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The Effects of Musical Intervals from Consonant to Dissonant on the Neural Activity of Earthworms

Charley L. Weaver

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THE EFFECTS OF MUSICAL INTERVALS FROM
CONSONANT TO DISSONANT ON THE NEURAL
ACTIVITY OF EARTHWORMS

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COLUMBUS STATE UNIVERSITY

THE EFFECTS OF MUSICAL INTERVALS FROM CONSONANT TO DISSONANT ON
THE NEURAL ACTIVITY OF EARTHWORMS

A THESIS SUBMITTED TO THE
HONORS COLLEGE
IN PARTIAL FULFILLMENT OF THE
REQUIREMENTS FOR HONORS IN THE DEGREE OF
BACHELOR OF ARTS
DEPARTMENT OF BIOLOGY
COLLEGE OF LETTERS AND SCIENCES

BY

CHARLEY L. WEAVER

COLUMBUS, GEORGIA

2016

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For Honors in the Degree of

BACHELOR OF ARTS

BIRDAWY

COLLEGE OF LETTERS AND SCIENCES

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Date 12/7/16

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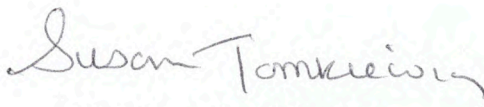


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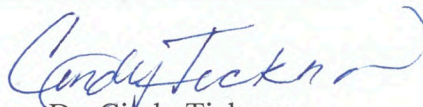


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ACKNOWLEDGMENTS ABSTRACT

Music is generated through vibrations of a medium creating what is perceived as tone. Two frequencies played simultaneously create intervals, and those ratios of frequencies determine what is consonant and dissonant. Studies have shown a greater neural response in human participants to consonant intervals (Bidelman and Heinz 2011, Bidelman and Krishnan 2009). In this study, the ventral nerve cord of *Lumbricus terrestris* was exposed to six dichotic intervals in the form of vibrations through a bass actuator. Nerve activity was recorded for amplitude (mV), duration (ms), and maximum depolarization (mV) of the action potential. There was no significant difference in amplitude, duration, or maximum depolarization across all six interval treatments.

INDEX WORDS: Earthworm, Neural Activity, Music, Interval.

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INTRODUCTION

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vocal folds of the larynx (Helmholtz 1954). These vibrations vibrate the air, which is transduced into sound through the ear and sensory nerves. Each note, or tone, has its own frequency of vibration. For example, the note A in the middle of the treble clef staff has a frequency of vibration of 440 Hz. The higher the pitch is, the higher the frequency and shorter the vibration period of the sound wave (Helmholtz 1954).

Considering the frequency of a note is important when beginning to pair notes together. Music is not always one pitch at one time. Polyphony, the beginnings of harmony in western civilization, introduced the pairing of notes and the inclusion of intervals in music in the Middle Ages. Ancient Greece also had a system of what was known as "harmonia", or intervals within an octave, even before the Middle Ages. This system of intervals was hard to distinguish aurally because of the differences in tuning of the lyre (Lippman 1964). The frequencies between pitches generate a mathematical ratio, which then can be classified into one of twelve standard intervals. A unison, or same pitch played at the same time, would have a 1:1 ratio. All frequency ratios within this category would have the same qualities. An octave, which is the same note name played at twice the frequency and the original note together, would have a 1:2 ratio. A ratio of 1:2 means that as the bottom note generates one vibration, the top note makes two vibrations

INTRODUCTION

The debate about what is music is ongoing. The broad conception of music is *sound within a set boundary of time*. However, for the purposes of this experiment, the focus will be on tonal music and harmonies and the mathematical relationships between them. Tone is considered a uniform sound with regular vibration of the air or comparable medium (Helmholtz 1954).

Musical tones are generated through vibrations of some physical substance. The vibrations can be made by the strings on a grand piano, the double reed of an oboe, or even the vocal folds of the larynx (Helmholtz 1954). These vibrations vibrate the air, which is transduced into sound through the ear and sensory nerves. Each note, or tone, has its own frequency of vibration. For example, the note A in the middle of the treble clef staff has a frequency of vibration of 440 Hz. The higher the pitch is, the higher the frequency and shorter the vibration period of the sound wave (Helmholtz 1954).

