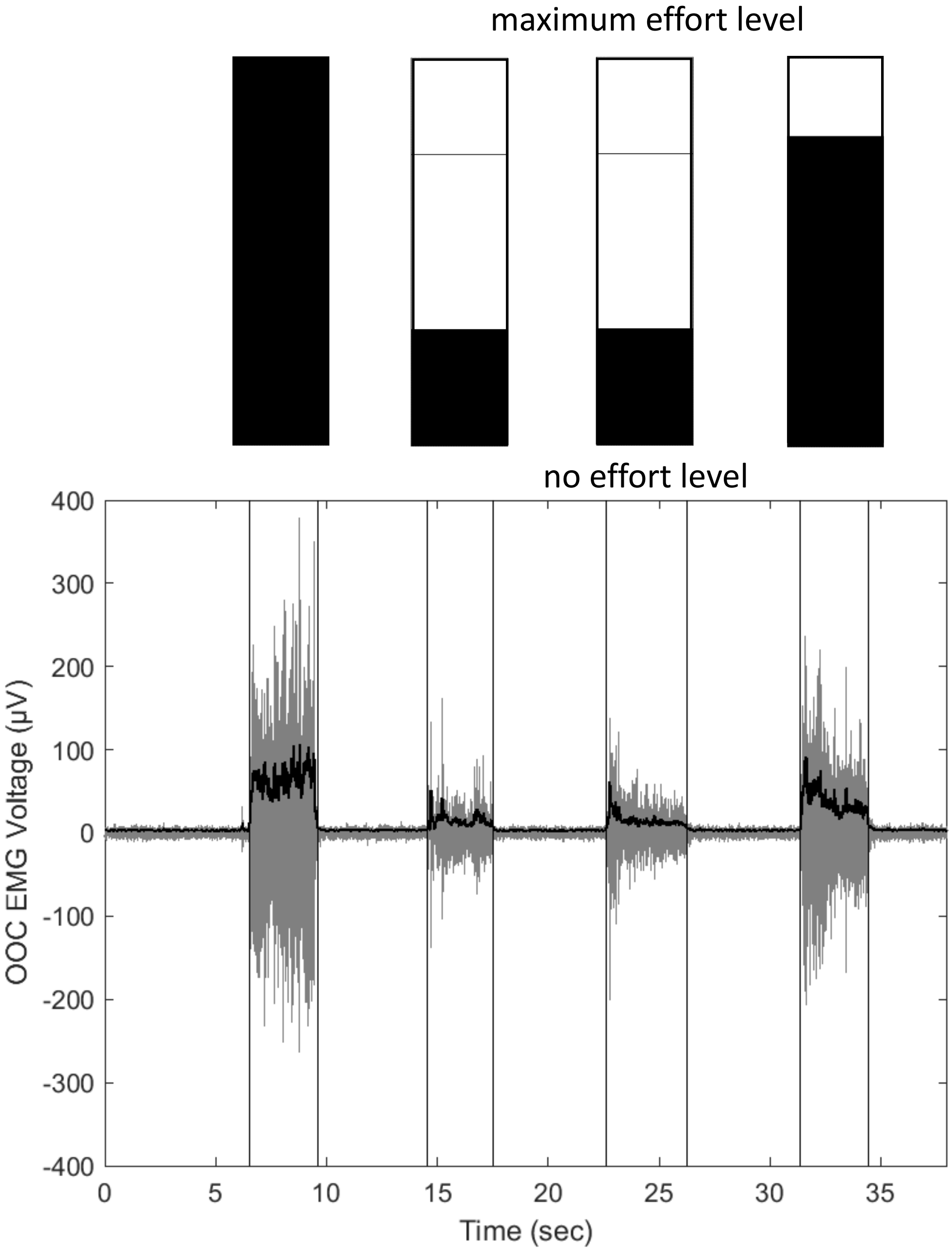

Figure 2. Raw (gray) and RMS (black) OOC EMG activity plotted as a function of time. This sample is drawn from the first 40 seconds of a 114 second record. The vertical lines represent the onset and offset of each eye close gesture. The black “thermometers” located above the plot represent the image presented to the participant to prompt eye closure of different effort levels. Each image was interleaved with an empty thermometer which instructed the participant to fully relax the eye closure gesture. Note that, for this example, the visual stimulus was successful in grading the level of OOC EMG activity.

Middle Ear Recordings

The eye closure onset and offset times were used to segment the entire 114-second record into a consecutive series of "baseline" and "elicitor" intervals. The baseline interval was defined as the period bound by the offset of the preceding eye close (or in the case of the first trial, the beginning of the record) to the time 500 ms prior the onset of the eye close gesture. The elicitor interval was defined as the period beginning 500 ms prior to the eye close onset and ending at eye close offset.

A change in middle ear status in response to eye closure was assessed using a method modified from Keefe et al. (2010). Briefly, this involved measuring the root-mean-square (RMS) differences between each click waveform developed in the ear canal during the elicitor interval and the ensemble-average of all clicks located within the previous baseline interval. The impedance change caused by MEMCs presented as a consistent change in the RMS response differences associated with the onset of eye closure. Figure 3 is a plot of a single trial. In this example, there is a clear correspondence between the OOC EMG activity and the magnitude of the RMS deviations recorded in the ear canal. The eye closure onset was used to time-align the RMS click deviations for all trials within an experimental condition (Figure 4). The 25th percentiles of the time-aligned RMS deviations indicated the presence of a reliable MEMC (Deiters et al., 2019). Three independent raters (authors GF, ST, and KD), who were blinded to the participant identity and eye closure effort level, examined all available 25th percentile traces (heavy line in Figure 3) for evidence of stimulus-linked changes in the RMS plot and provided a binary judgement of presence or absence of a middle ear muscle contraction regardless of response amplitude. Two criteria were used to determine the occurrence of MEMCs. A "loose" criterion required 2/3 agreement among the judges and a "strict" criterion required unanimous agreement among the judges.

