Spectral and synchrony differences in auditory brainstem responses evoked by chirps of varying durations

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The chirp-evoked ABR has been termed a more synchronous response, referring to the fact that rising-frequency chirp stimuli theoretically compensate for temporal dispersions down the basilar membrane. This compensation is made possible by delaying the higher frequency content of the stimulus until the lower frequency traveling waves are closer to the cochlea apex. However, it is not yet clear how sensitive this temporal compensation is to variation in the delay interval. This study analyzed chirp- and click-evoked ABRs at low intensity, using a variety of tools in the time, frequency, and phase domains, to measure synchrony in the response. Additionally, this study also examined the relationship between chirp sweep rate and response synchrony by varying the delay between high- and low-frequency portions of chirp stimuli. The results suggest that the chirp-evoked ABRs in this study exhibited more synchrony than the click-evoked ABRs and that slight gender-based differences exist in the synchrony of chirp-evoked ABRs. The study concludes that a tailoring of chirp parameters to gender may be beneficial in pathologies that severely affect neural synchrony, but that such a customization may not be necessary in routine clinical applications. © 2010 Acoustical Society of America. [DOI: 10.1121/1.3483738]

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I. INTRODUCTION

To compensate for temporal dispersion down the cochlea, recent studies have suggested that ‘click’ stimuli be replaced by rising-frequency ‘chirp’ stimuli when acquiring the auditory brainstem response (ABR). Rising-frequency chirp stimuli compensate for temporal dispersion by delaying the higher frequency content of the stimulus until the lower frequency traveling waves are closer to the cochlea apex. This compensation theoretically results in simultaneous displacement maxima along the entire length of the basilar membrane within the inner ear, allowing all regions to contribute to the ABR. The result is an ABR waveform that is longer, can be recorded in less time, and potentially has more diagnostic power. 1

Improvements to the amplitude of the ABR waveform could infer that chirp-evoked responses exhibit more neural synchrony than click-evoked ABRs. However, measuring ABR amplitude alone is not necessarily a robust measure of neural synchrony. Larger ABR peak amplitudes may be the result of larger contributions from individual neurons, contributions from more neurons, and/or better neural synchrony. 2 A stimulus that causes synchronicity in the generators of wave V does not necessarily imply that the same stimulus also causes maximum synchronicity at the basilar membrane (BM) level. 3

In a previous study we demonstrated that, at high intensities, chirp stimuli in fact evoke less synchronous ABRs than click stimuli. 4 The reduction in synchrony was shown to decrease the magnitude spectra of the ABR at higher frequencies and to increase response ‘jitter’. We concluded that those chirp-evoked ABRs had larger wave V s due to increased neural recruitment, but that neural synchrony was reduced due to local phase interactions between cochlea filters. It was also suggested that a further reduction in overall neural synchrony occurred due to the broadening of BM excitation with increased intensity, consistent with previous reports. 1, 5

The purpose of the present study is to extend these findings and examine chirp-evoked neural synchrony at a lower intensity. We also investigate the sensitivity of the chirp-evoked response to variations in stimulus duration and frequency sweep rate, effectively altering the degree of temporal compensation offered by chirp stimuli. In doing so we can evaluate the effect of varying sweep rate on neural synchrony.

Other studies of chirp-evoked responses to varying sweep rates have inferred the effect on neural synchrony from latency measurements. 7 In the present study we extend this analysis to the magnitude and phase spectra. It is known that ABRs with greater jitter, such as those acquired using low frequency tone bursts, have broader peaks in the averaged ABR. 8 Conversely, when neural synchrony is high, the averaged ABR has peaks that are narrower and higher—more nearly approximating the response of individual neurons. 9 Narrower peaks have higher frequency components, and therefore the fast Fourier transform (FFT) magnitudes of the higher frequencies may reflect the level of neural synchrony.
An additional technique we will use to determine the level of neural synchrony is the ‘Synchrony Measure’ (SM), originally described by Fridman et al.\textsuperscript{10} and further experimented with by Chen et al.\textsuperscript{11} Sakai et al.\textsuperscript{12} The Synchrony Measure represents the degree of reproducibility for group averages of the evoked response by examining the phase variance in particular Fourier components.\textsuperscript{10} Smaller phase averages of the evoked response by examining the phase Measure represents the degree of reproducibility for group variances indicate greater synchronization in the origin of the ABR\textsuperscript{15} and that the particular Fourier component is time-locked with stimulus onset. Components that are time-locked with stimulus onset are more likely to reflect auditory-evoked potentials rather than background physiological noise.\textsuperscript{10}

If we apply multiple measures of synchrony to chirp-evoked ABRs we can provide a good indication of whether chirp stimuli evoke more synchronous responses. Furthermore, by varying the duration of the chirp we may be able to examine the relationship between chirp sweep rate and response synchrony. The aims of this study were therefore as follows: (1) to use wave V amplitude, wave V latency variance, ABR frequency content (as shown by the FFT) and the SM to investigate differences in neural synchrony between click-evoked ABRs and ABRs evoked by chirps of varying durations; and (2) to discuss factors contributing to changes in synchrony and the FFT. The experiments completed for this study are described below.