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over the same period time (Helmholtz 1954). Frequency ratios are the basis of what is consonant and what is dissonant, or what sounds pleasing versus what sounds unstable.

Consonances are determined by the overtone series, or harmonic series. An overtone series is series frequencies that occur above the note being played. These can be determined by multiplying the initial pitch frequency by 1,2,3,4, etc. The higher the frequency, the closer the notes of the overtone series become. The order of the series is an octave, followed by a fifth, fourth, major third, and minor third in that order (Bain 2003). The closer to the beginning of the series, the more “consonant” the interval. The list of ratios of these intervals is shown in Figure 1 (Helmholtz 1954).

Interval	Ratio
Octave	1:2
Fifth	2:3
Fourth	3:4
major Third	4:5
minor Third	5:6

Figure 1. Relationship between consonant intervals and frequency ratios.

While consonant intervals contain a sense of stability with simple frequency ratios, dissonance contains the opposite. Dissonance is found further in the overtone series and has higher frequency ratios. These ratios are listed in Figure 2 (Bidelman and Krishnan 2009).

Interval	Ratio
major Seventh	15:8
minor Second	16:15
Tritone	45:32

Figure 2. Relationship between dissonant intervals and frequency ratios.

Dissonant interval frequencies do not line up with each other. They contain conflicting waves, which cause the interval to not be stable. Because of this instability, the resulting interval is more difficult for the brain to analyze (Errede 2002).

In a study by Bidelman and Krishnan (2009), consonant intervals had a larger neural response than dissonant intervals in human participants. The combined notes were not played in both ears. One note was played in one ear, while the other note of the interval was played in the opposite ear. Summation would have needed to occur for the interval to be processed. These intervals would not normally be isolated notes in either ear in the environment. However, the presentation of one note to one ear insured correct readings from frequency-following responses (FFR) in the brainstem.

Another study also showed a similar pattern of strength of neural activity of consonant intervals in humans. In this study, pitch salience of auditory nerve responses was compared over all twelve intervals within an octave for normal hearing and hearing impaired (Bidelman and Heinz 2010). Pitch salience is *the measure of periodicity of a neural stimulus standardized over a period* (Deutsch 3013). The neural pitch salience of all twelve intervals not only showed peaks at the most consonant intervals and valleys at the dissonant intervals, but it also showed the hierarchy of consonance. The octave had the highest neural pitch salience followed by the fifth, fourth, and so forth (Bidelman and Heinz 2010). However, subjects with impaired hearing showed little discrimination between the intervals (Bidelman and Heinz 2010).

The sensory organ for hearing and vibration detection in vertebrates and invertebrates is similar in regards to the mechanoreceptors that transduce the sound waves into action potentials. Mechanoreceptors are also used for touch reception and balance. For vertebrates, the

mechanoreceptor is a bundle of hair cells consisting of microvilli, or stereocilia, of increasing height from one side to the other. When the microvilli are bent toward the largest stereocilia, a depolarization occurs and more neurotransmitter is released. When the microvilli are bent in the opposite direction, the cell is hyperpolarized and less neurotransmitter is released (Hill et. al 2012).

Invertebrates use a similar mechanoreceptor, but instead of hair cells made of microvilli, they are made of cilia. When the tympanum is vibrated in response to a sound wave, the cilia are bent and a depolarization occurs. The tympanum and its associated mechanoreceptors do not have to be located on the head. For crickets, the mechanoreceptor is located on the legs, and for some moths it is on the thorax (Hill et. al 2012).

Earthworms, however, do not have a tympanum and cannot hear (Darwin 1882). Instead of hearing, earthworms are very sensitive to vibrations (Mitra 2009). However, if hearing is defined as *an animal's response to a mechanical disturbance of their environment by special organs that are meant to transduce those vibrations to action potentials*, then earthworms can hear to a certain extent (Roy 1994).

Earthworms contain "sensory buds" along the length of their body. These sensory buds are clusters of ciliated mechanoreceptors, which are elongated and synapse with a peripheral nerve deep within the epithelium (Langdon 1895). In a study by Moment and Johnson (1979), the sensory buds were found to be clustered within the anterior half of the worm. While there were few sensory buds on the posterior segments, the buds were located the anterior portion of those segments.