Calibration of Audiometric Equipment

The calibration of all acoustic transducers was verified at the beginning and end of each test day. A G.R.A.S. RA0045 occluded ear simulator (G.R.A.S., Holte, Denmark) was used to acquire calibration recordings for the OAE probe and insert earphone and a G.R.A.S. type 43AA ear simulator was used for the audiometric headphones. A Quest QC-20 acoustic calibrator (Quest, Oconomowoc, WI) was used to present calibration signals into the OAE probe microphone and a field microphone (G.R.A.S. 40AC) that was used to monitor the experiment. Microphone signals were preamplified (G.R.A.S. Type 26AC) and routed through a power supply (G.R.A.S. Type 12AA) prior to digitization using an NI hybrid PXI/PXIe-4461 dynamic signal analyzer module mounted within an NI PXIe-1082 chassis. The Quest calibrator was regularly validated against a G.R.A.S. Type 42AP Intelligent Pistonphone.

Statistical Analysis

Stata® software (StataCorp LLC, College Station, TX) was used to perform descriptive and inferential statistical analysis as well as assess rater agreement using Cohen's kappa (Cohen, 1960). Logistic regression analysis was used to examine univariable relationships between demographic variables, hearing thresholds, clinical acoustic reflex results, as well as measures of eye closure and occurrence of MEMCs. The univariable results were then used to discard variables unlikely to retain significance in a multivariable logistic models using conventional procedures (Hosmer, Lemeshow, & Sturdivant, 2013). A final multivariable logistic regression model was then developed by sequentially discarding variables that failed to retain significance in the multivariable context.

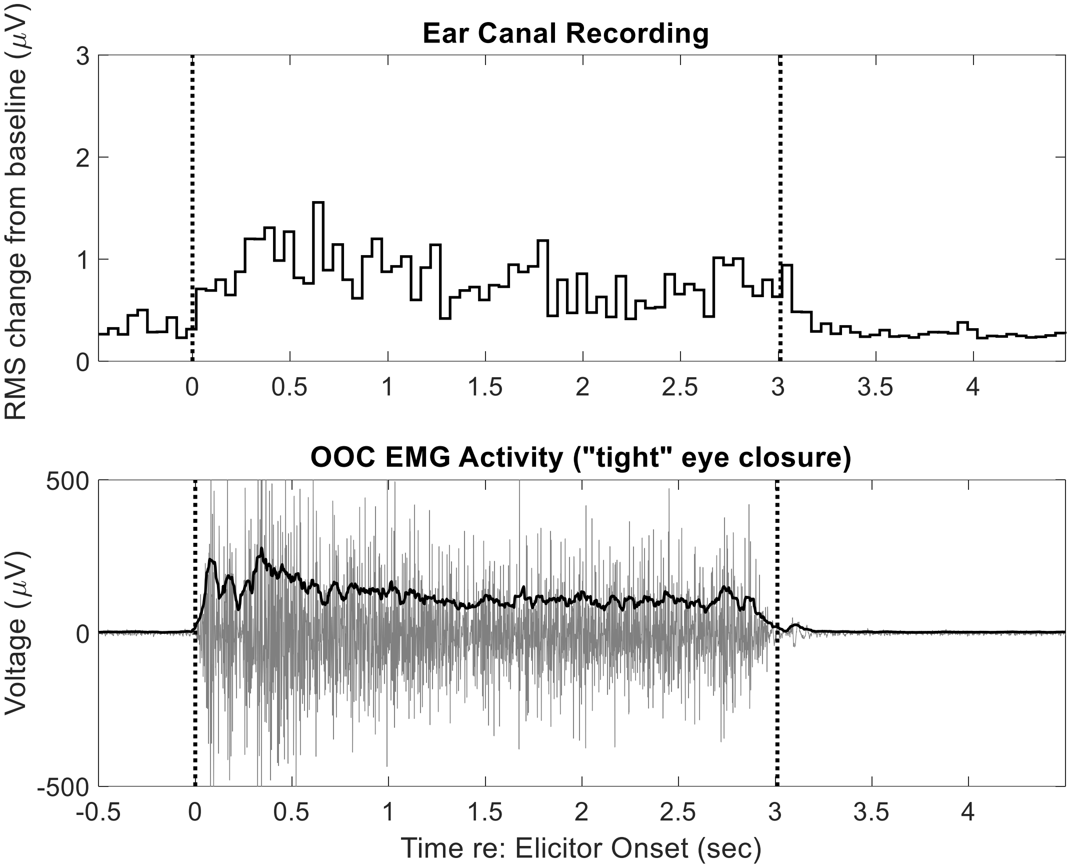

Figure 3. The top and bottom panels are synchronous plots of ear canal recordings and OOC muscle activity, respectively, for a single eye close trial at tight eye closure effort level. The top panel is a stair plot with RMS click deviations plotted as a function of time, relative to the eye close onset. Each step in the stair plot represents the RMS click deviation for successive 50-msec clicks. The bottom panel is a plot of raw (gray) and RMS (black) OOC EMG activity for the same time interval. The broken vertical lines represent the onset and offset of eye closure. Note that the ear canal recorded RMS click deviation pattern approximates the OOC EMG activation pattern.

