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Best Practices for Bioacoustic Analysis of Wood Frog (*Rana sylvatica*) Advertisement Calls Over a Suburbanization Gradient

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Best Practices for Bioacoustic Analysis of Wood Frogs (*Rana sylvatica*) Advertisement Calls over a Suburbanization Gradient



ABSTRACT

- Identifying how species respond to anthropogenic changes in their environment is important for understanding human impact.
- I developed bioacoustics methods to analyze how wood frog (*Rana sylvatica*) advertisement calls vary in pitch, duration, and number across a suburbanization gradient (Figure 1).
- I examined call structure and different measures of frequency. Center and 95% frequency have more variability when compared to peak and 5% frequency.
- Wood frog calls are difficult to analyze for many reasons, but I have developed a method to quantify these calls and capture their variability.

INTRODUCTION

- Wood frogs' distribution makes them an excellent species to test the impacts of environmental change on their behavior and physiology¹.
- Wood frog (Figure 4) advertisement calls are hard to analyze as they are short, overlapping, and lack complex call structure¹.
- The hypothesis is that wood frogs found in more urban areas will have higher pitches (Figure 2), decreased durations, and a lower number of calls when compared to frogs from isolated areas².



Figure 1. Left to right: rural to suburban gradient of wood frog breeding ponds (light blue) surrounded by 2.5%, 25.4%, 40.4% and 74.7% suburban cover in a 200 m radius (circles). Used with permission from Holgerson et al, in prep.

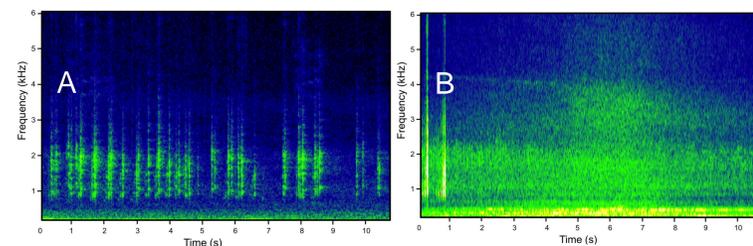


Figure 2. A) Spectrogram of wood frog (*Rana sylvatica*) calls and B) traffic noise recorded at a roadside vernal pool



Figure 3. Diagram showing how audio data were collected. Three wood frogs were placed in a kiddie pool with a shotgun microphone and recording equipment and were allowed to perform mate-searching behavior. Made using Biorender.



Figure 4. Wood frogs (*Rana sylvatica*) in amplexus, male (top) and female (bottom). Photo by L. Swierk.

METHODS

- A total of 42 trials (Figure 3) from Madison, Connecticut were recorded with 2-3 male wood frogs each.
- Audio collected was analyzed with the program Raven Pro 1.6.4.
- Spectrograms have a y-axis that shows pitch (frequency, kHz), and an x-axis that shows time (Figure 5).
- The darker the color, the more intense the sound (greater "power").

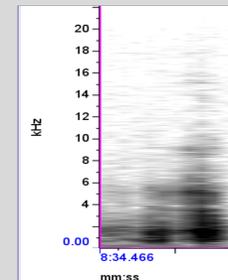


Figure 5. Wood frog advertisement call in spectrogram form, made using Raven Pro

PRELIMINARY RESULTS

Peak Frequency vs Center Frequency

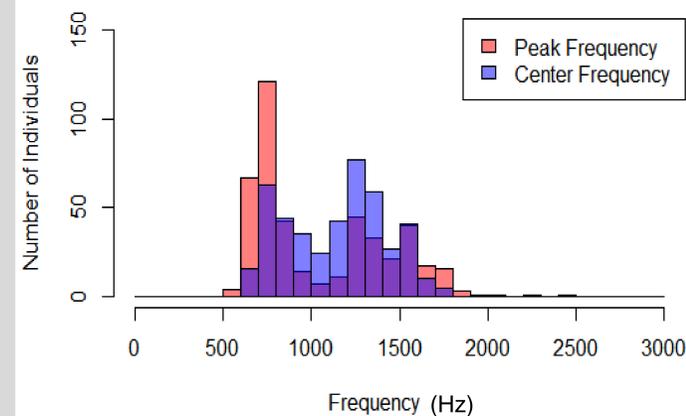


Figure 6. A comparison of peak frequencies and center frequencies from individual advertisement calls. Peak frequency is the frequency at which max power occurs in a selection, and the center frequency is the frequency that splits a selection into two parts of equal energy.