II. METHODS

A. Subjects

Thirty two normal hearing subjects (12 males and 20 females) participated in the experiments. The age distribution was 25.7 ± 4.3 years for males and 28.4 ± 5.2 years for females (28.4 ± 5.0 years overall). Participants had audiometric thresholds of 15 dB HL or better for octave frequencies between 250 and 6000 Hz and no self-reported otological concerns. Anyone not meeting these criteria was excluded. All subjects were volunteers and written consent was obtained from each participant prior to commencement of the study.

B. Equipment

We used a low-noise CED 1902 bio-amplifier and a CED Power1401 data acquisition system, both from Cambridge Electronic Design (CED) to acquire ABR waveforms. The 1902 bio-amplifier contained onboard filters and could provide up to 120 dB of gain. We presented auditory stimuli to test subjects via high-frequency ER-2 insert earphones, driven by a Tucker-Davis HB7 headphone buffer. We used a custom-coded Mathworks Matlab\textsuperscript{TM} interface to drive the data acquisition and analysis.

The relationship between the custom built ABR unit’s variable gain and output dB SPL for a 1000 Hz tone was determined using an Aurical system (EN60645–1, –2 type: 2, A; monitor version 1.00; program version 2.40; DSP program version 2.40) running Aurical REMUL module version 2.50, and this system’s 2 cc coupler. All stimuli produced by the unit were calibrated before each ABR waveform acquisition using a loopback method whereby the output to the headphone amplifier was simultaneously monitored by the analog-to-digital converter. The gain required to achieve the required dB peSPL was determined by successive approximation, using the previously calibrated 1000 Hz tone value for dB SPL.

C. Stimuli

We included four rising-frequency sweeps (chirps) in this study, the first of which was the ‘ABR-based’ chirp, ‘A-Chirp.’\textsuperscript{3} For more details on the construction of the A-Chirp, we refer the reader to the original paper.\textsuperscript{3} However, as an overview, the A-Chirp is intensity-specific and is based on previously published wave V latencies from tone bursts at various intensities.\textsuperscript{14,15}

Neely et al.\textsuperscript{12} assumed that ABR latency was comprised of a fixed neural component, \(a\), and a frequency- and level-dependent basilar membrane (BM) delay, \(\tau_{BM}\). They estimated this BM group delay by:

\[
\tau_{BM}(i, f) = bc^{-i}f^{-d},
\]

where \(i\) represents the tone-burst intensity (in dB SPL divided by 100), \(f\) represents tone-burst frequency (in kHz), and \(b\) and \(c\) and \(d\) are constants with the values \(b=12.9\) ms, \(c=5.0\), and \(d=0.413\), according to a power-law fit.\textsuperscript{15} Applying the transform \(\tau_{BM}(i, f) \rightarrow t_0-t \) to invert the time relationship, the A-Chirp compensates for the BM group delay by increasing the instantaneous frequency, \(f(t)\), at a rate commensurate with the above power-law:

\[
f(t) = (t_0-t)^{(-1/d)}(bc^{-i})^{(1/d)}.
\]

The duration of the A-Chirp can be altered by adjusting either the \(b\) or \(c\) constants, the latter of which is preferable to adjust as it is a unitless constant. The value of \(c\) in the A-Chirp at 25 dB SL is fitted to traveling wave delay data obtained by Gorga et al.,\textsuperscript{14} from a population of unspecified gender distribution. Previous investigations into traveling wave delays have found gender differences in the order of 10%.\textsuperscript{16} Therefore in this study, to investigate possible differences in the chirp-evoked response between genders and the effect on synchrony of varying chirp duration, we calculated three additional ‘custom’ chirps with durations that deviated approximately 15% from the A-Chirp. At an intensity of 25 dB SL, with an estimated reference equivalent sound pressure level (RETSPL) of 40 dB peSPL,\textsuperscript{3} the A-Chirp is 9.98 ms long. The custom chirps had \(c\) values of 6.40, 4.98, and 4.02—giving durations of 8.5, 10, and 11.5 ms respectively. The frequency sweep for all these chirps ranged from 0.1 to 10 kHz. We adjusted the temporal envelope of the chirps to weight all frequencies equally in the power spectrum, as was prescribed for ‘flat-chirps’ by Dau et al.\textsuperscript{1}

We also included a 100 \(\mu\)s click in this study, commonly used in audiology, for subjective comparisons to the chirp stimuli. The intensity of chirp stimuli was 65 dB peSPL. The clicks were presented at 72.2 dB peSPL, so all five stimuli had the same Sensation Level (SL), 25 dB SL, according to Fobel and Dau.\textsuperscript{3} The 7.2 dB difference between click and chirp sound pressure level ‘reflects temporal integration of signal energy involved in behavioral threshold

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measures that probably occurs at more central stages of auditory processing and is most likely not reflected in ABR—a theory supported by Nabelek.

The pre-transducer electrical time courses of our stimuli are shown in Fig. 1 (arbitrary vertical scale). Additionally, we recorded the spectra of our stimuli using a 4152 artificial ear and a Brüel & Kjær 2250 hand-held analyzer (Fig. 2).

