



# Practiced Perfect or Spontaneous Singing: Do Zebra Finches Warm Up Before Their Songs?

Andrew Magee, Rima Abram, Kevin Bien, Hannah Scheffer, & Sam Shah

Advisor: Dr. Brent Cooper



## Introduction

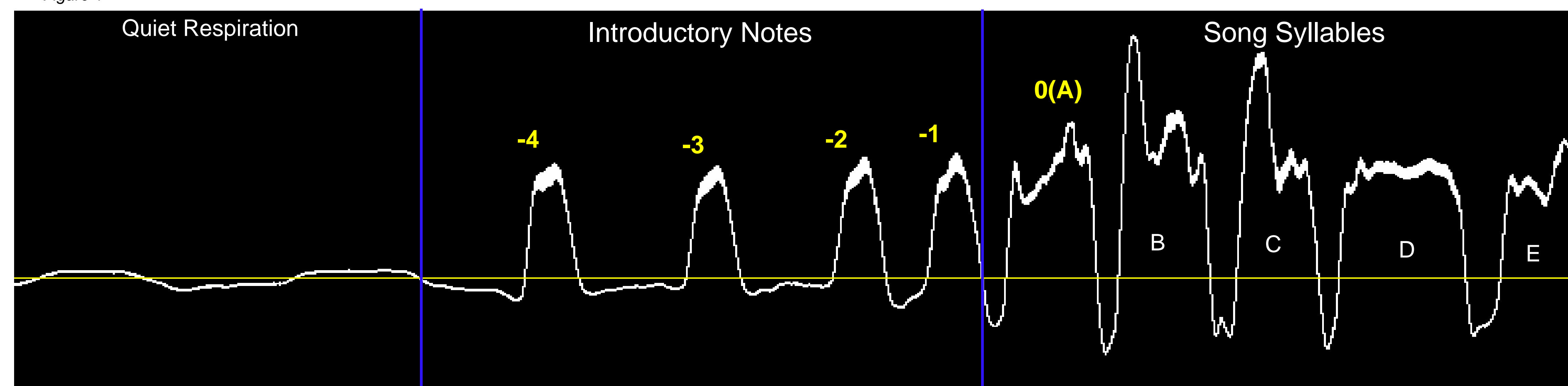
- Language has a strong connection to human cognition, allowing for:
  - conceptualization and abstract thinking
  - conveying ideas
  - enhanced social understanding, symbolic thinking, and logic
  - numerous other advanced cognitive actions
- Language must be considered, understood, and researched for human cognition to be fully understood.
- As speech is a complex and sophisticated vocal behavior, it requires a great deal of preparation, bodily coordination, and motor planning.
  - Mentally, speakers plan what they will say in multi-word chunks
  - Neurologically, the ventral premotor cortex and premotor cortex engage in motor planning
  - Physiologically, respiration is modulated, and voluntary control of the larynx produces and controls the voice
  - Speech deficits seen in people with autism, speech apraxia caused by stroke, stuttering, and aphasia are linked to deficits in motor planning
  - Improved understanding of motor preparation for learned vocal production could lead to improved treatment for these disorders
- Zebra finches and other songbirds demonstrate vocal production learning, a capability that is uncommon in non-human animals. Zebra finch song parallels human speech in several important aspects:
  - As humans learn language through vocal imitation of their parents, male zebra finches learn song from their father (only male zebra finches sing)
  - The syrinx, a vocal organ analogous to the larynx, produces sound by modulating respiration and the vibrations of the labial pairs, similar to the function of human vocal folds
  - A number of neurological similarities and analogs exist between human and zebra finches:
    - The HVC in zebra finches is analogous to the premotor cortex in humans
    - Humans and zebra finches share have similar GABAergic neurons in vocal-related brain regions
    - Humans and zebra finches have more similar vocal pathways in the forebrain than either have to their closer evolutionary relatives
- Zebra finch song also requires motor planning and preparatory respiratory behavior.
  - Introductory notes and song syllables are separated by quick, silent gaps, during which birds take mini-breaths: inspiratory pulses that are short in duration and high in amplitude
  - Interneurons and sets of basal-ganglia projecting neurons change their activity several hundred milliseconds before song bout onset (i.e., first introductory notes are sung), and the changes correlate with successful song sequence initiation, suggesting preparatory neural activity for song production
  - Introductory notes gradually increase in speed and converge on a highly stereotyped acoustic endpoint. Based on these findings, introductory notes been argued to comprise a period of preparatory behavior as the brain reaches a "ready" state before singing. However, introductory notes, particularly those later and closer to the song syllables, could also be part of song rather than separate preparatory behavior. Further investigation is required to explore these possibilities.
- The current study aims to examine the relationship between pressures of inspiratory pulses (mini-breaths) and expiratory pulses (intro notes and song syllables) during song in zebra finches, aiming to understand the relationship between them, and further, the possible functions they serve for preparation before song. Further, the current study aims to provide evidence that may be used to determine whether intro notes are preparatory behaviors prior to song or a part of song itself.