The medium with which the vibration moves through is selective for earthworms. Earthworms do not respond to sound waves and vibrations moving through a gaseous medium.

Earthworms are most receptive to vibrations of a solid medium (Darwin 1882). In a study by Darwin (1882), earthworms were placed in respective pots on top of a piano. When the piano played the notes C at 130.81 Hz and G at 783.99 Hz, the worms burrowed into the soil each time a note was played.

Likewise, in a study by Mitra (2009), vibrations were created below 500 Hz, and the abundance of emergence of earthworms decreased the further away from the source of the vibrations. Instead of burrowing, the earthworms emerged. Therefore, the responses to vibration can be varied. Earthworms can emerge from the soil or burrow into the soil in response to a stimulus (Mitra 2009).

This study aimed to compare the neural activity of earthworms across eight dichotic interval treatments; starting from the octave, which is the most consonant, to a tritone, which is the most dissonant. I predicted the neural activity will be lowest with the most dissonant interval because of the conflicting vibration patterns of the two notes through the medium. For example, as one note vibrates 45 times, the other will vibrate 32 times in the same time period in a tritone. However, I predicted the consonant intervals will give a more consistent vibration pattern and a stable waveform. This will cause a "louder" stimulus, and ultimately a larger response in amplitude and duration.

MATERIALS AND METHODS

Lumbricus terrestris.

The large earthworms (Carolina Biological: 14-1620) were kept in their respective containers filled with moist soil until dissected. The worms were kept at 15°C or lower to ensure survival. Earthworms were anesthetized in 10% ethanol for five minutes. The earthworms were

dissected by placing the earthworm dorsal side up and making an incision through the midsection of the worm to expose the ventral nerve cord. Once the nerve cord was exposed and isolated, a section approximately 4 cm in length was extracted. The ~4 cm section was placed in a Biopac nerve chamber on top of the Aura bass actuator (ACT-50-4) dorsal side facing up. Earthworm ringer's solution was used periodically to keep the nerve viable.

Interval Treatments.

There were six dichotic interval treatments: three consonant and three dissonant. The intervals were chosen based on their position in the overtone series. The consonant intervals were the octave, perfect fifth, and perfect fourth. The dissonant intervals were the major seventh, minor second, and tritone. The lowest note in the interval was set at 261.63 Hz, or middle C. The second note within the interval was placed above middle C. For example, a perfect fifth will contain middle C at 261.63 Hz and G at 392.00 Hz. Placing the intervals within the fourth octave ensured frequencies below 500 Hz as described in the study by Mitra (2009), which elicited a neural response. There were ten trials consisting of all the interval treatments in different orders. The different orders of interval treatments ensured that the interval itself, rather than the order of intervals, elicited the neural response. The interval treatments simulated vibrations through the soil because the sound vibrations will travel through a solid medium, the nerve chamber and the vibration device.

The notes were played simultaneously by a tone generator through the Aura bass actuator. Each interval was played for two seconds, and stimulation at 0.8 V using a Biopac stimulator (BSLSTMB) was applied immediately along with the continuing vibration stimulus. As a control, I tested singular notes along the fourth octave to make sure similar nerve activity

was occurring for each note alone without conflicting vibrations that occur between two notes in an interval. Vibration alone did not reach the threshold for an action potential, so vibration was coupled with a 0.8 V electrical stimulation to detect an action potential.

Neural activity analysis.

Neural activity was measured by placing dissected earthworms in a Biopac nerve chamber. Each interval was played for two seconds, and the neural response for each interval was recorded via BSL MP35. Each of the ten individual worms was exposed to all six interval treatments in different orders. The neural response was analyzed for average (\pm 1 S.E.) amplitude (mV), duration (ms), and maximum depolarization (mV). Results were analyzed using a 1-way ANOVA, and a Post-hoc Least Significance Differences test.

