

# The role of Cl<sup>-</sup> channel-inhibition in the brain on the phonotactic selectivity of female crickets

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## ABSTRACT

The goal of this project is to determine if picrotoxin (PTX) has an effect in the supraesophageal ganglion (the location of the secondary and tertiary auditory interneurons) of female crickets, *Acheta domestica*. Females are pretested for phonotaxis on a non-compensating treadmill over a range of syllable periods. Following exposure of the supraesophageal ganglion, 9.2 nL of either saline (control) or 10<sup>-5</sup> M picrotoxin is nano-injected into the supraesophageal ganglion of females. Post-tests following saline injection (10 min later) are similar to the pre-tests. Females treated with picrotoxin become more phonotactically selective preferring the shorter syllable periods.

## INTRODUCTION

Phonotaxis of female crickets toward the calling songs of males is controlled by a number of neurotransmitters and modulators. Picrotoxin, a non-selective blocker of Cl<sup>-</sup> channels, causes unselective females to become selective to the syllable period of male calling songs when nano-injected into the prothoracic ganglion (the location of the primary auditory interneurons). In addition to the prothoracic ganglion, the supraesophageal ganglion has also been shown to be involved in phonotaxis (Pires and Hoy, 1992; Schildberger, 1984). Markovic (2010) nano-injected juvenile hormone III (JHIII) into the supraesophageal ganglion (Fig. 1) and found that unselective females became more selective post-injection. The goal of this project is to determine if picrotoxin has an effect in the supraesophageal ganglion (the location of the secondary and tertiary auditory interneurons) of female *Acheta domestica*.

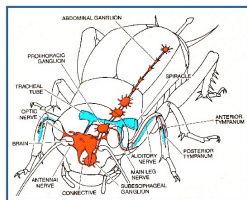


Fig. 1

## METHODOLOGY

- Used 20-32 day old virgin female crickets of *Acheta domestica*.
- Performed pre-tests on a cricket treadmill (Fig. 2) while playing randomized order of male calling songs
- Recorded the phonotactic response of females using Optical Kugel software (Fig. 3).
- Dissected crickets after pre-test by cutting out a flap on the top of their heads in order to expose the supraesophageal ganglion (brain).
- Nano-injected 9.2 nL of either saline (control) or 10<sup>-5</sup> M picrotoxin (experimental) into the brain of the crickets (Fig. 4).
- Performed post-tests identical to the pre-tests.
- Used a paired two-tailed *t*-test to analyze the results of the nano-injection.



Fig. 2

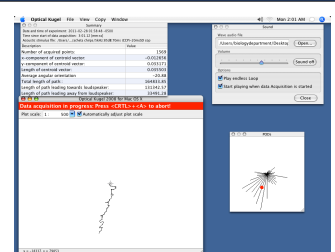
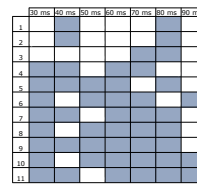


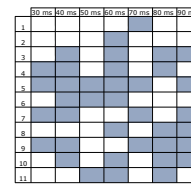
Fig. 3

## RESULTS

### Saline Pre-Test

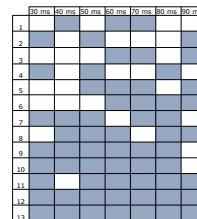


### Saline Post-Test

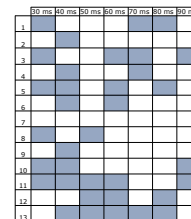


Results indicate that nano-injection of saline into the supraesophageal ganglion of female crickets has no significant effect ( $t=2.09, p=0.064, d.f.=10$ ).

### PTX Pre-Test

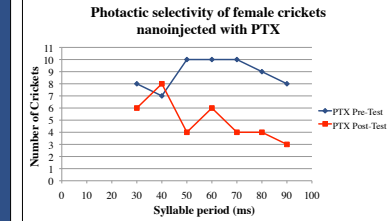


### PTX Post-Test



Results indicate that nano-injection of PTX into the supraesophageal ganglion of female crickets has a significant effect ( $t=3.96, p=0.002, d.f.=12$ ).

## RESULTS Cont'd



Increased selectivity caused by PTX results from decreased responsiveness to longer syllable periods

## CONCLUSIONS

- Picrotoxin causes female crickets to become more phonotactically selective.
- Females prefer shorter syllable periods after nano-injection of picrotoxin.
- Inhibitory inputs in the supraesophageal ganglion dependent on Cl<sup>-</sup> channel inhibition are involved in modulation of selective phonotaxis.
- These findings do not match the changes that occurred with JHIII in the supraesophageal ganglion (Markovic, 2010) or the changes observed from nano-injecting PTX and JHIII into the prothoracic ganglion (Atkins et al., 2008). Thus, these results suggest a different mechanism for controlling phonotactic selectivity in the brain compared to the prothoracic circuit.

## LITERATURE CITED

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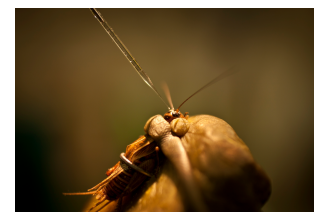


Fig. 4