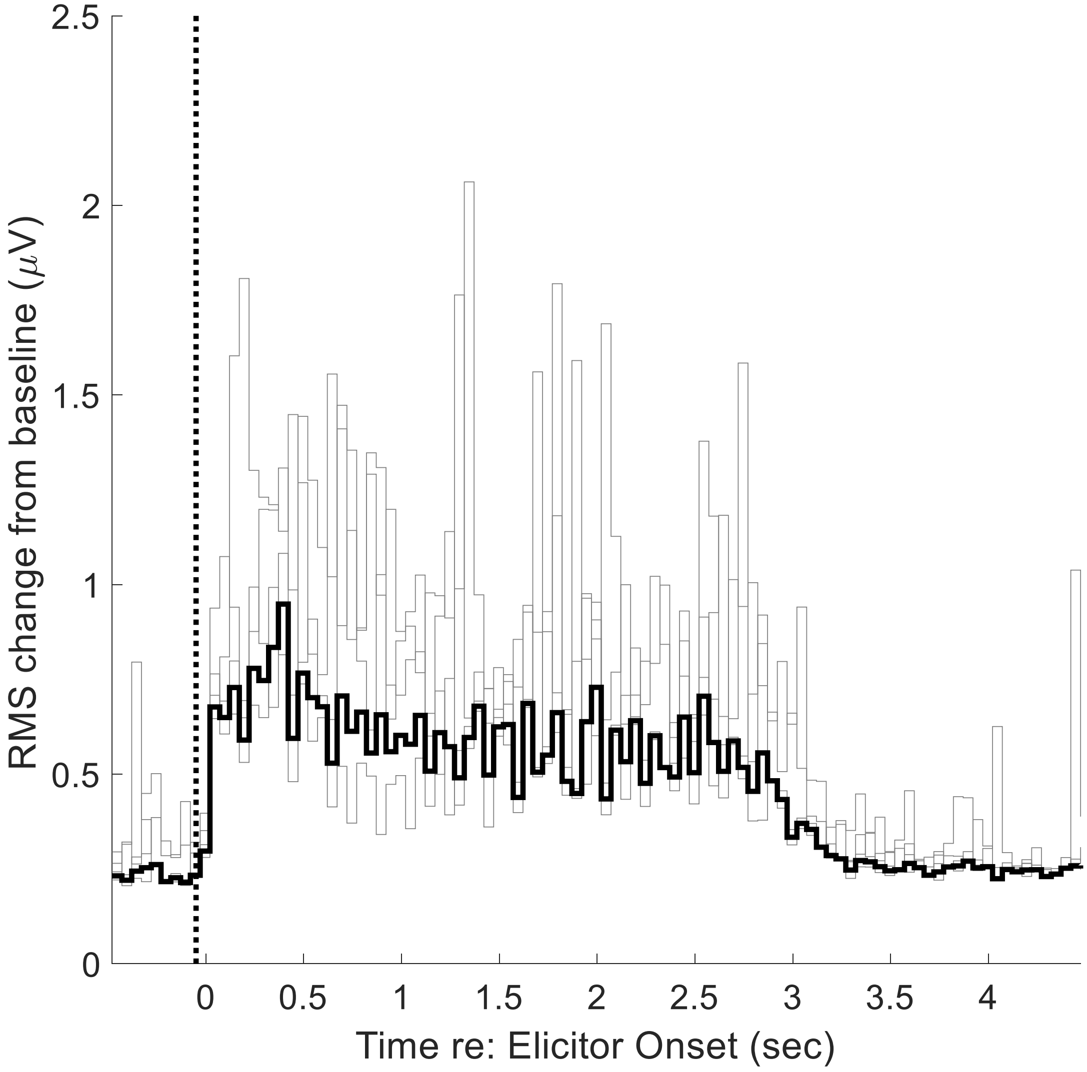

Figure 4. A stair plot with RMS click deviation plotted as a function of time, relative to the eye close onset (broken vertical line). Each light gray line represents the RMS click deviation series for an individual trial within a single condition (tight eye closure effort level), relative to the mean RMS during the baseline period. The heavy black line represents the 25th percentile of the data distribution (i.e., trial-level data for this participant on this task) and was used by judges to assess for the presence or absence of MEMCs.

Results

Inter-rater Reliability

The presence of MEMCs was based on visual inspection of the 25th percentile trace of the ensemble of responses for a given effort level within a given participant (heavy black line in Figure 4). A panel of three judges independently evaluated results for each participant at each effort level. Each judge evaluated a total of 545 25th percentile traces across a total of 190 participants. Not all participant-effort level combinations were available for review. This was typically due to an inability to identify clear onsets or offsets of the eye close gesture in the EMG recordings. A total of 185 participants underwent judgements for the light and tight effort eye closure conditions and 175 participants were judged for the maximum effort eye closure condition. Inter-rater reliability across the three combinations of judge pairings was measured using Cohen's kappa. Absolute agreement was 90.3, 90.9 and 96.2% for each pair-wise combination of the three raters. Expected agreement based on chance alone for the same three judge pairings was determined to be 80.5, 80.9 and 87.5%, respectively. Though absolute agreement was extremely high, due to the high expected agreement based on chance, Cohen's kappa values for the three judge pairings were 0.50, 0.52 and 0.70, which placed agreement levels in the moderate to substantial range (Cohen, 1960).

Descriptive Analyses

Orbicularis Oculi EMG Levels Across Nominal Levels of Eye Closure Effort

The procedure of matching eye closure effort to a visual display was successful in systematic grading of muscle activity (Figure 5). The light eye closure effort condition elicited smaller amplitude signals than the tight eye closure effort condition, which in turn elicited smaller amplitude signals than the maximum effort eye closure condition. This pattern was observed regardless of whether muscle activity was estimated from the mean or maximum level, or whether activity was expressed in microvolts or as a percentage of maximum voluntary contraction. The distributional similarities in the different estimates of muscle activity was likely to result in predictor variable multicollinearity in subsequent regression modeling. Therefore, the mean OOC activity level expressed in microvolts was selected as the estimate of muscle activity level for subsequent inferential analysis.

Occurrence Rates of MEMCs

Occurrence rates of MEMCs were lowest for the light eye closure, higher for tight eye closure, and highest for the maximum closure effort (Figure 6). Depending upon the criterion used, for the light eye closure effort condition, MEMCs were observed in 55 to 75% (loose and strict rating results, respectively) of the participants. Between 89 and 93% (loose and strict rating results, respectively) of participants exhibited MEMCs when generating tight eye closure, and for maximum eye closure, MEMCs were observed in 96 to 98% (loose and strict rating results, respectively). As expected, estimated occurrence rates using the loose criterion were higher than for the strict criterion. The differences in estimated occurrence rates as a function of criteria was largest for light eye closure, indicating that judges agreed least for responses associated with this condition. In summary, these results indicate that MEMC occurrence rates systematically vary as a function of nominal eye closure effort level. However, even for the maximum eye closure effort, the lower bound of the 90% confidence intervals was less than the 95% level required to be considered a pervasive phenomenon (Figure 6).