D. Recording

Testing took place in a sound-treated and electrically-shielded room. The test subjects lay down on a clinic bed and the lights were turned out to assist the subject to sleep if possible. Electro-encephalic activity was recorded from the scalp via disposable silver/silver chloride electrodes. The electrodes were placed in a vertical montage such that one electrode was attached to the forehead, one on the nape of the neck (reference), and one below the middle of the left clavicle (ground). We used the vertical electrode montage as it does not change with test ear, and because of previous reports that it highlights wave V. Inter-electrode impedance was maintained below 5 kΩ, and in most cases was below 2 kΩ.

Stimuli were interleaved between ears to allow responses from both ears to be recorded almost concurrently. The delay between stimuli presented to opposite ears was 22.6 ms, whereas the delay between stimuli presented to the same ear was 45.2 ms, giving an effective stimulus repetition rate of 22.1 stimuli per second at each ear. The stimuli were presented with alternating polarity—to minimize stimulus artifacts and the cochlear microphonic—and were digitally generated with a reconstruction frequency of 263.2 kHz. Responses were amplified (89.5 dB) and filtered (100 Hz to 5000 Hz Butterworth band-pass, 12 dB/oct roll-off) before being digitized by the Power 1401 ADC unit (set to 32.9 kHz, 16 bit resolution).

The order of presentation was randomized and responses were recorded in bursts of 100 epochs per stimulus. The varying N values in the present study reflect the fact that the responses to some stimuli reached the SNR threshold consistently faster than for other stimuli. ABR acquisitions took approximately 60 min per test subject.

E. Signal processing

Recorded data was analyzed post-hoc using a custom-coded Mathworks Matlab™ interface. Artifact rejection was performed, such that epochs with data that exceeded a threshold of 19 μV within a window of interest were excluded from further analysis. Following this, the raw data was filtered using a 100 Hz–3 kHz software band pass filter.

The window of interest for artifact rejection was 0 to 12 ms following stimulus offset, accounting for the 0.86 ms tubing delay of the ER-2 earphones. Rejecting epochs with data that exceed a threshold has been shown to be the most straight-forward and effective in artifact rejection.

We sought to keep the signal-to-noise ratio (SNR) of each ABR at a similar level across all stimuli and test subjects. To achieve this, we made use of the single point variance statistic, $F_{SP}$. When constructing the ensemble average for each ABR, we only summed as many epochs as were required to reach a $F_{SP}$ value of 3.1. This criterion infers that the probability of an ABR being present is at least 99%. In this manner, all ensemble averages had a similar SNR; 1.6 dB, using the approximation,

$$SNR = 10 \log_{10}(\sqrt{(F_{SP} - 1)}).$$

The analysis window used when calculating the $F_{SP}$ statistic was the same as for the artifact rejection, and the ‘single point’ for each $F_{SP}$ calculation was automatically chosen to be the strongest peak in the overall ensemble average (within that same analysis window). The window parameters were appropriate to resolve low frequency noise contributions to the ABR, avoid any contribution from stimulus artifact, and include the entire ABR signal.

The characteristics of physiological noise are such that the noise falls in and out of phase with the acquisition window and stimulation rate. This “noise memory bias” can cause oscillations in SNR estimations. For this reason, we determined the number of epochs required to reach our $F_{SP}$ threshold from a fitted slope of the increase in $F_{SP}$ over time. A robust linear regression function iteratively removed out-
F. Analysis of wave V amplitude and latency

We determined the location of the peak of wave V visually, and used a graphical interface to measure the amplitude from the peak of wave V to the next major trough.

The click-evoked wave V latencies are reported here relative to stimulus onset since the click-evoked ABR is suggested to be an onset-drive response. However, in the case of the chirps, the stimuli have been designed to give maximum basilar membrane (BM) deflection at stimulus offset. Therefore, we subtracted the stimulus duration from these ABRs to give an ‘offset latency’ relative to stimulus offset. This reflects the notion that displacement maxima on the BM should occur in all frequency channels at the same time, and thus, the latencies for the chirps and the click should be similar if expressed relative to stimulus offset.

We tested per-stimulus differences in means across wave V latencies and amplitudes. We also analyzed latency variance using Bartlett’s test, which is designed to test for equality of variances across groups against the alternative that variances are unequal for at least two groups. The variable under test was the inter-subject variance of wave V latency across stimuli groups.

G. FFT spectrum analyses

To minimize artifacts when calculating the FFT we applied a Tukey cosine-tapered window, aligned relative to the location of the wave V peak. The window began 5.6 ms prior to wave V and ended 3.6 ms after; including as much of the ABR as was possible for the longer chirps without risk of exceeding the interleaving period. For all responses this encompassed waves I to V, the large trough following wave V, and a portion of wave VI. The window length was 301 samples and had a ratio of tapered sections to constant sections of 0.15. Pre-FFT windowing is important to reduce high-frequency artifacts that appear when start or end points are nonzero.

We zero-padded the data to extend the duration to 2048 samples before calculating the FFT. Zero-padding the ABR in this way increased the resolution of the FFT to approximately 16 Hz.

We calculated the FFT across frequencies for each subject and analyzed the locations and magnitudes of the three major peaks that appeared in the FFT spectrum. Overall FFT spectra were generated by averaging the FFT for each test subject, as opposed to calculating the FFT of the Grand Average, as individual FFT spectra were required for statistical testing.