We made the following hypotheses:

H1: The first song syllable will account for a greater amount of variance of the preceding introductory notes than the last introductory note as measured by model  $R^2$ .

H2: Features of introductory notes will converge on the last intro note.

Figure 1



## Method

### Subjects

- 7 adult male zebra finches

### Procedure

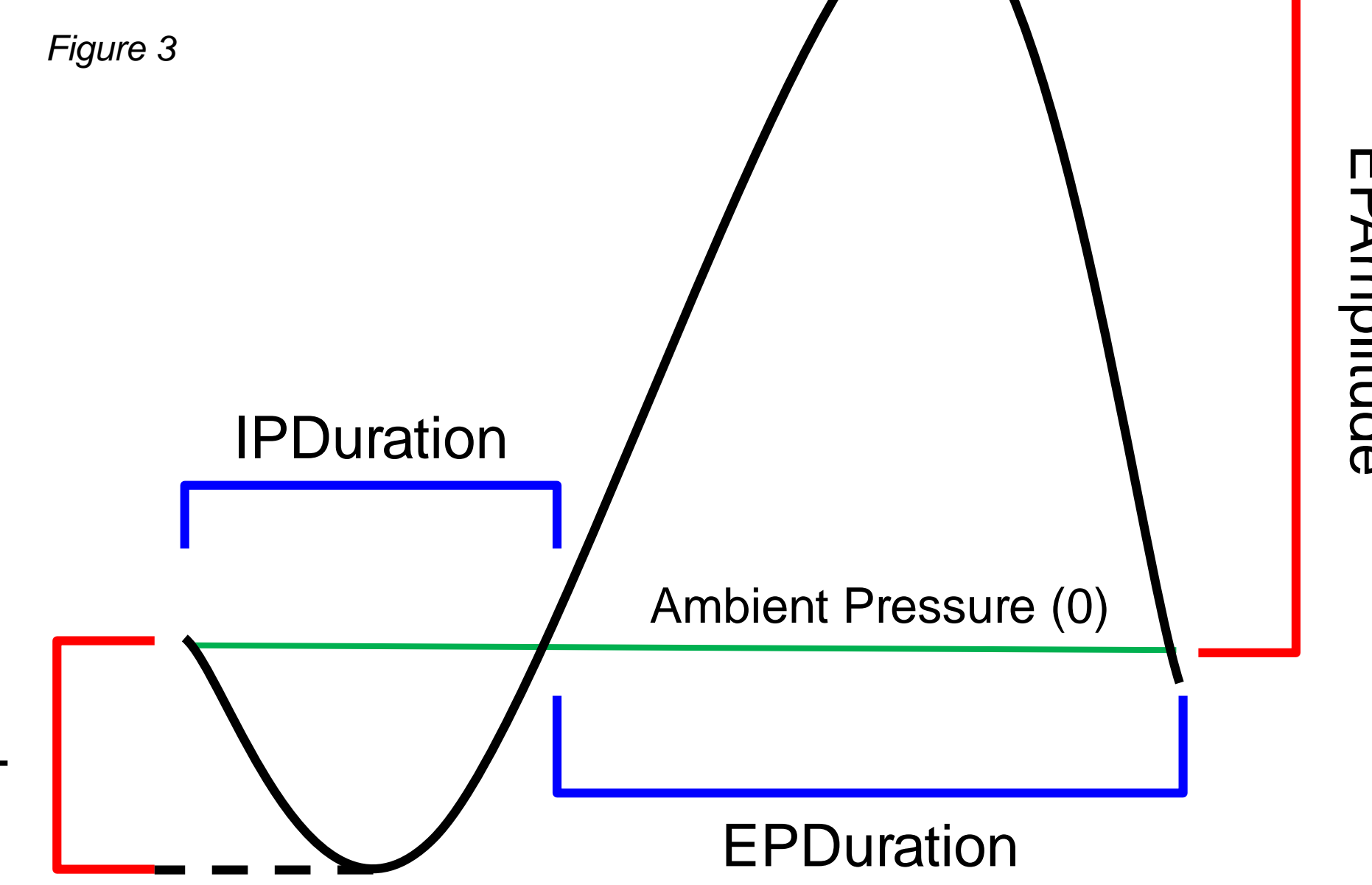
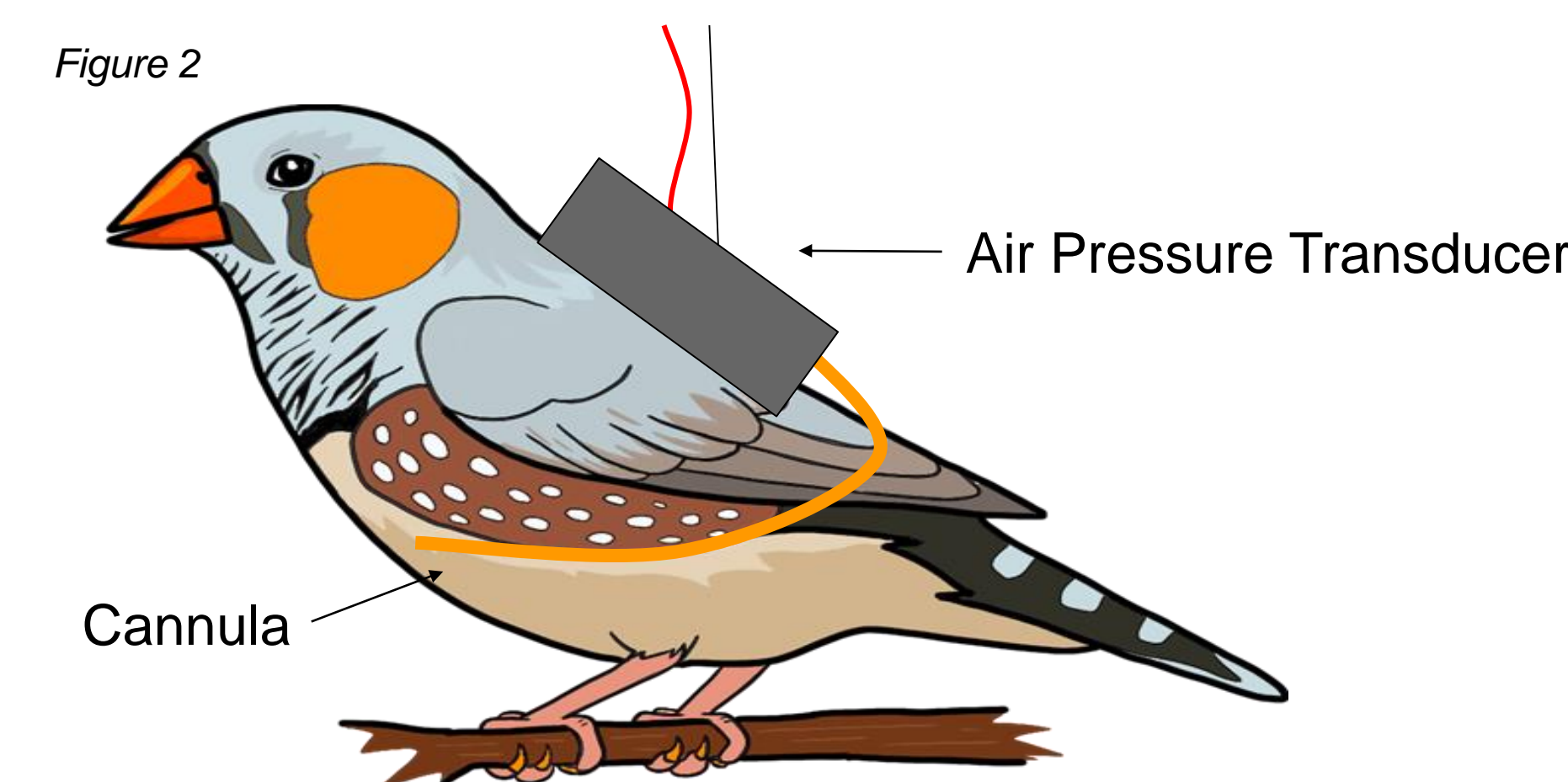
- Recorded audio and subsyringeal air pressure via surgically inserted cannula and pressure transducer (see Figure 1)
- Both directed (female present) and undirected (no female present) singing included