Data for Frequency 5% and 95%

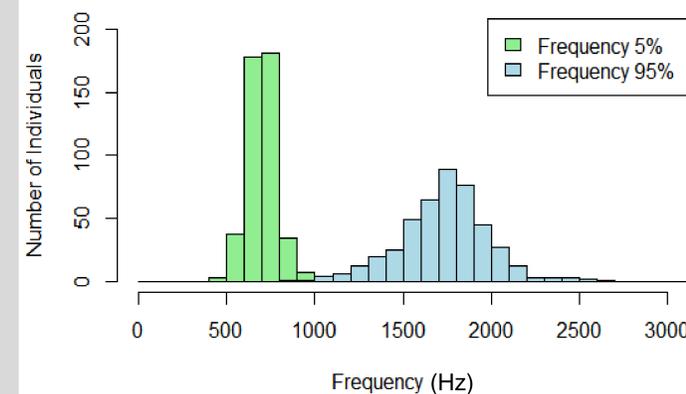


Figure 7. A comparison of frequencies 5% and 95% from individual advertisement calls. Frequency 5% is the frequency at which 5% of the energy in a spectrogram selection has been reached. Frequency 95% is the frequency at which 95% of the energy in a spectrogram selection has been reached.

PRELIMINARY RESULTS (CONT.)

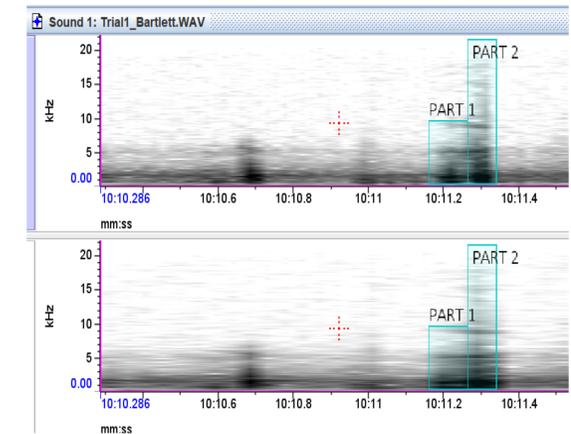


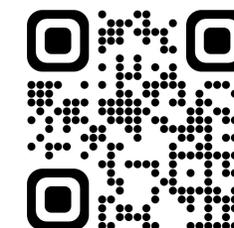
Figure 8. Example of how a single advertisement call was split into two parts to be analyzed. The first spectrogram has a Hann window of 2048 (frequency resolution) and the second spectrogram has a Hann window of 4096 (time resolution).

- Boxes to analyze calls were hand-drawn around both parts of each wood frog call. Splitting the calls into two parts made analysis more accurate.
- Center frequency has more variability than peak frequency; individuals vary more when it comes to center frequency in calls (Figure 6).
- Frequency 95% has more variability than frequency 5%; individuals vary more when it comes to 95% frequency in calls (Figure 7).
- The Hann window of 2048 has better frequency resolution, and the Hann window of 4096 has better time resolution (Figure 8).
- 90% duration (time measurement based on the dominant sound energy) was used to account for any subjectivity in hand-drawn boxes.

DISCUSSION

- Some aspects of wood frog advertisement calls have more variability than others.
- This variability is important when it comes to changes in advertisement calls since it suggests which call components are more plastic.
- Some aspects of calls appear to be more consistent or less affected by the environment.
- These methods can be used to analyze calls in other frog species.
- In the future, the reason for differing variability in aspects of wood frog calls can be examined.

REFERENCES



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