RESULTS

All results are expressed as average \pm standard deviation. There was no significant difference in the amplitude (mV) of the action potentials across all six interval treatments (1-way ANOVA, $F_{6,63}=0.496$, $P=0.809$; Figure 3). There was no significant difference between amplitude of neural response with exposure to the octave, perfect fifth, or perfect fourth (19.61 \pm 1.99, 20.22 \pm 2.16, and 20.13 \pm 2.07, respectively). Likewise, there was no significant difference between amplitude of neural response with exposure to the major seventh, minor second, and tritone (20.42 \pm 2.27, 19.51 \pm 2.02, and 19.55 \pm 1.32, respectively).

Duration (ms) of action potentials also showed no significant difference across either the three consonant or three dissonant interval treatments (1-way ANOVA, $F_{6,63}=0.240$, $P=0.961$; Figure 4). There was no significant difference of duration of action potentials with exposure to

the octave, perfect fifth, or perfect fourth (0.44 ± 0.06 , 0.43 ± 0.08 , and 0.46 ± 0.09 , respectively). Continuing with the trend, there was no significant difference in duration of neural response with exposure to the major seventh, minor second, or tritone (0.44 ± 0.11 , 0.43 ± 0.07 , and 0.46 ± 0.09 , respectively).

A 1-way ANOVA also showed no significant difference in maximum depolarization (mV) for all six interval treatments ($F_{6,63}=0.390$, $P=0.883$; Figure 5). There was no significant difference in maximum depolarization of neural response with exposure to the octave, perfect fifth, or perfect fourth (13.39 ± 1.30 , 13.50 ± 1.48 , and 13.83 ± 1.11 , respectively).

Likewise, there was no significant difference in maximum depolarization of neural response with exposure to the major seventh, minor second, and tritone (13.47 ± 1.59 , 14.02 ± 1.04 , and 13.88 ± 1.52 , respectively). Overall, there were no significant differences between the interval treatments for all three parameters studied: amplitude (mV), duration (ms), and maximum depolarization (mV).

Figure 3: Average (± 1 S.E.) action potential amplitude across six interval treatments. Earthworms ($n=10$) were exposed to the two tones of the interval simultaneously for two seconds and a stimulus coupled with the vibration after to generate the action potential. There was no significant difference in amplitude of the action potentials across all six treatments (1-way ANOVA, $F_{6,54}=0.496$, $P=0.889$).