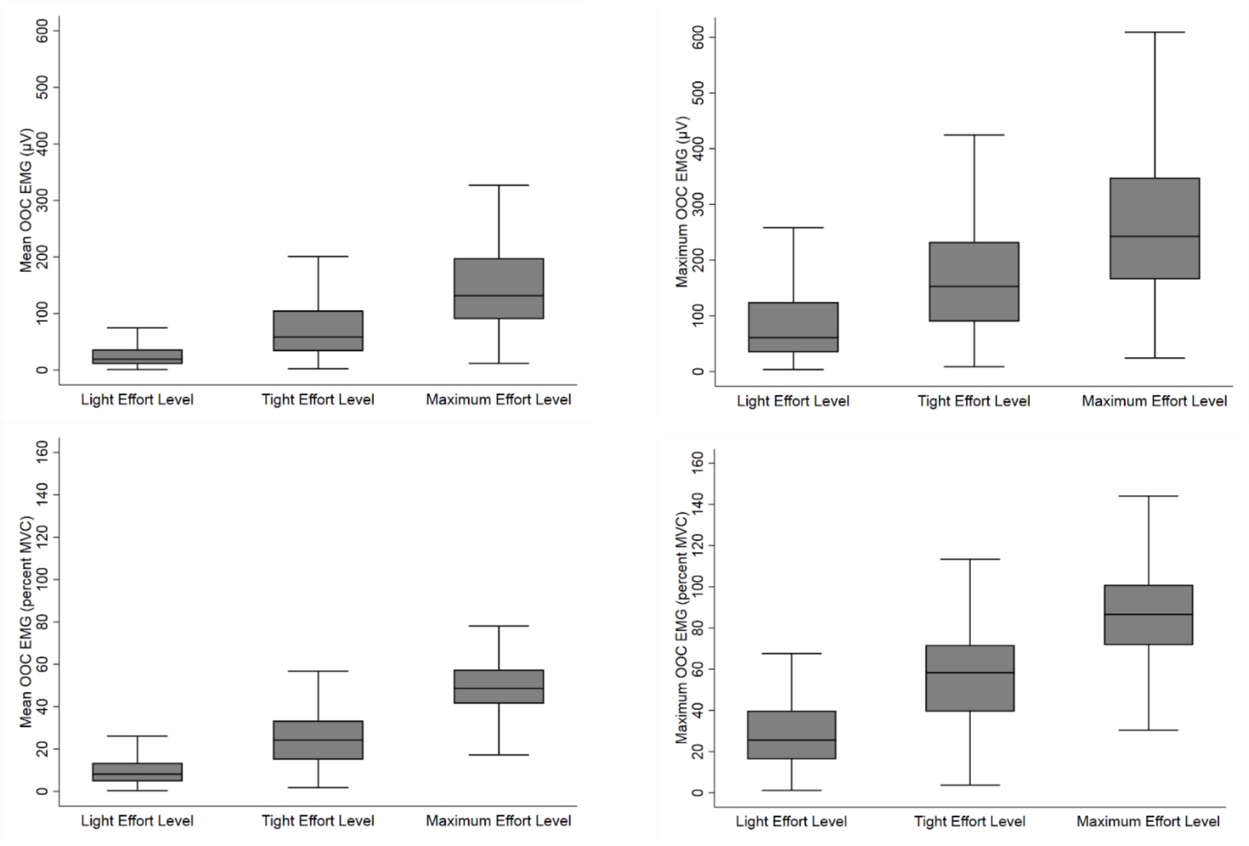

Figure 5. Box plots of four different estimates of OOC RMS EMG levels for the light, tight and maximum eye close effort levels. The upper panels represent the data in microvolts and in the lower panels the data are expressed as a percentage of maximum voluntary contraction (MVC). The left panels express EMG level based on the mean level across the gesture and the right panels express level based on the maximum EMG level drawn from the eye close gesture. The horizontal line within each box represents the mean value, the gray box bounds the interquartile range and the whiskers represent the lower and upper adjacent values.

Inferential Analyses

A multivariable logistic regression model was used to examine the quantitative relationship between OOC muscle activity and MEMC occurrence rates while controlling for potentially influential factors such as hearing thresholds, clinical middle ear testing results, participant characteristics, and experimental design features. The development of this model was a multi-step process (Hosmer et al., 2013). First, univariable analyses were conducted with all potential predictor variables. Any univariable model with a near-significant *p*-value (≤ 0.20) was identified and the associated predictor variable was retained for inclusion in the multivariable model. Non-significant univariable models ($p > 0.20$) were discarded as they were considered unlikely to retain significance in the multivariable model. A final multivariable model was developed by initial forced-entry of all near-significant variables. Then variables that failed to retain significance in the multivariable context were sequentially discarded until the model contained only significant ($p \leq 0.05$) variables using robust standard errors (Huber, 1967).