H. Component synchrony measures (CSM) and synchrony measure (SM)

The first step to obtaining the SM is to calculate Component Synchrony Measures (CSMs) at each frequency. The CSM is a measure of phase variance of a given component frequency over a number of sub-averages of the evoked response. The CSM ranges from 0 (for large variances and apparent disynchrony) to 1 (for responses that are perfectly synchronized).

To calculate the CSMs for each evoked response we followed the methods of Fridman et al., separating all available data, post artifact-rejection, into N sub-averages of 100 repetitions. Sub-averages with less than 100 repetitions were excluded. After removing any DC offset from each sub-average, we applied the same zero-padding and Tukey window as for the previous FFT spectrum analyses. We calculated the FFT of each windowed sub-average and measured the phase of each component frequency. The variances of all phase measurements were calculated as:

$$\text{var}(\varphi(m)) = 1 - \left( \frac{1}{N} \sum_{n=1}^{N} \cos \varphi(n) \right)^2 - \left( \frac{1}{N} \sum_{n=1}^{N} \sin \varphi(n) \right)^2,$$

where $N$ is the number of sub-averages, and $\varphi(m)$ is the phase of the $m$-th Fourier component of the $i$-th sub-average. The CSM was then taken as:

$$\text{CSM}(m) = 1 - \text{var} \varphi(m).$$

We report only three representative CSMs, the frequencies of which were chosen to correspond to the three major peaks of the FFT. CSM measurements above 3 kHz can be considered noise, as they are in the stop-band of our low pass filter, and are not relevant to our comparisons. Therefore, the Synchrony Measure (SM) was calculated to be the average of CSMs from every available frequency up to the 3 kHz limit of our low pass filter:

$$\text{SM} = \frac{1}{M} \sum_{m=1}^{M} \text{CSM}(m),$$

where $M$ is the number of Fourier components included in the calculation.

If the SM calculated for test data significantly exceeds the expected SM value for noise, one can assume the presence of a signal. For click-evoked ABR, Fridman et al. prescribed a SM threshold of 0.22, at which the probability that an evoked response is present is very high (99.9%).

I. Statistical testing

Statistical comparisons in this study were performed with non-parametric tests, since not all data sets satisfied the normality and homoscedasticity assumptions required for the use of parametric analyses. We preferred the Friedman test, also known as a two-way analysis of variance by ranks, as it is appropriate for analyzing three or more repeated measurements of ordinal data. To compensate for the exclusion of
ensemble averages that did not reach our $F_{SP}$ threshold we utilized an extension of the Friedman test that allowed for unequal numbers of observations per test subject.\textsuperscript{38}

Whenever the Friedman result was significant (i.e., the null hypothesis was rejected) we followed with multiple Mann-Whitney Ranked Sum analyses on samples evoked by each of the stimuli. We included the Bonferroni correction for multiple comparisons but, to maintain statistical power, limited our planned comparisons to only those that directly involved previously reported stimuli; the click and the A-Chirp. Our comparisons were therefore; each stimuli to the click, and each stimuli to the A-Chirp. This gave 7 permutations and a Bonferroni corrected significance level of 0.05/$7=0.007$.\textsuperscript{32}

In all our measurements and comparisons, we report the result for the overall population and then separate the results for each gender to determine if the observed effects were specific to physiological differences between genders, such as cochlear length.\textsuperscript{39}

\section*{III. RESULTS}

\subsection*{A. Grand averages}

In the Grand Averages (Fig. 3) an ordinate offset has been added to the male and female traces for clarity. The Grand Averages for the male subpopulation (solid curve) had consistently later wave V latencies than for the female subpopulation (dotted curve). Wave Vs for chirp stimuli are clearly larger than for click stimuli. One feature that stands out in the female subpopulation is that wave IIIs for the A-Chirp, the 10 ms chirp, and the 11.5 ms chirp are not followed by a clearly defined trough. In the male subpopulation waves I and III for chirp stimuli are of comparable amplitude to those evoked by the clicks.

\subsection*{B. Time domain analyses}

The Friedman analysis of wave V amplitudes confirmed that the effect of stimulus choice on wave V amplitude (Fig. 4, top) was very highly significant ($p<0.001$). Pair-wise Mann-Whitney comparisons found that all the chirps evoked significantly ($p<0.05$) larger amplitudes than the clicks for males, females, and the overall population—consistent with previous reports.\textsuperscript{1,3,5,40}

The effect of stimulus choice on offset wave V latencies (Fig. 4, bottom), according to the Friedman analysis, was very highly significant ($p<0.001$) for all populations. Subsequent Mann-Whitney comparisons confirmed that offset wave V latencies for the chirps were significantly smaller ($p<0.05$) than those of the clicks. When comparing the A-Chirp to the other chirps, the Mann-Whitney comparison suggested that the longer chirp (11.5 ms) had significantly smaller offset latencies. Additionally, for females and the overall population, the shorter chirp (8.5 ms) had significantly longer offset latencies (than A-Chirps) when data from both ears were pooled together.