### Measures

- Duration (seconds) of inspiratory and expiratory pulse
- Amplitude (collapsed variable: mean amplitude\*maximum amplitude; mean amplitude\*minimum amplitude) of inspiratory and expiratory pulse (see Figure 2)
- Labels are organized by
  - Pulse type (Inspiratory [IP] or Expiratory [EP])
  - Measure type (Duration [Dur] or Amplitude [Amp])
  - Note position relative to the first song syllable (first song syllable = 0, last intro note = -1, second to last intro note = -2, and so on)

### Data Analyses

- Graphed each individual bird's introductory notes and song syllable duration by amplitude (see Figures 3a-3d)
- Conducted 8 forward regressions comparing the relationships among the outcome variables



Outcome Variable	Independent Variables
IPDuration0	IPDuration-1, IPDuration-2, IPDuration-3, IPDuration-4
IPAmplitude0	IPAmplitude-1, IPAmplitude-2, IPAmplitude-3, IPAmplitude-4
EPDuration0	EPDuration-1, EPDuration-2, EPDuration-3, EPDuration-4
EPAmplitude0	EPAmplitude-1, EPAmplitude-2, EPAmplitude-3, EPAmplitude-4
IPDuration-1	IPDuration-2, IPDuration-3, IPDuration-4
IPAmplitude-1	IPAmplitude-2, IPAmplitude-3, IPAmplitude-4
EPDuration-1	EPDuration-2, EPDuration-3, EPDuration-4
EPAmplitude-1	EPAmplitude-2, EPAmplitude-3, EPAmplitude-4

Table 1: Regressions performed by outcome variable and independent variables

## Results

Outcome Variable	$R^2$	F	p	df <sub>1</sub>	df <sub>2</sub>	Variables Included
IPDur0	-	-	-	-	-	-
IPAm0	.902	18.372	.010	2	4	IPAm-1, IPAm-2
EPDur0	-	-	-	-	-	-
EPAm0	.657	9.559	.027	1	5	EPAm-1
IPDur-1	.597	7.420	.042	1	5	IPDur-2
IPAm-1	-	-	-	-	-	-
EPDur-1	.914	53.412	<.001	1	5	EPAm-2
EPAm-1	.941	80.165	<.001	1	5	EPAm-3

IPDur0, EPDur0, and IPAm-1 were not significantly predicted by any variables.

- The strongest relationships were seen in the expiration of the last intro note.
- The last introductory note accounted for greater variance than the first song syllable in all comparisons (excepting IPAm0 and EPAm0), contradicting our first hypothesis.
- All significant predictors (excepting EPAm-3) is immediately preceded by the outcome variable or a significant predictor.
- Five out of seven birds followed an pattern where introductory notes approximated a linear arrangement in chronological order, supporting our second hypothesis. Two birds displayed no clear pattern with disordered introductory notes