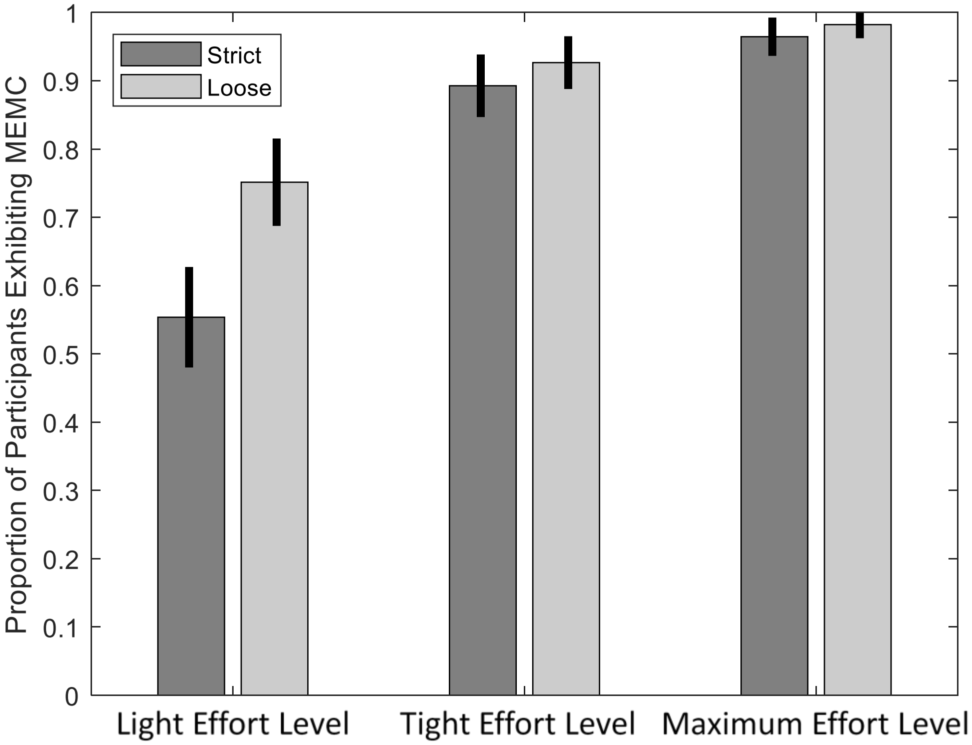

Figure 6. Proportion of participants exhibiting MEMCs coincident with the onset of eye closure-related orbicularis oculi EMG activity for light eye closure effort level, tight eye closure effort level and maximum eye closure effort level. The dark bar represents proportions of participants based on a strict agreement across judges (3/3 judges must agree MEMCs are present) and the light bar represents proportions based on a less strict criterion (2/3 judges agree MEMCs are present). The thick, black vertical lines represent the range of the 95% confidence intervals.

Identification of Potential Predictor Variables Using Univariable Analysis

The univariable results, although helpful in determining variable inclusion in a multivariable model, should be assessed cautiously as results do not control for effect modifications by other variables. Hearing thresholds appeared to be minimally predictive of MEMCs in response to eye closure during initial univariable analyses (Table 1). Only three of a possible 13 variables met the near-significant threshold for inclusion in the multivariable model. These included 0.5 and 8 kHz hearing thresholds in the ear contralateral to the OAE probe and the bone conduction threshold at 2 kHz. In summary, any influence of hearing threshold on

Table 1. Univariable logistic regression results for hearing threshold variables. Each odds ratio represents the likelihood of observing MEMCs for each 5-dB change in hearing threshold. Odds ratios and lower- and upper-95% confidence intervals (CI) that are in bold represent those variables with a near-significant *p*-value of 0.20 or less.

Hearing Thresholds (5 dB change)	Odds Ratio	Odds Ratio 95% CI Lower	 Upper
Air conduction (ear contralateral to OAE probe)			
0.5 kHz	**1.21**	**0.96**	**1.52**
1 kHz	1.01	0.81	1.26
2 kHz	0.99	0.82	1.20
4 kHz	1.00	0.83	1.20
8 kHz	**0.88**	**0.76**	**1.03**
Air conduction (ear ipsilateral to OAE probe)			
0.5 kHz	1.01	0.82	1.24
1 kHz	1.09	0.87	1.37
2 kHz	1.07	0.89	1.29
4 kHz	0.93	0.77	1.12
8 kHz	1.00	0.87	1.16
Bone conduction			
0.5 kHz	1.08	0.92	1.26
1 kHz	1.11	0.94	1.32
2 kHz	**1.17**	**1.02**	**1.33**
4 kHz	1.02	0.86	1.21

MEMC likelihood appeared to be intermittent, small in effect and, due to differing direction of effects, difficult to interpret. Similarly, for clinical acoustic reflex variables, only a handful of potential variables met the near-significant *p*-value threshold for model inclusion (Table 2). Those variables included the acoustic reflex onset latency at 1 and 2 kHz, acoustic reflex duration at 2 kHz, and static admittance. Acoustic reflex magnitude, the area under the response curve, and the maximum level at which the acoustic reflex was tested based on a stopping point of a repeatable reflex of .05 mmhos or greater, appeared to have no influence on eye close related MEMCs. Similar findings were observed for tympanometric volume, pressure, and gradient. Age, self-reported firearm or nail gun use, and self-reported tinnitus did not meet the near-significant threshold *p*-value (Table 3). Additionally, the presence of MEMCs in response to pneumotactile stimuli (another experimental condition) was not predictive of eye close related MEMCs. Two variables in this grouping did meet the near-significant threshold for inclusion in the multivariable model. These variables were the order in which the eye close condition occurred in the larger experiment and the interstimulus interval between successive eye close gestures. Together, these results suggest that eye close related MEMCs may be influenced by the timing of previous MEMC elicitors. The univariable models used to assess nominal levels of eye closure effort exhibited *p*-values that were well below the threshold for inclusion in the multivariable model (Table 4). The light effort level served as the reference condition. The odds ratios for both the tight effort level and maximum effort level were quite large relative to other variables included in the univariable analysis.

Table 2. Univariable logistic regression results for clinical acoustic reflex variables. For each variable, the odds ratio represents the likelihood of observing MEMCs for each unit change in the parentheses for that variable category. Odds ratios and lower- and upper-95% confidence intervals (CI) that are in bold represent those variables with a near significant *p*-value of 0.20 or less.

Clinical acoustic reflex variables (contralateral)	Odds Ratio	Odds Ratio 95% CI Lower	 Upper
Peak acoustic reflex magnitude (1 mmho change)			
0.5 kHz	2.88	0.01	1569.16
1.0 kHz	3.89	0.02	881.94
2.0 kHz	0.40	0.00	46.54
4.0 kHz	0.58	0.00	71.75
Area under response curve (1 mmho·ms change)			
0.5 kHz	1.16	0.92	1.46
1.0 kHz	1.10	0.92	1.32
2.0 kHz	1.04	0.86	1.25
4.0 kHz	0.98	0.81	1.19
Acoustic reflex onset latency (100 ms change)			
0.5 kHz	0.94	0.82	1.08
1.0 kHz	**0.84**	**0.73**	**0.98**
2.0 kHz	**0.87**	**0.77**	**0.98**
4.0 kHz	0.99	0.87	1.13
Acoustic Reflex duration (100 ms change)			
0.5 kHz	1.07	0.95	1.21
1.0 kHz	1.01	0.91	1.13
2.0 kHz	**1.07**	**0.97**	**1.19**
4.0 kHz	0.98	0.88	1.10
Maximum presentation level (5 dB change)			
0.5 kHz	0.96	0.75	1.24
1.0 kHz	0.98	0.75	1.29
2.0 kHz	1.08	0.85	1.35
4.0 kHz	1.01	0.84	1.21
Tympanometric volume (1 ml change)	1.41	0.69	2.87
Static admittance (1 mmho change)	**0.84**	**0.64**	**1.10**
Tympanometric pressure (1 decaPa change)	0.99	0.98	1.01
Tympanometric gradient (1 decaPa change)	1.00	1.00	1.01

Table 3. Univariable logistic regression results for demographic, self-report and features of the experiment. Eye closure was one of eleven different experimental conditions. Odds ratios and lower- and upper-95% confidence intervals (CI) that are in bold represent those variables with a near-significant *p*-value of 0.20 or less.