For Bartlett’s test of difference in wave V latency variances, we rejected the null hypothesis that all stimuli groups had equal variance, at the very highly significant level ($p<0.001$) for the overall population and the female population. A subsequent pair-wise F-test on data from these populations found that the A-Chirp, the 10 ms chirp, and the 11.5 ms chirp had significantly greater ($p<0.05$) wave V latency variance than the clicks, in the case of both ears pooled. The result was similar for left and right ears treated alone, except comparisons involving the A-Chirps and 10 ms chirps did not always reach significance. Wave Vs evoked by the longer 11.5 ms chirp also had significantly greater latency variance than the A-Chirp, for the case of both ears combined in the female and overall populations. There were no significant results for Bartlett’s test in the male subpopulation.

\subsection*{C. Frequency domain analyses}

In the FFT (Fig. 5), the three predominant peaks (at the center of each ‘band’) had frequencies and relative magnitudes consistent with previous reports.\textsuperscript{35,41,42} The CSM versus Component Frequency plots (Fig. 6) have peaks at similar frequencies as for the FFT spectra, but the magnitude of each peak now reflects the level of synchrony at that particu-
lar frequency. We also observed an additional fourth peak in the CSM plots, around 830 Hz, which was inconspicuous in the FFT spectra.

FFT magnitudes in band A were significantly larger for all of the chirp stimuli, when compared to the click. In band B, the FFT magnitudes for the chirps were significantly larger than for the clicks in all comparisons. When comparing other chirps to the A-Chirp, the 11.5 ms chirp had significantly smaller FFT magnitudes in band B for the female population when data for both ears were combined. The FFT magnitudes in band C were larger for the A-Chirp than for the clicks in all subpopulations when data for both ears were combined. Additionally, in the overall population, the 10 ms and 11.5 ms chirps evoked significantly larger FFT magnitudes in band C than the clicks, when data for both ears were combined.

In the FFT analyses the central frequencies of band A were significantly greater \( p < 0.05 \) for the 11.5 ms chirps than for the clicks in all comparisons. When comparing other chirps to the A-Chirp, the 11.5 ms chirp had significantly smaller FFT magnitudes in band B for the female population when data for both ears were combined. The FFT magnitudes in band C were larger for the A-Chirp than for the clicks in all subpopulations when data for both ears were combined. Additionally, in the overall population, the 10 ms and 11.5 ms chirps evoked significantly larger FFT magnitudes in band C than the clicks, when data for both ears were combined.

In band B there were significant differences again in central frequency for the male and overall populations. The central frequencies for the A-Chirp and the 11.5 ms chirp were significantly larger than for the click in the male population for the right ear, and also when both ears were combined. This carried over to the overall population, where the central frequencies of band B for the A-Chirp, 8.5 ms chirp, and 11.5 ms chirps were significantly higher in the right ear than for the clicks. The A-Chirp and 11.5 ms chirp also had significantly higher band B frequencies than the clicks when data for both ears were combined. The Friedman statistic indicated that there were no significant differences in the left ears of these populations.

In band C, the central frequencies for ABRs evoked by the 10 ms chirp were significantly higher than for the clicks, in the male subpopulation, as well as in the overall population. Again, however, this result was not significant in left ears, and the difference in means tended to be less than the 16 Hz resolution of our FFT calculation.

For Component Synchrony Measures in band A, the A-Chirps had significantly more synchrony than the clicks for males when data for both ears were combined. Similarly, the A-Chirp and the 10 ms chirp had significantly more synchrony than the click for females and the overall population, when data for both ears were combined. In band B, the CSMs for all chirps were significantly larger than clicks in the male subpopulation when both ears were combined, except for the shortest (8.5 ms) chirp. Conversely, the CSMs for all the chirps in the female subpopulation were significantly larger than for clicks except the longest (11.5 ms) chirp (both ears combined). For the females the apparently reduced synchrony in band B of the longest (11.5 ms) chirp was also significantly smaller than the A-Chirp. The results were similar in band C; all chirps except the shortest (8.5 ms) in the male subpopulation had significantly larger CSMs.
than for clicks, and the 8.5 ms and 10 ms chirps had significantly larger CSMs than the clicks in the female subpopulation. None of the custom chirps had significantly different CSMs to the A-Chirp in band C.

After analyzing the CSMs, they were summated to give the overall SM. We found the chirp SMs for males were significantly greater than the click for all chirps except the shortest (8.5 ms) chirp, for both ears combined. Conversely, the chirp SMs for females were significantly greater than the click for all chirps except the longest (11.5 ms) chirp for both ears combined.

IV. DISCUSSION
A. Synchrony of chirp-evoked ABRs

In this study we analyzed chirp- and click-evoked ABRs at low intensity, using a variety of tools to measure synchrony in the response. Contrary to our previous study at higher intensities, the present results strongly suggest that the chirp-evoked ABRs in this study exhibited more synchrony than the click-evoked ABRs. This was evidenced by the larger wave V amplitudes, larger FFT magnitudes, larger CSMs, and larger overall SMs.

