## Supplementary Information

## Inhibition within a premotor circuit controls the timing of vocal turntaking in zebra finches

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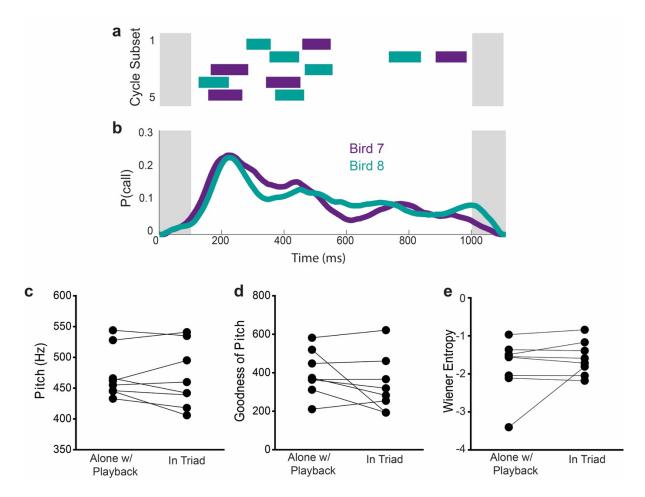
## This PDF file includes:

Supplementary Figs. 1–4

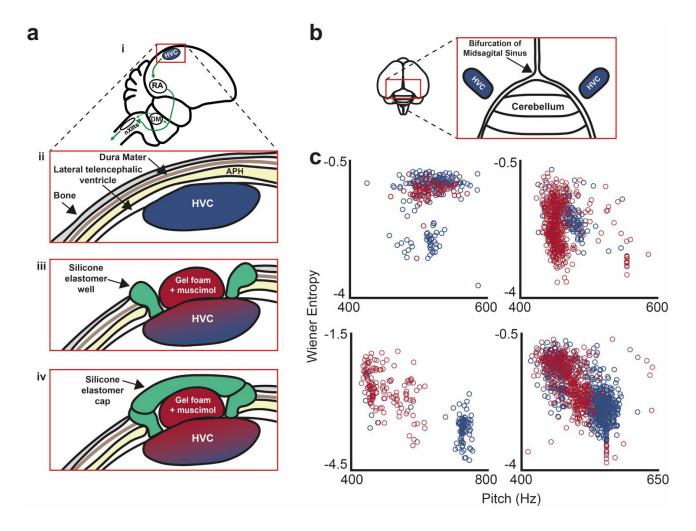
Captions for Supplementary Video 1

Other Supplementary Materials for this manuscript include the following:

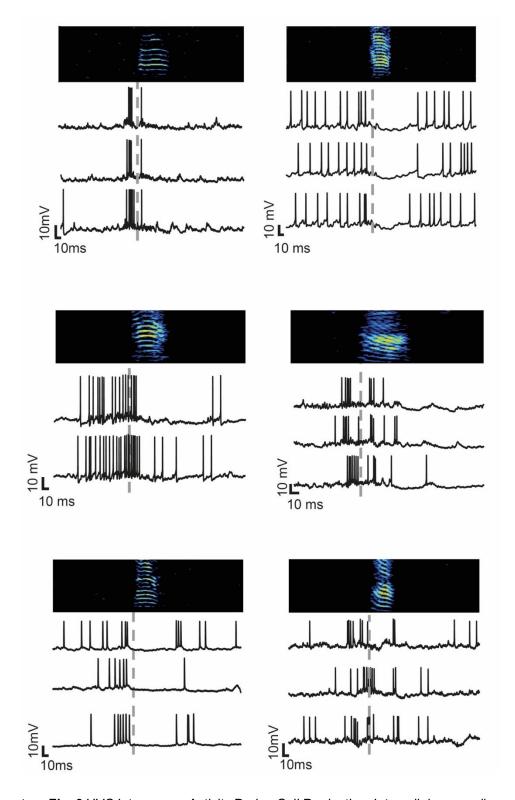
Supplementary Video 1



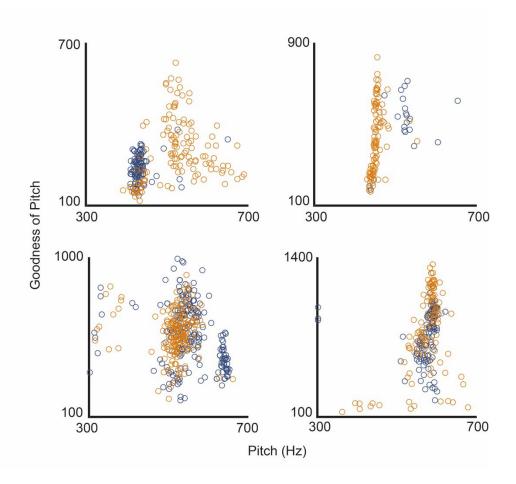
**Supplementary Fig. 1** Call Coordination and Call Acoustic Structure in Vocal Triads. **a** A subset of sequential call cycles demonstrating an alternating response pattern between Bird 7 (purple) and Bird 8 (turquoise) during the vocal triad condition. **b** Call response probability distributions for Birds 7 and 8 for a full 10-minute session when in triad with call playbacks (in grey). Peak response latencies remain similar, although the pair's alternating call sequences result in lower than expected simultaneous calling. **c** Mean call pitch for n = 8 birds while alone with playbacks vs. in the triad condition (Alone w/playbacks = 472 ± 40 Hz, in triad = 467 ± 51 Hz, Wilcoxon sign rank test, p = 0.4609). **d** Harmonic structure of calls as measured by mean Goodness of Pitch for n = 8 birds while alone with call playbacks vs. in the triad condition (mean (alone w/playback) = 397 ± 118, (in triad) = 337 ± 146, Wilcoxon signed rank test, p = 0.3125). **e** Average Wiener Entropy of calls for n = 8 birds while alone with call playbacks vs. in triad condition (mean(alone w/playback) = -1.81 ± 0.74, (in triad) = -1.59 ± 0.45, Wilcoxon signed rank test, p = 0.8438). Note: Each bird's average was calculated from 54-243 calls for all acoustic features.



Supplementary Fig. 2 a) Schematic of pharmacological infusion from the dorsal surface of HVC. i) Illustration of sagittal section of zebra finch vocal motor pathway (RA:robust nucleus of the arcopallium; DM: dorsomedial part of the intercollicular nucleus; nXIIts: 12<sup>th</sup> cranial (tracheosyringeal) nerve. ii) Bone, dura mater, APH (area parahippocampalis), and the lateral telencephalic ventricle are situated above HVC. iii) A craniotomy is cut, dura mater and APH are resected, and a silicone elastomer well is created around the perimeter of the craniotomy. A gel foam sponge soaked in 40°C 5mM muscimol is placed directly on the surface of HVC. iv) A silicone elastomer cap seals the well and the animal is returned to the recording chamber for behavioral testing. b) Schematic of a dorsal view of the zebra finch brain, indicating positions of bilateral craniotomies above HVC. c) Acoustic Features of Calls During Control and Muscimol Conditions. Changes in acoustic features of calls for 4 birds during infusion with musicmol (red) vs. saline controls (blue, as in Fig. 2g); entropy(control) = -2.52 ± 1.13, entropy(muscimol) = -2.21 ± 0.79, 2d-Kolmogorov-Smirnov test ¹, p < 0.01 Bonferroni correction).



**Supplementary Fig. 3** HVC Interneuron Activity During Call Production. Intracellular recordings of six HVC interneurons during multiple renditions of call production. Dotted grey lines indicate onsets of calls.



**Supplementary Fig. 4** Acoustic Features of Calls During Control and Gabazine Conditions. Changes in acoustic features of calls for 4 birds during infusion with saline controls (blue) vs. gabazine (orange), (as in Fig. 5g), (2d-Kolmogorov-Smirnov test <sup>1</sup>, p < 0.01 Bonferroni correction (4/5 birds)).

## References

1. Peacock JA. Two-Dimensional Goodness-of-Fit Testing in Astronomy. *Mon Not R Astron Soc* **202**, 615-627 (1983).