	Odds Ratio	Odds Ratio 95% CI	
		Lower	Upper
Age (Decade)			
18-19	1.00		
20-29	0.75	0.42	1.34
30-39	0.52	0.16	1.66
40-49	1.00	0.11	9.14
50-59	1.00	0.11	9.14
Self-reported firearm/nail gun use			
No past use	1.00		
Past use, but not in the last 30 days	0.77	0.50	1.19
Past use in the last 30 days	1.15	0.50	2.61
Self-reported tinnitus	0.96	0.63	1.45
Presentation order of Experimental condition			
1st in overall sequence	1.00		
2nd in overall sequence	**0.45**	**0.24**	**0.84**
10th in overall sequence	0.78	0.41	1.50
11th in overall sequence	0.63	0.34	1.16
Interstimulus Interval (sec)	**3.32**	**1.15**	**9.61**
MEMCs in response to pneumotactile stimulus	1.32	0.60	2.90

Table 4. Univariable logistic regression results for the nominal levels of eye closure effort level. Odds ratios and lower- and upper-95% confidence intervals (CI) that are in bold represent those variables with a near-significant *p*-value of 0.20 or less.

	Odds Ratio	Odds Ratio 95% CI	
Univariable Analysis		Lower	Upper
Light Effort Level	1.00		
Tight Effort Level	**6.70**	**3.83**	**11.74**
Maximum Effort Level	**21.90**	**9.20**	**52.11**

Multivariable Analysis

The predictor variables from all univariable regression models with *p*-values at or below the near-significant threshold of 0.20 were included in a multilevel mixed-effects logistic regression model. The only exception was the eye closure effort data. Instead, the OOC EMG data were transformed into quintiles based on the mean muscle activity level, expressed in microvolts (left side of Table 5), thus providing a more direct estimate of muscle activity. Wald tests of multiple equation maximum likelihood were used to verify which variables significantly improved the model fit. The final, best fit model only included OOC muscle activity broken out by quintile ($W = 35.93$; $p < 0.00005$). None of the variables related to hearing threshold, clinical middle ear testing, participant characteristics or experimental design features significantly improved the model fit. Activity of the OOC muscle was linked strongly with MEMCs (right side of Table 5). The odds ratios associating OOC muscle activity with eye close related MEMCs were multiple orders of magnitude larger than for any other predictor variable, ranging from an odds ratio of approximately 20 (comparing 2nd to 1st quintile) to an odds ratio over 7000 (comparing 4th and 5th quintiles to 1st quintile).

Table 5. The left side of the table is a summary of the OOC EMG levels after division into quintiles of the overall distribution. The quintiles were included in the final multivariable logistic regression model along with other variables with a *p*-value at or below the near-significant level of 0.20. The only variables that significantly contributed to the final model fit were the OOC EMG quintiles. The right side of the table is a summary of the odds ratios (relative to 1st quintile) and the lower and upper 95% confidence intervals (CI).

Multivariable Analysis OOC EMG Quintile	Minimum (μV)	Maximum (μV)	N	Odds Ratio	Odds Ratio 95% CI Lower	Upper
1 (1st -20th percentile)	0.8	18.2	109	1.00		
2 (21st-40th percentile)	18.4	40.8	109	**20.08**	**3.98**	**101.28**
3 (41st-60th percentile)	41.3	81.6	109	**942.63**	**72.00**	**12341.31**
4 (61st-80th percentile)	81.7	136.7	109	**8724.86**	**405.12**	**187903.50**
5 (81st-100th percentile)	137.7	449.4	109	**7897.06**	**337.53**	**184765.20**

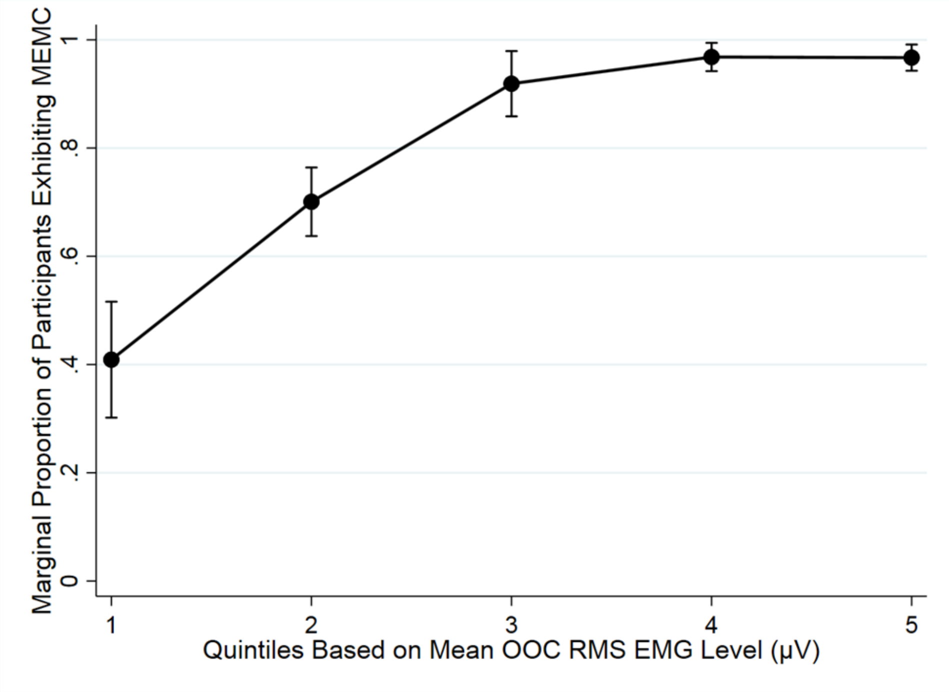

Figure 7. Marginal proportion of participants exhibiting MEMCs (strict criteria) for each quintile based on the mean orbicularis oculi RMS EMG level, expressed in microvolts. The solid circles represent the mean marginal proportion for each quintile and the error bars represent the 95% confidence interval. Each adjacent quintile is connected by a straight line.