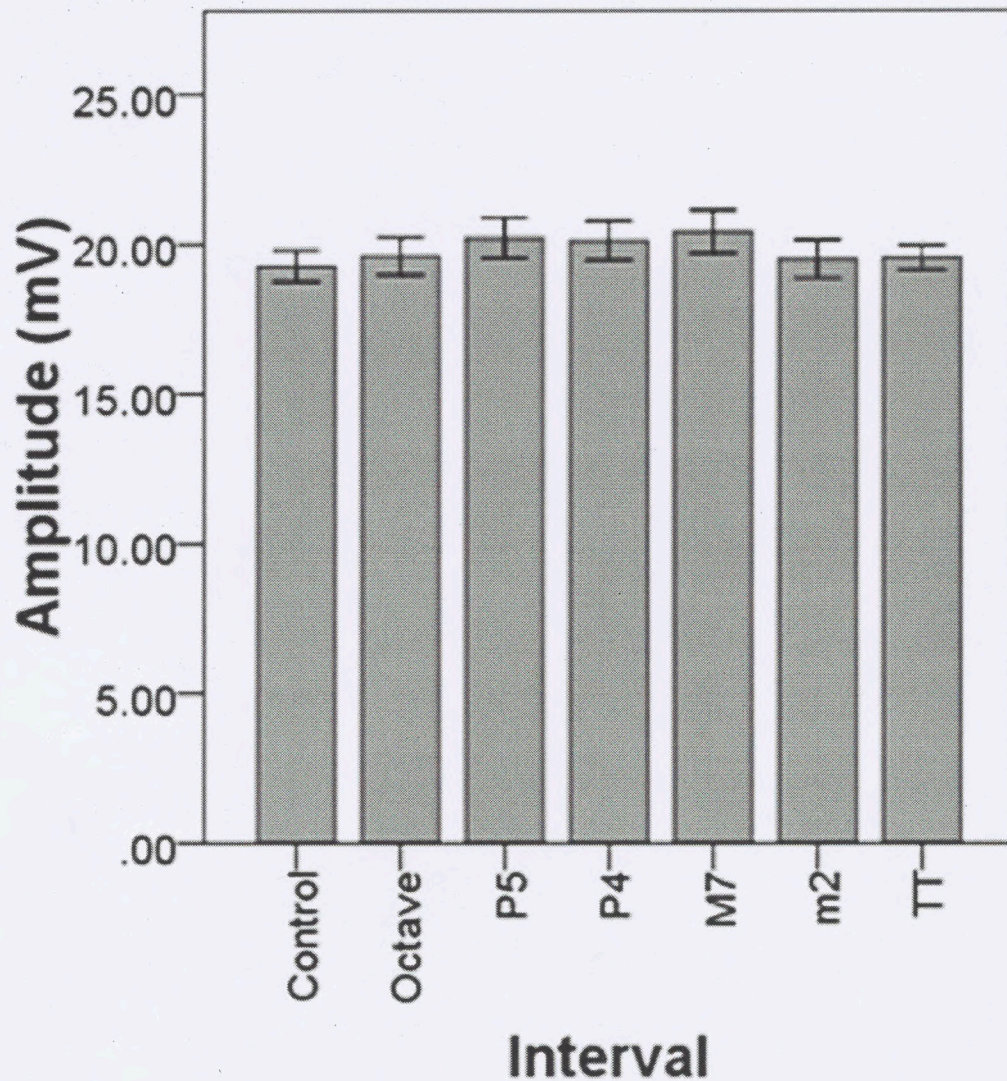


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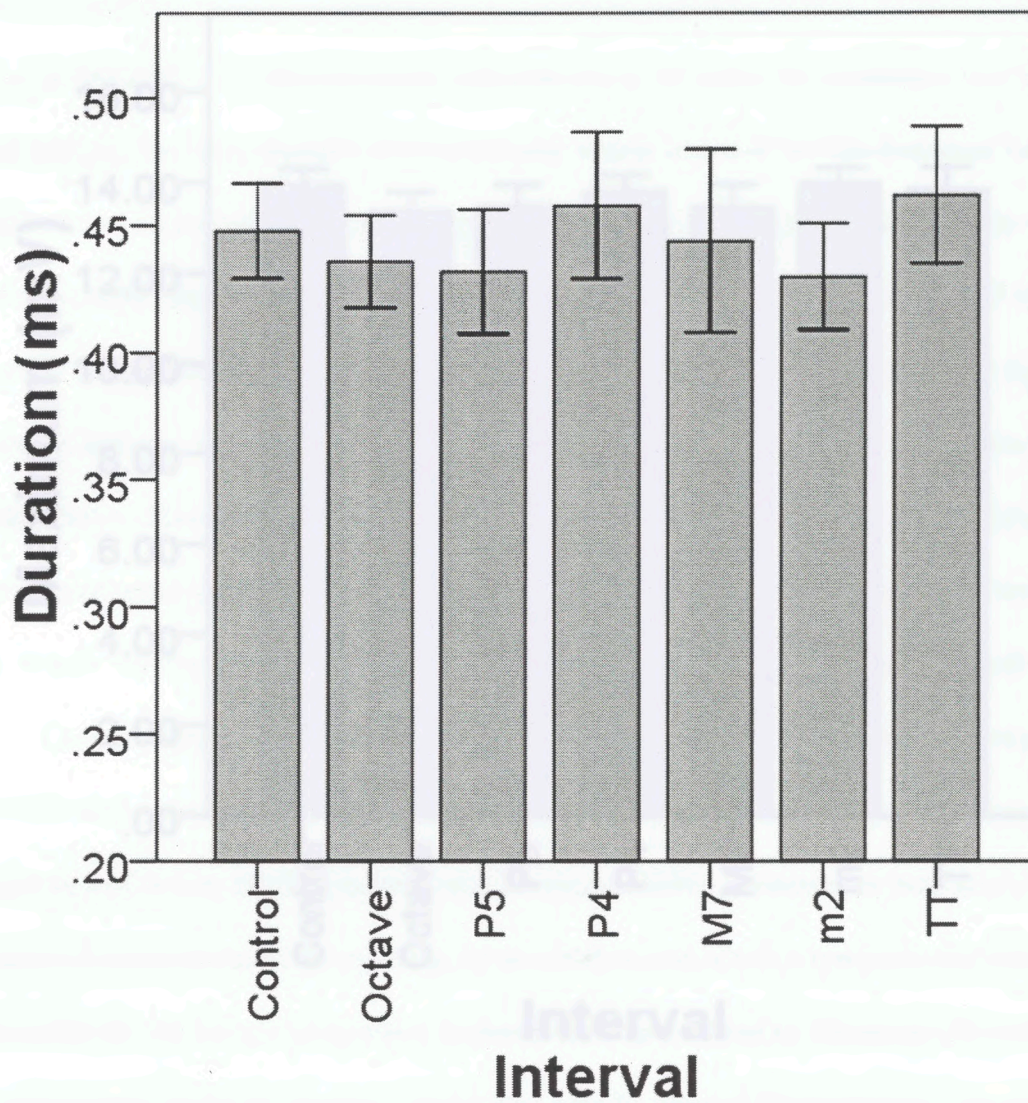