We also examined the relationship between chirp sweep rate and response synchrony by varying the duration of the chirp. It was expected that differences in cochlea dimensions between test subjects would cause a given chirp length to exhibit better or worse synchrony, dependent on whether compensation for BM temporal dispersion was enhanced or diminished. The results obtained when varying the length of the chirps suggest that differences in resulting synchrony, dependent on chirp duration, exist between males and fe-
males. These observations may be similar to what a clinician could expect if a given chirp was (or was not) suited to a test subject.

1. Observations in the time domain

Consistent with previous reports, we found that rising-frequency chirps evoke significantly larger wave V amplitudes than click stimuli. This was also consistent with the result that the FFT magnitudes of the lower frequency band A for all chirp stimuli were significantly larger than for clicks, as band A is known to contribute the most to wave V. However, waves I and III in the Grand Averages for the chirps and the clicks appeared visually to have comparable amplitudes, suggesting that the enhancements offered by the chirp relate predominantly to wave V. The effect on individual ABR waves has been modeled previously by Dau, who determined that chirp-evoked contributions to wave V are integrated across a relatively large temporal window and are mainly enhanced by net synchrony across BM channels rather than phase synchrony within channels. Conversely, enhancements to waves I and III require synchrony of neural discharges within individual channels as these waves are integrated over a much shorter window.

The broadening of wave III for females in the Grand Averages of the longer chirps could indicate a comparatively larger III-V inter-peak latency (IPL) variance across that subpopulation, commensurate with the significantly greater wave V latency variances observed in the contributing ABRs. The IPL variance is known to increase with alternating stimuli, but is also likely to be exaggerated with the longer chirps if the chirp duration exceeds the temporal compensation required for synchronous BM displacement. Particular also to the female subpopulation, inter-subject variance in IPLs would be expected if our test subjects were at varying stages of their menstrual cycle.

2. Observations in the frequency domain

We observed that ABRs evoked by chirps had significantly larger FFT magnitudes in bands A and B than for the clicks. Spectral power in band A has a major contribution from spontaneous brain activity, EMG and movement artifacts. However, the significantly larger band A magnitudes we observed for chirps are more likely linked to their significantly greater wave V amplitudes, since components in band A are also known to contribute to the compositions of waves IV and V.

In band C the A-Chirp had larger FFT magnitudes than click-evoked ABRs, indicating an increase in information pertaining to waves I through V. The significantly larger FFT magnitudes for the A-Chirp-evoked ABRs could imply that the A-Chirps recruited more neurons into the response, or that the response was more synchronous. The latter hypothesis was confirmed with our CSM results; in all bands the A-Chirp-evoked responses had significantly larger CSM values than for the clicks, implying increased synchrony.

In contrast with our previous study at higher intensities, the chirp-evoked ABRs in the present study tended to have higher CSM and SM values than the click stimuli. This difference between studies can be explained by the fact that, at high intensities, the tuning of each cochlear filter becomes broader, and the mechanical interactions between coupled sections of the BM become complex and nonlinear. Transfer functions of the cochlear filters exhibit phase dispersion in the form of frequency ‘glides,’ which have been reported in BM vibrations and auditory nerve (AN) firing rates. The reduction in stimulus intensity between our two studies appears to have lessened the impact of this phase dispersion on neural synchrony within the chirp-evoked responses.

3. Synchrony variation with chirp duration

For the female subpopulation, we observed that ABRs evoked by the shorter chirps (10 ms or less) tended to have higher FFT magnitudes and synchrony measures than for the clicks. The longest chirp (11.5 ms) had synchrony measures and FFT magnitudes in the female subpopulation that were statistically no better than for clicks. These observations suggest that shorter chirps compensate more exactly for traveling wave delays down female cochleae, as female cochleae have shorter traveling wave delays than males, corresponding to shorter cochleae and larger stiffness gradients.

Conversely, for the male subpopulation, the shortest chirp (8.5 ms) performed worse than the other chirp lengths and did not display any significant differences from click stimuli in our analyses of FFT magnitudes and synchrony measures. Fobel and Dau, who originally described the A-Chirp, proposed that the duration of chirp stimuli should be adjusted to match intensity-dependent dispersions down the BM. Their A-Chirp was calibrated to the hearing thresholds of 7 males and 2 females, with no provision made for differences between genders. There is no evidence in the present study that the A-Chirp is inferior to the other chirp lengths, but we do find it noteworthy that significant differences between click- and chirp-evoked responses were only observed in the male subpopulation for chirps equal or longer in length than the A-Chirp. Our results for overall SM suggest that longer chirps evoke more synchronous ABRs.

FIG. 7. Synchrony Measures (both ears combined) for the overall population, male subpopulation, and female subpopulation.
than clicks for males, and that shorter chirps evoke more synchronous ABRs than clicks for females.