Marginal proportions and pairwise comparisons using Bonferroni adjustment were used in post-hoc analyses. These comparisons revealed that the 1st and 2nd quintiles were significantly different from the 3rd through 5th quintiles, and the 3rd, 4th and 5th quintiles were not significantly different from each other (Figure 7). Marginal proportions of participants exhibiting MEMCs ranged from approximately 0.4 (1st quintile) to greater than 0.95 (4th and 5th quintile). The relationship between marginal proportions and quintiles of OOC muscle activity were similar to a quadratic function, showing a linear increase in the marginal proportions between the 1st and 3rd quintiles, followed by a flattening off between the 3rd and 5th quintile. Therefore, OOC muscle activity beyond the 3rd quintile was very likely to elicit MEMCs.

Discussion

Voluntary eye closure is a robust elicitor of MEMCs

The aim of this study was to assess the relationship between voluntary eye closure and MEMCs in a group of participants with excellent hearing sensitivity and normal middle ear function. Results indicated that voluntary eye closure was a robust elicitor of MEMCs and that the rate of MEMCs was positively associated with greater eye closure effort (Fig. 6) and resultant OOC muscle activity (Fig. 7). Further, multivariable analysis revealed that this relationship between eye closure and MEMCs was unaffected by other variables such as participant hearing thresholds and clinical middle ear testing results, as well as experimental design variables (Table 5). Even at the lowest measurable levels of mean OOC activity (0.8 to 18 µV), MEMCs were observed in many participants. The rate of MEMC identifications rose steadily such that when mean OOC activity exceeded 40 µV, MEMCs were observed in greater than 90% of the participants. Mean OOC activity greater than 80 µV nearly assured MEMCs. These results are consistent with and expand upon previous studies that have examined relations between the eye-related motor activity and MEMCs. Salomon and Starr (1963), who made direct recordings of middle ear muscles in two adult humans, observed tensor tympani responses were consistently present during active voluntary closure, increased in magnitude with an increase in eye closure force, typically lasted the duration of the eye closure gesture, and exhibited little habituation across repeated closures. Although the magnitudes of MEMCs were not the objective of the current study, the likelihood of observing contractions depended strongly on eye closure effort/muscle activity and is consistent with the view that contractions were larger in magnitude and therefore easier to identify when eye closure effort level was greater. Additionally, the impedance changes used to identify MEMCs frequently had a time course very similar to that of the eye closure gesture (Fig. 3). Habituation to repeated eye closure gestures could not be assessed in the current study because identification of MEMCs relied on using the 25th percentile trace of the whole distribution of individual responses. Assessing MEMC habituation to repeated eye closure would require analysis at the trial level. Efforts to identify MEMCs at the trial level are currently underway.

MEMCs are more commonly elicited by eye closure than brief acoustic elicitors

Deiters et al. (2019) assessed rate of MEMCs in response to brief acoustic elicitors in the same participant group used for the current study. Acoustic elicitors included brief tones, white noise and recorded gunshots. Data processing was identical to the current study as was the method for identification of MEMCs. There were notable differences in the results for the two different elicitor conditions. Overall rates of MEMCs in response to brief acoustic elicitors were much lower than rates for voluntary eye closure. Specifically, rates ranged from as low as 20-30% to as high as 70-80% depending upon the characteristics of the acoustic elicitor. None of the rates for the acoustic elicitors approached the rates of MEMCs for even moderate levels of OOC activity. Additionally, unlike the current study, Deiters et al. (2019) found that rates of MEMCs for brief acoustic stimuli were significantly influenced by clinical middle ear testing results (i.e. tympanometric volume as well as clinical acoustic reflex magnitude and latency). The comparison of the current results with that of Deiters et al. (2019) clearly indicates that voluntary eye closure is a more robust elicitor of MEMCs than brief acoustic elicitors in participants with excellent hearing sensitivity and normal middle ear function, including the presence of clinical acoustic reflexes. What is less clear are the reasons for these differential rates of MEMCs. The acoustic reflex is elicited by sound mediated over the auditory pathways. The originating source of voluntary eye closure-mediated MEMCs is not so well-defined. It is possible that a copy of a cortically derived motor command used to generate eye closure is also sent to the middle ear muscles. Alternatively, the response may be mediated via proprioceptive responses arising from the periorbital movement. This issue would be best addressed by comparing relative timing of OOC muscle activity and middle ear impedance changes. Unfortunately, that requires a level of temporal precision not possible using the current study's methodology for detecting MEMCs. The pioneering study of Salomon and Starr (1963) and more recent work by Gruters et al. (2018), who examined eye saccade and middle ear changes, suggest that eye and ear motor activity most likely arise from a common motor command. The effector muscles are likely different for the two types of responses. The stapedius muscle response is generally agreed to be the primary effector mediating the acoustic reflex threshold (Borg et al., 1984). Salomon and Starr (1963) reported that eye closure-related periorbital muscle activity most frequently elicited tensor tympani activity and the stapedius muscle was not typically active during eye closure. The method used in this study, or any non-invasive study of middle ear muscle function, cannot discern relative contribution of tensor tympani and stapedius. However, the key point is that the acoustic reflex represents a single example of a wide range of complex sensorimotor responses that involve the middle ear muscles. This point has wide-ranging implications for studies that employ acoustic reflex measures and assume a protective function for them during noise exposure.

