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This document is part of a collection that serves two purposes. First it is a public archive for data and documents resulting from evolutionary, ecological, and behavioral research conducted by the Ketterson-Nolan research group. The focus of the research is an abundant North American songbird, the dark-eyed junco, *Junco hyemalis*, and the primary sources of support have been the National Science Foundation and Indiana University. The research was conducted in collaboration with numerous colleagues and students, and the objective of this site is to preserve not only the published products of the research, but also to document the organization and people that led to the published findings. Second it is a repository for the works of Val Nolan Jr., who studied songbirds in addition to the junco: in particular the prairie warbler, *Dendroica discolor*. This site was originally compiled and organized by Eric Snajdr, Nicole Gerlach, and Ellen Ketterson.

Context Statement

This document was generated as part of a long-term biological research project on a songbird, the dark-eyed junco, conducted by the Ketterson/Nolan research group at Indiana University. For more information, please see IUScholarWorks (https://scholarworks.iu.edu/dspace/handle/2022/7911).

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Female release of luteinizing hormone (LH) in response to short- and long-range song in a songbird, the dark-eyed junco (*Junco hyemalis*)



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Introduction

- Male courtship signals affect female reproductive physiology and condition.¹
- In songbirds, hearing male song is known to activate HPG-axis leading to sex steroid production and follicle development in females.^{1,2}
- High-amplitude (loud) songs, also known as long-range songs (LRS), have been the focus of all research to date.
- Males of many species also sing lowamplitude (quiet) songs or short-range songs (SRS).
- SRS can be directed exclusively towards females during courtship and are often strikingly divergent in structure from LRS.³

Methods Long-range Song (LRS) or Short-range Song (SRS) 45-min playback + 15-min silence Hypothalamus GnRH Pituitary LH, FSH Sex Steriods

- Free-living females, early breeding season
- LRS, SRS, or Silent Control
- Blood Sample after 60-min stimulus
- Plasma assayed for Luteinizing Hormone (LH)