### Ordered Introductory Notes

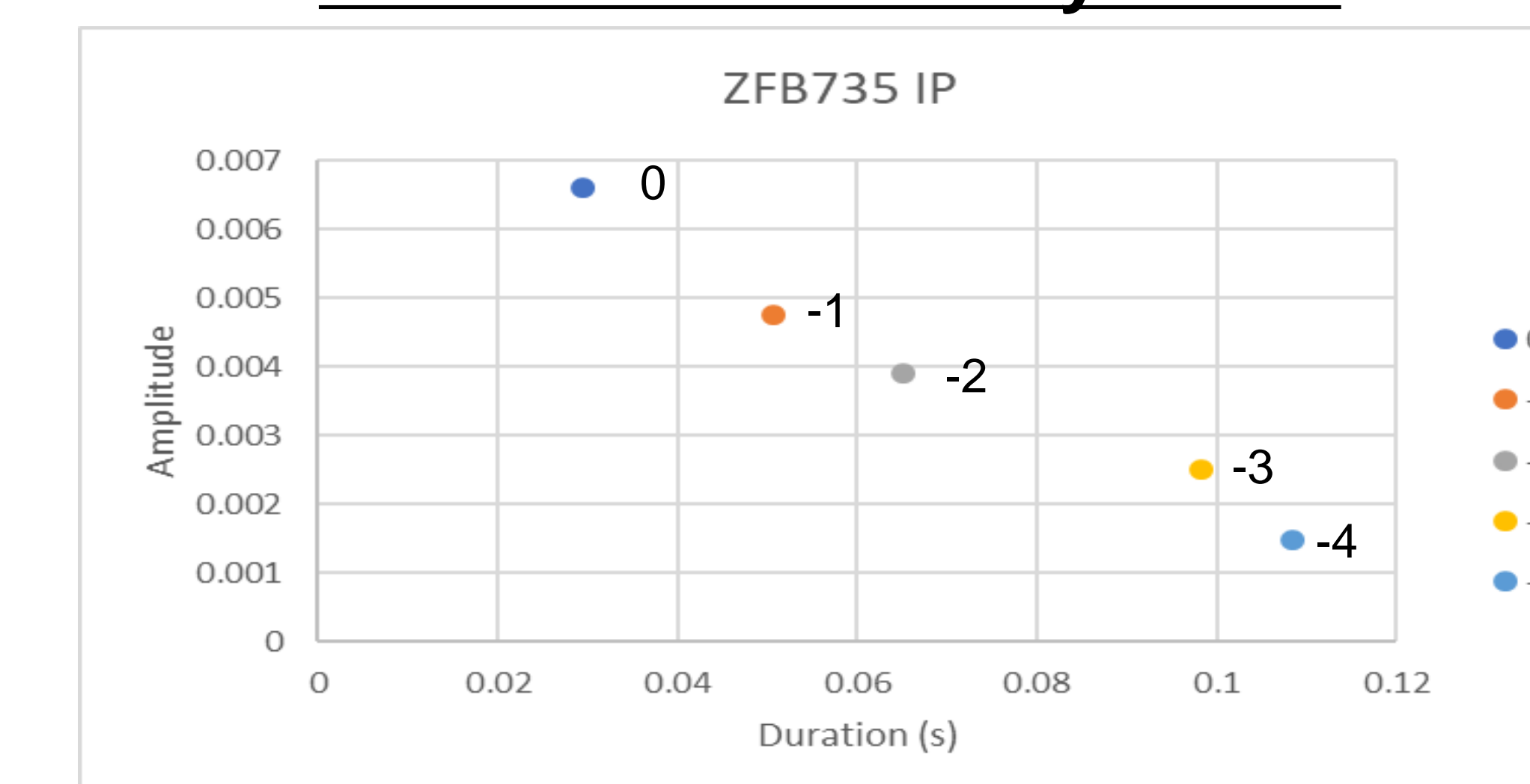


Figure 4a

### Disordered Introductory Notes

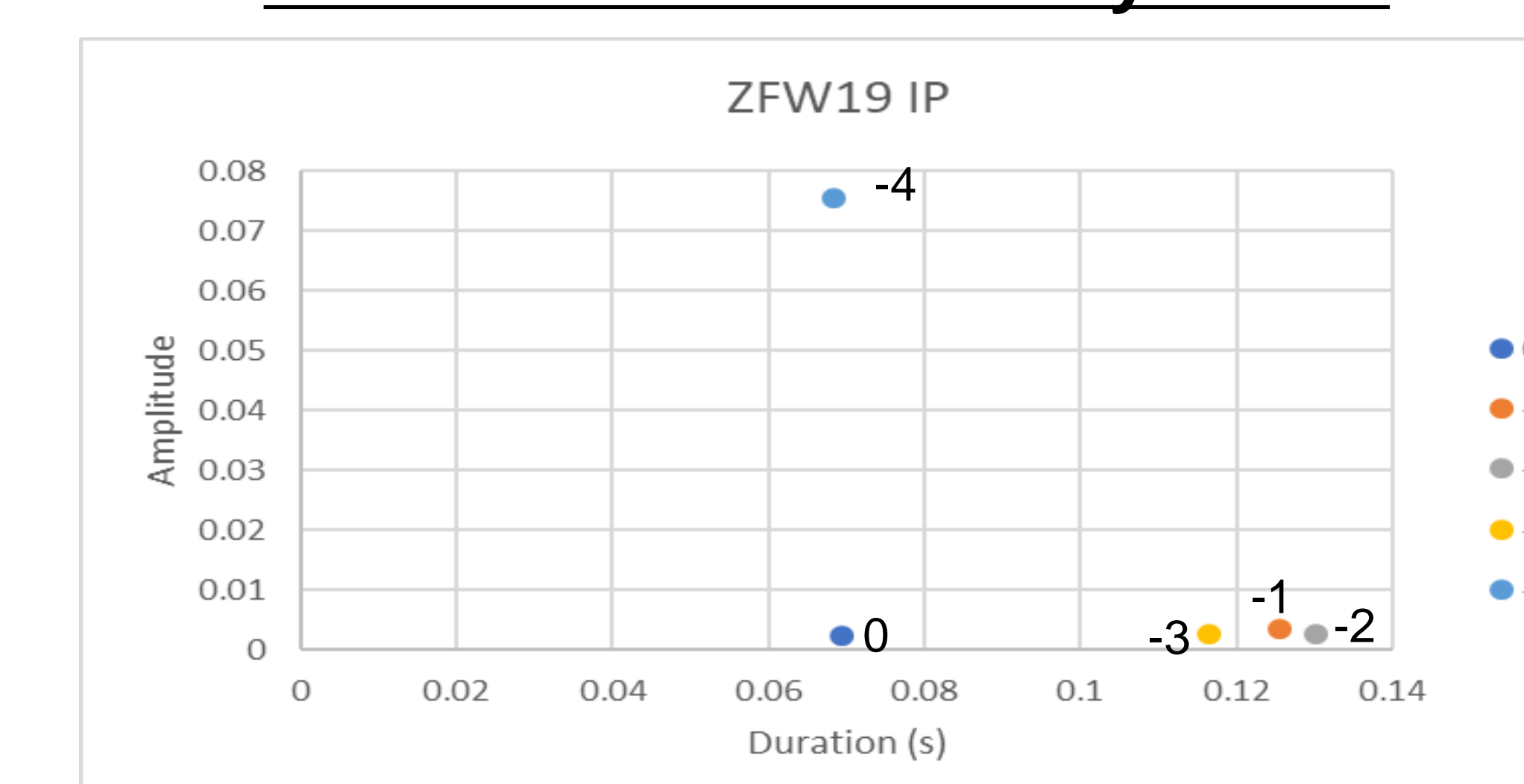


Figure 4b

## Discussion

The results of the final model comparisons appear to contradict our first hypothesis. Three outcome variables for the last introductory note had significant relationships, while the first syllable had two. Additionally, the average variance accounted for was higher for the introductory note analyses (81.7%) than for the syllable (78.0%). Although this averaged value may not be necessarily meaningful, and the difference is small, this might imply that introductory notes have a stronger relationship to the last introductory note than to the first syllable. Nevertheless, the pattern seen with the majority of the birds provides support for preparatory behavior preceding zebra finch song.

The current study has a few limitations. The sample size was small for regression analyses. The data is better analyzed by analyses besides regression, such as hierarchical linear modeling, or linear mixed effect modeling. A greater number of introductory notes and further examination of song syllables would may also provide further information.

## References

- Aamodt, S. (1999) Singing in the brain: Song learning in adult zebra finches. *Nature Neuroscience*, 2, 590.