Implications for studies of MEMCs

The powerful influence of voluntary motor activity, such as eye closure, on middle ear muscle activity serves as a potential experimental confound for acoustic reflex studies. In such experiments, little, if any attention has focused on explicitly monitoring concomitant motor activity other than to instruct participants to sit quietly and keep movement to a minimum. Depending on the timing of concomitant motor activity such as eye closure, there is a potential to obscure the presence of actual acoustic reflexes or misinterpret as an acoustic reflex when one did not occur. This is a timely issue given a resurgence of interest in middle ear muscle function. Recent studies suggest that narrow-band acoustic elicitors of MEMCs may be a highly sensitive indicator for synaptopathy in mice (Valero, Hancock, & Liberman, 2016; Valero, Hancock, Maison, & Liberman, 2018). Assessing the viability of measures of MEMCs as a marker of synaptopathy in humans is currently underway (Mepani et al., 2019; Wojtczak, Beim, & Oxenham, 2017). Moving forward, sensitive and specific procedures for testing MEMCs will require careful control for influential variables. Motor activity, such as eye-related motor activity, needs to be monitored.

Implications for damage risk criteria for impulsive noise

The results of this study also have implications for the evaluation of damage risk criteria (DRC) for impulsive noise because some DRC assume a protective role for MEMCs. The most notable example is the Auditory Hazard Assessment Algorithm for Humans (AHAAH), which is an electro-acoustic model of sound transmission through the ear (Price & Kalb, 2018). Though the AHAAH model distinguishes between warned and unwarned exposures, both exposure types assume a role for MEMCs. For an unwarned exposure, the initial impulse will elicit maximum and sustained contraction of the middle ear muscles as a reflexive response. While the latency of the contraction will not provide protection for the initial impulse, a protective function for subsequent impulsive exposures is assumed. For a warned exposure, the model assumes MEMCs will reach maximum effect prior to the arrival of the initial impulse and will sustain a maximum contraction throughout an exposure series. Finally, regardless of whether the exposure is warned or unwarned, the model assumes MEMCs are present and identical for all exposed persons and do not vary across individuals or situational context (Price and Kalb, 2018). The results of the current study, along with the recent finding by Gruters et al. (2018) that suggest visual tracking elicits MEMCs, call into question this last assumption. As a DRC, the AHAAH model is designed to be applied across a wide range of operational environments. In such settings, the warfighter will be responding to a continuously varying set of environmental demands that in one moment may imply tight eye closure in anticipation of a blast, while in the next moment demand a visual scan of the field, followed by aiming and firing a weapon with both eyes open. The experimental results from the current study indicate that the level of eye closure has a significant impact on the likelihood of MEMCs, and that it is unlikely that MEMCs are present and identical across all warfighter operational contexts.

Finally, the current results also have implications for studies designed to compare different DRC for impulsive noise. Such studies are designed to incrementally increase, or “walk up” the number of impulsive noise exposures to participants while carefully monitoring hearing status (Johnson, 1997). The tight control of exposure level is critical to properly assess candidate DRC. Such studies must monitor and control for non-acoustic factors, such as eye-related motor activity, that can influence MEMCs and resultant exposure at the cochlea.

Study Limitations

This study inferred MEMCs based on RMS changes in the amplitude of a 50-ms broad-band click (.2-8 kHz) probe stimulus relative to levels drawn from a baseline period. Because RMS calculations do not account for the direction of impedance change, there may be cases where impedance decreased during the period of elicitor presentation, thus providing increased sound transmission into the cochlea. Additionally, although the ear canal recording was high-pass filtered at 0.1 kHz to remove noise unrelated to the probe stimulus, one cannot rule out the possibility that eye closure activity may have introduced some exogenous noise within the bandwidth of the probe signal, which could contribute to the RMS deviation. Studies are currently underway that employ a frequency-based analysis to determine if the RMS deviation is frequency-dependent.

The primary dependent measure of MEMCs was their occurrence rate. The occurrence rate was based on the binary judgement of 25th percentile traces of effort level-specific responses from three separate raters. While this procedure has the advantage of face validity, it does present some limitations to data interpretation. First, as with any procedure that relies on a subjective rating, rater error and/or bias is a possibility. The high degree of absolute agreement in these data (90-96%) suggests the likelihood that rater error and/or bias influenced the results is small. Second, the binary nature of the data did not capture the variation in MEMC magnitude, which is necessary for estimating functional impact. However, the purpose of this study was to determine the likelihood of any response. Having demonstrated that MEMCs are commonly elicited by voluntary eye closure, future efforts will examine the variability and functional impact of the MEMCs elicited in this way. The main purpose of the study was to determine the likelihood of a response. Third, the use of the 25th percentile trace for identification of MEMCs prevented estimating the occurrence rate at the individual trial level and assessing for response habituation. The development of algorithms for trial-level detection of MEMCs is underway.

The current results are based on a large group of participants with excellent hearing sensitivity and normal clinical middle ear function. Additionally, all participants exhibited robust acoustic reflexes to clinical acoustic reflex stimuli. It is not clear how well these results generalize to individuals with no clinical acoustic reflex responses, poorer hearing sensitivity, and/or middle ear anomalies.

Recommendations

Although voluntary eye closure is a strong elicitor of MEMCs, occurrence rates do not reach the threshold of being pervasive (95% confidence of 95% prevalence). Further, degree and type of eye motor activity is likely to be highly dependent upon the operational context. Therefore, DRC for impulsive noise should not include a protective role for eye closure related MEMCs. However, MEMC occurrence rates in response to eye closure are high enough that motor activity, such as eye closure, should be monitored as a complicating factor in auditory exposure studies.

Conclusions

In this laboratory study, MEMCs were measured in response to voluntary eye closure at three different effort levels in a large group of participants with excellent hearing sensitivity and normal middle ear function. Orbicularis oculi (OOC) EMG recordings were used to monitor and quantify eye closure effort level. MEMCs were commonly observed during EMG-verified eye closure gestures. MEMCs were more frequently observed for higher levels of eye closure effort. Multivariable analysis revealed that OOC EMG activity was a strong predictor of the likelihood of observing MEMCs. The occurrence rates of MEMCs at even moderate levels of eye muscle activity are much greater than are observed for impulse-like acoustic stimuli. MEMCs were nearly certain to occur at high levels of eye muscle activity. These results suggest that eye-related motor activity is a powerful elicitor of MEMCs that can serve as a confounding factor for acoustic reflex studies. These results also complicate the interpretation of impulsive noise DRC that assume MEMCs serve as a consistent, uniform protective factor.

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