Regarding evoked wave V amplitudes, the effect of varying chirp duration was not significant. All chirps in this study evoked larger ABRs than the clicks, but none of the ABRs evoked by custom chirps had significantly different amplitudes from the population-based A-Chirp. This lack of sensitivity of evoked amplitudes to chirp duration can be explained by redundancy in the auditory system—each higher order neuron is activated by the endings of many lower order neurons, and thus only a small number of activated lower order neurons are required to cause near maximal excitation.\(^52,53\) However, an increase in the number of contributing lower order neurons is still desirable since variability (jitter) in the firing rate of higher order neurons is reduced when many lower order neurons contribute.\(^54\) For example, neural synchronization in the cochlear nucleus (AVCN) is dependent on the convergence of inputs from two or more auditory nerve (AN) fibers and is enhanced by coincident input spikes.\(^55\)

In pathologies that severely affect neural synchrony, the effect of varying chirp duration may become more significant. For example, the demyelination in multiple sclerosis compromises the synchrony of impulse firing in groups of affected axons.\(^56,57\) It is conceivable that a gender-specific customization of chirp parameters could improve synchrony and increase the chances of recording a usable ABR. This customization may be as simple as correlating chirp duration with head size measurements, as these dimensions have been previously shown to correlate with ABR wave latencies.\(^58\) Customization may also be important when performing ‘stacked-ABR’ to detect neurinomas,\(^59\) since that particular application assumes a correlation between derived-response latencies and site of BM activation.

### B. Chirp ‘goodness-of-fit’

The observation that the offset wave V latencies were significantly \((p<0.05)\) smaller for chirps than clicks suggest that the chirps in our study were either evoking a response from more proximal (high frequency) sections of the cochlea than the clicks, or that the response was evoked prior to the chirp offset. Although chirp stimuli were expected to elicit simultaneous displacement of all portions of the basilar membrane, we observed that increasing the chirp duration led to a decrease in offset wave V latencies. In particular, the longer (11.5 ms) chirp had offset latencies that were significantly shorter \((p<0.05)\) than for the A-Chirp, suggesting that it evoked a response prior to stimulus cessation. If this is the case, the longer chirps would have evoked a response from predominantly lower characteristic frequency (CF) portions of the BM as the higher frequency components would have arrived at their CF place too late to contribute to the response. An alternative hypothesis is that the low frequency components of the chirp stimuli stimulated high CF neurons, but previous reports suggest that this effect is not substantial at the relatively low intensity of 25 dB SL used in this study.\(^60\)

In the female subpopulation, the 8.5 ms chirp had offset latencies that were significantly longer \((p<0.05)\) than the A-Chirp, and closer to those of the clicks. This suggests that the shorter chirp evoked a response in females that was initiated in similar frequency portions to the click stimuli. Therefore, the sweep rate of the 8.5 ms chirp may have compensated more exactly for frequency-specific BM group delays in the female subpopulation than the longer chirps, and hence could be assumed to be a ‘better fit’. The results for Bartlett’s test for equal variance also suggest that the 8.5 ms chirp is well suited to female cochleae at this intensity, since there were significantly smaller \((p<0.05)\) variances in wave V location for that chirp in that subpopulation. Conversely, the 11.5 ms chirp seems the least suited to female cochleae, since responses from that stimulus had significantly greater wave V variance than both the click and the A-Chirp. Chirp stimuli are designed to compensate for traveling wave delays down the cochlea partition, but if the compensation is inexact, we expect that dissynchronous excitation of the BM would lead to increased jitter and a larger variance in wave V latency. This is especially the case if the chirp duration is too long, as the response will come from predominantly lower CF portions\(^7\) where the neural synchrony is known to be less than at high CFs.\(^61\)

An additional contribution to neural disynchrony that may be relevant to the chirp-evoked response is ‘two-tone suppression’. This refers to the inhibition of a nerve fiber’s response to a CF tone by an off-frequency tone that lies outside its excitatory area, and is a feature of normal cochlear function that presumably serves to sharpen frequency resolution.\(^54\) Responses to frequency components of the chirp that align to their CF first may suppress those frequency components that do not reach their CF until slightly later (or vice-versa). Previous studies of chirp-evoked responses have found that, while certain sweep rates minimize the temporal delay between unit responses, slower sweep rates only partially cancel travel time; initiating a response from the lower frequency portions before the higher frequency portions.\(^7\) At higher intensities the effect is reversed; low frequency components of the chirp will stimulate high CF neurons, inhibiting their response when the high frequency components of the chirp eventually arrive.\(^9,17\)

There is a strong suggestion in our results for the overall population that variances in wave V location for chirp-evoked ABRs are significantly greater than for clicks. However, the absence of significant differences in the male subpopulation does not support this. Human cochleae lengths are highly variable among populations,\(^62\) and the length is significantly longer in males than females.\(^63\) This leads to significantly longer delays in male cochleae for lower frequencies to reach their characteristic place, in comparison to female cochleae,\(^16\) and a subsequent decrease in neural synchrony.\(^39\) Therefore, consistent with the present study, wave V latencies are later for males and have greater variability than for females.\(^64\) Previous researchers have suggested also that latency variances for chirp-evoked ABR are exaggerated due to inter-subject cochlea differences in travel time to the lower frequency portions.\(^5\)
C. Additional findings