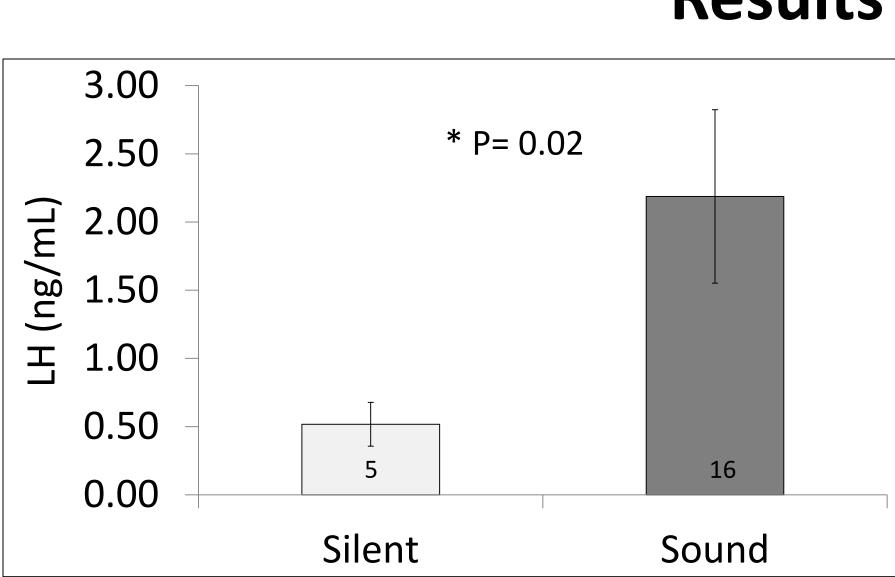
ABSTRACT

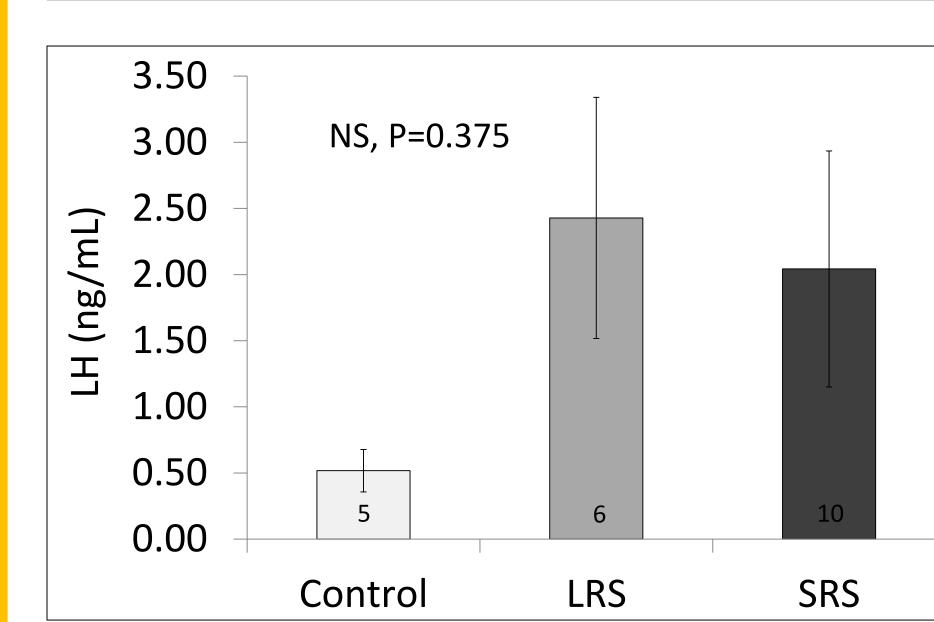
Male courtship signals and displays can have strong stimulatory effects on female reproductive physiology. In songbirds, males use vocal signals, or songs, both to attract and stimulate females into a copulation or lasting pair bond. Previous research has shown that hearing male song can activate the hypothalamic-pituitary-gonadal (HPG) axis leading to the production of sex steroids that drive reproductive development in both sexes. However, studies of the physiological impacts of male song have focused exclusively on high-amplitude, long-range songs (LRS) that project over long distances and overlooked low-amplitude, short-range songs (SRS) that occur during directed courtship. To test the effect of SRS on female physiology, we captured free-living, female dark-eyed juncos (*Junco hyemalis*), brought them into captivity and played 45 minutes of LRS or SRS, or left them in silence as a control. After an additional 15 minutes of silence, we collected plasma samples that were later assayed for luteinizing hormone (LH), the hormonal link between the pituitary gland and the gonads in the HPG axis. When focusing only on females with LH levels detectable by the assay, females hearing song had significantly higher circulating LH than controls. Also, a higher proportion of females hearing SRS exhibited LH levels above the minimum detection limit of our assay than did females receiving LRS or the silent control. These results suggest that females can rapidly elevate their circulating LH in response to male song and highlight the potential for SRS to elicit a larger release of LH and possibly sex steroid production than LRS. We conclude by arguing that future research on male vocal signals should focus on both LRS and SRS.

Research Questions

- 1. Do female dark-eyed juncos upregulate their HPG-axis in response to male song?
- 2. Do females upregulate differentially to long-range song (LRS) or short-range song (SRS)?

Results





Treatment		
Control	LRS	SRS
4	7	4
5	6	10
9	13	14
	Control 4	Control LRS 4 7 5 6

Females receiving sound (LRS or SRS) had significantly higher circulating LH than females left in silence.

Females did not detectably differ in circulating LH levels between treatments.

SRS elicited detectable
LH levels in a higher
proportion of females
than did LRS or
silence.

Discussion

- Female juncos can upregulate circulating
 LH rapidly in response to male song
- Lack of detectable difference between silence and playback birds may be due to
 - Small sample size
 - Variable Reproductive Condition of subjects (no brood patch to full brood patch)
 - Stress of capture
- Both LRS and SRS appear to elicit LH release and SRS may be a more potent activator of the HPG axis than LRS.
- Results highlight the importance of both SRS and LRS in studies focusing on female preference for male courtship signals.

Future Directions

- Replicate with captive birds and a larger sample with repeated measures design.
- Measure female behavioral preferences for each song type.
- Is a visual stimulus necessary to activate the HPG axis? Possibly augment response?



Literature Cited

- 1. Bentley et al., 2000. Horm. Behav. 37: 179-189
- 2. Kroodsma, D.E., 1976. Science 192: 574-575
- 3. Reichard et al., 2011. Am. Nat. 178: 478-487

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