- Beecher, P. M. & Berke, A. (1997) *Alpha*. In: *Neuroscience P. D., editor. Handbook of Neurophysiology and Aging*. (pp. 299-314). New York and London: Plenum Press.
- Berke, G. S. & Long, J. L. (2010) Functions of the larynx and production of sounds. In: *Avian Vocalization*. S. M. editor. *Handbook of Behavioral Neuroscience*. (Vol. 19, pp. 419-450). Elsevier.
- Chow, H. M., Garnett, E. O., Ratner, N. B., & Chang, S. E. (2023) Brain activity during the preparation and production of spontaneous speech in children with persistent stuttering. *NeuroImage: Clinical*, 38, 103413.
- Colclaitt, B. M., Merullo, D. P., Konopka, G., Roberts, T. F., & Brainard, M. S. (2021) Cellular transcriptomics reveals evolutionary identities of songbird vocal circuits. *Science*, 372.
- Colclaitt, B. M., Tachibana, R. O., Cooper, B. G., Halverson, R. H., R. J., Kojima, S., Sober, S. J., & Roberts, T. F. (2019) Transitioning between preparatory and precisely sequenced neuronal activity in production of a skilled behavior. *eLife*, 8, e43732.
- Jarvis, E. D. (2004) Learned birding and the neurobiology of human language. *Annals of the New York Academy of Sciences*, 1016, 749-777.
- Jarvis, E. D. (2007) Neural systems for vocal learning in birds and humans: a synopsis. *Journal of Ornithology*, 148, 35-44.
- Miró, J. M., Dulcis, J., & Cooper, B. G. (2022) Preparing to sing - Respiratory patterns underlying motor readiness for song. *Journal of Neurophysiology*, 128(6), 1646-1662.