Figure 4. Average (± 1 S.E.) action potential duration in response to six interval treatments from consonant to dissonant. Earthworms ($n=10$) were exposed to the two notes making up the interval for two seconds before a 0.8 V electrical stimulus was coupled with the vibration to generate an action potential. There was no significant difference in duration of action potentials across all six interval treatments (1-way ANOVA, $F_{6,63}=0.240$, $P=0.961$).

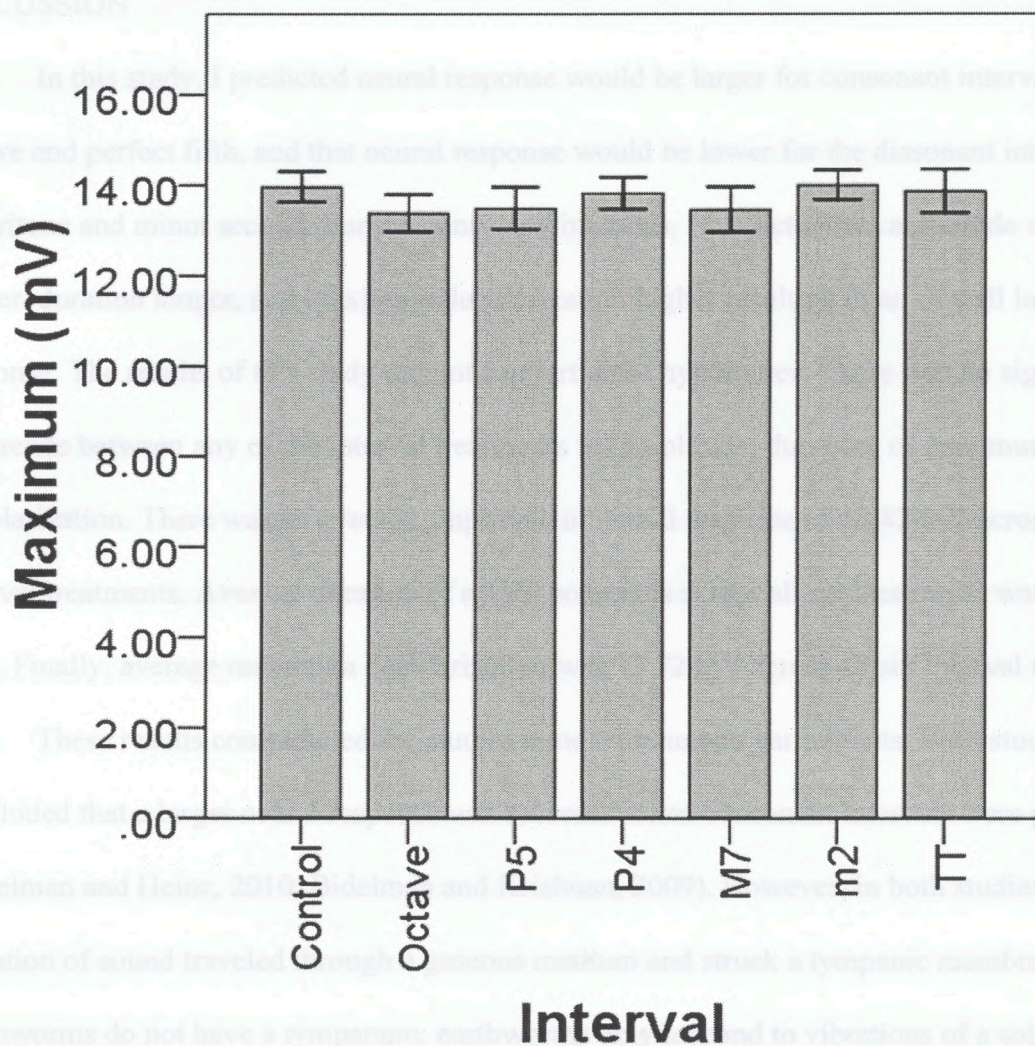


Figure 5. Average (± 1 S.E.) maximum depolarization for six interval treatments from consonant to dissonant. Earthworms ($n=10$) were exposed to each interval treatment in a different order. Exposure to the vibration of the interval occurred for two seconds, and an electrical stimulus coupled with the vibration generated the action potential. There was no significant difference in maximum depolarization across all interval treatments (1-way ANOVA, $F_{6,63}=0.390$, $P=0.883$).

DISCUSSION

In this study, I predicted neural response would be larger for consonant intervals, like the octave and perfect fifth, and that neural response would be lower for the dissonant intervals, like the tritone and minor second. For the consonant intervals, I predicted that amplitude would be higher, duration longer, and maximum depolarization higher resulting in an overall larger neural response. The results of this study did not support these hypotheses. There was no significant difference between any of the interval treatments for amplitude, duration, or maximum depolarization. There was an average amplitude of neural response of 19.82 mV across all interval treatments. Average duration of action potentials across all six treatments was 0.44 ms long. Finally, average maximum depolarization was 13.72 mV across all six interval treatments.

These results contradicted the studies done with human participants. Both studies concluded that a larger neural response was recorded when consonant intervals were played (Bidelman and Heinz, 2010; Bidelman and Krishnan, 2009). However, in both studies, the vibration of sound traveled through a gaseous medium and struck a tympanic membrane. Earthworms do not have a tympanum; earthworms only respond to vibrations of a solid medium. The earthworm's response to sound could very well be much different than a human because of the medium the sound waves pass through.