1. FFT frequency shifts

We observed significant upward shifts in central frequencies for bands A and B in the male subpopulation when using the 11.5 ms chirps. These ABRs had central FFT frequencies that were significantly higher than for 100 μs clicks and A-Chirps in band A, and significantly higher than for 100 μs clicks in band B. Head trauma, coma, epilepsy, multiple sclerosis, and neurinomas of the acoustic nerve are all conditions that are known to affect the location of each of the major FFT peaks, but these conditions were absent in our healthy subjects. The position of the recording electrode is also known to shift the frequencies of the FFT peaks, as is stimulus polarity. Some researchers have suggested that decreasing stimulus intensity decreases the frequencies of the spectral peaks, but other studies have not had sufficient FFT resolution to confirm this. These effects do not adequately explain the results of the current study, since electrode position was maintained throughout the acquisition and all stimuli were calibrated to equal dB SL. Pre-FFT windowing can affect the location of the frequency peaks, but again this does not explain why some ABRs were affected more than others. Finally, the post auricular muscle reflex (PAMR) has also been reported to shift peaks in the FFT, dependent on state of relaxation, but the vertical electrode montage used in the current study is unlikely to have recorded the PAMR. We can conclude that the observed shifts in peak FFT frequencies were not systematic, and are therefore more likely to be physiologically based.

One possible physiological contribution to the observed frequency shifts could come from the manner in which responses to varying frequency stimuli are neurally encoded. Auditory nerve fibers typically communicate two different codes for the frequency of sound to the central auditory nervous system: a place code as a result of the BM separating sounds according to their frequencies and a temporal code that reflects the frequency of vibration of each individual point along the BM. Temporal coding means that discharges of individual nerve fibers are phase-locked to the frequency of vibration of individual segments of the basilar membrane. A shift in the frequency of peaks in the FFT could reflect a shift in phase interactions between BM segments, and hence a shift in the firing pattern of nerve fibers carrying temporal coding. Computer-models of basilar membrane motion (BMM) and neural activity patterns (NAPs) show that the chirp aligns the point of maximal vibration across the frequency channels of the basilar membrane, but that the chirp also causes a ringing of the response within BM channels. This ringing is exaggerated particularly in the low frequency channels below 1000 Hz and conceivably has a phase spectra dependent on chirp sweep rate.

The frequency following response (FFR) has also been reported to affect the location of peaks in the magnitude spectrum. If there is a contribution from FFRs in our chirp-evoked responses then an upwards shift in FFT frequencies is compatible with the augmented duration of low frequencies in the 11.5 ms long chirp as FFRs to low-frequencies mainly arise from neural activities generated in the mid- and high-frequency channels.

2. Additional peak at 830 Hz

Synchrony measures proved more sensitive than magnitude, in that an additional CSM peak around 830 Hz (between band B & C, Fig. 6) was observed that was not immediately discernible in the magnitude spectra—presumably because this magnitude peak was below the residual background noise. Phase information, as in the CSM, has been shown previously to be a better predictor of auditory function than amplitude information. Some researchers have suggested that the presence of more than three main FFT peaks is indicative of a pathology such as epilepsy or neurinoma of the acoustic nerve. Moller also suggested that the splitting of FFT peaks is due to phase-locked synchronization between nerve bundles as a result of myelin injury, as in multiple sclerosis. Given that our study included only healthy ears, we propose that the ‘additional’ peak around 830 Hz contains information pertaining to normal auditory function. Potential sources for this component include the generators of peaks I to V, since these peaks are spaced around 1.2 ms apart (the inverse of 830 Hz). Further work is needed to identify the source of these synchronous neural discharges.

3. Interaural differences

There were occasional differences between ears observed in this study, which we confirmed were significant using the Friedman statistic. We would rule these differences out as not being functionally significant—particularly given the large variance in amplitudes among our populations. However, in every instance of an observed interaural difference, the right ear was found to have an advantage over the left ear. Significant interaural differences in wave V latencies tended toward the right ear having shorter wave V latencies than the left, although the differences were within a 0.3 ms criterion previously defined as ‘normal.’ Previous studies have also reported a right-ear bias in the evoked amplitudes and FFT magnitudes, and a tendency for the right ear to have shorter latencies. This bias has also been observed in otocochlear emissions. It has been suggested that asymmetric activation of the medial olivocochlear system (MOC), which modulates the cochlear amplifier, is responsible for this phenomenon. Significant frequency shifts in the power spectrum that we observed in our results occurred mostly in right ears, and may feasibly be linked to increased activity in the MOC.

V. CONCLUSIONS

Collectively, our measures for neural synchrony (wave V latency variance, FFT analyses, and synchrony measures) suggested that, at the intensities of the current study, chirp stimuli evoke more synchronous responses than click stimuli. There was also strong suggestion of differences between genders in the required compensation of frequency-
specific temporal delays down the BM. The shortest chirp in our study (8.5 ms) was shown to evoke significantly less synchronous responses in male cochlea, whereas the longest chirp (11.5 ms) was shown to evoke significantly less synchronous responses in female cochlea. These results imply that a tailoring of chirp parameters to gender would be beneficial—particularly in pathologies that severely affect neural synchrony (e.g., multiple sclerosis) or in applications that assume a correlation between response latency and site of BM activation (e.g., tumor detection via stacked-ABR). However, when comparing results for custom chirps against those of the normative A-Chirp, there were very few statistical differences in synchrony measures between chirp lengths. We must conclude that a population-based chirp, such as the A-Chirp, is appropriate for most clinical work.

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