Singular notes, while not the focus of this study, did elicit a consistent 2 mV increase in response when a 0.8 V electrical stimulus was also applied along with the vibration (data not shown). However, two frequencies played simultaneously do not significantly differ from the electrical stimulus alone. Studies by Mitra (2009) and Darwin (1882) showed behavioral responses to singular notes, and thus only one frequency was applied to the environment at one time. The study by Mitra showed emergence of earthworms when a 500 Hz frequency or lower

was applied directly in the soil (2009). Contrastingly, Darwin's study showed burrowing of earthworms as vibrations of a piano string traveled through many different objects: piano lid, container, soil, and eventually to the earthworm (1882). Earthworms may respond easier to singular notes. Similar to dissonance proving difficult for the human brain to analyze, intervals may be difficult for earthworms to analyze.

Future Studies

While intervals did not seem to elicit a different neural response, going forward, it would be interesting to discover the true range of frequencies that elicit a response in earthworms. It has been shown that earthworms respond to frequencies below 500 Hz and up to 700 Hz (Mitra 2009, Darwin 1882).

A study by Shannon et al. explained another method of recording action potentials of the ventral nerve cord in earthworms that included the anterior portion of the worm, with the most sensory buds, in vibration exposure (2013). In this method, the earthworm remains intact on top of a solid surface with electrodes placed along the length of the body directly adjacent to the ventral nerve cord (Shannon et al. 2013). This solid surface could be placed on top of the bass actuator for the entire worm to be exposed to the different frequencies of vibration. The exposure of the anterior portion of the worm would give a higher probability that mechanoreceptors in the sensory buds would be activated and a response recorded.

Finally, another model organism, such as fish, could be used in place of the earthworm. An organism that has a tympanic membrane for the vibrations to strike would correlate more similarly to humans than an organism that does not have a tympanum. While the sound would

pass through a liquid medium, fish may elicit a differing response to consonant and dissonant intervals.

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