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Factors Affecting Phonotaxis in Male House Crickets, Acheta domesticus

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Abstract:

This research addresses some of the possible factors that affect male *Acheta domesticus* phonotactic responses to calls of males of the same species. The study examines cricket age and social interaction with females as possible factors affecting this behavior. Through Friedman rank test analysis, the study attempts to demonstrate a link between these factors and the responsiveness of the male crickets in the study. As the crickets aged, they responded to a wider range of syllable periods, which may imply that they follow a similar pattern of decreasing selectivity in their response with age as their female counterparts. A Friedman's rank test was done on the data to determine whether there was a statistically significant difference within any of the syllable periods as the crickets aged. This test showed slight significance in the syllable periods 50ms and 90ms. Socialization with females had minimal effects on male phonotactic behavior.

Introduction:

The goal of this study was to investigate the factors that influence male *Acheta domesticus* phonotactic responses to conspecific male calls. I examined the effects of age and social interaction with females on the males' phonotactic responses to calls of other males of the same species (conspecific males). Hopefully this research, which is part of an ongoing examination of cricket neurobiology studies, will complement similar studies involving female *Acheta domesticus* also done by other members of the neurobiology team in the Biology department at Andrews University (Walikonis et al. 1991). An understanding of the factors that influence male responses to conspecific males will lay the foundation for further investigations and analysis of brain cells that encode these calls. Since there is very little work done to document the relative importance of different features of the male call in *A. domesticus*, my work has been largely

exploratory. I analyzed the by applying statistical tests such as the Friedman rank and chi – square tests. By investigating factors that influence the phonotactic behavior of the male *Acheta domesticus*, I hope to advance knowledge in the field of neuroethology.

Materials and Methods:

Newly molted male crickets were maintained in a separate bin and fed cricket chow and water *ad libitum*. A 12/12 dark-light cycle was maintained using a timer and the temperature was set at room temperature (20 to 25 degrees Celsius). I then tested male responses to conspecific male calls in the cricket arena. For this experiment I am using the cricket arena in order to simulate a more authentic environment (closer to nature) where the cricket can actually move around rather than on the cricket treadmill where the cricket does not actually move or get closer to the calling speaker. I tested a range of syllable periods (30ms, 40ms, 50ms, 60ms, 70ms, 80ms, and 90ms). The normal syllable periods for female phonotactic response are from 50ms to 70ms, but we are including the other syllable periods which are considered less desirable to females in order to see if males prefer different syllable periods. The test protocol involved releasing two male crickets into the arena per trial and allowing 5 minutes of acclimatization to the arena before beginning to play the male call that had been synthesized electronically. The order of syllable period presentation was 50ms, 90ms, 70ms, 40ms, 60ms, 30ms, and 80ms. The calls were presented for 3 minutes each with a two minute break between calls for the crickets to rest and readjust to their environment. I tracked the crickets' movement using a video camera mounted on the ceiling directly above the arena and recorded the data by tracing the path taken by each cricket towards the center of the arena (where the speaker presenting the sound was located). I recorded only the paths that ultimately led a cricket to the center. In order for the path to be listed as positive phonotaxis, the cricket's path could not vary more than 60 degrees from its original course to the

speaker. This is to eliminate the events where a cricket would randomly walk to the center of the arena not in response to the call being played.

For the second portion of the experiment, the crickets will be fed water and cricket chow with no restrictions and be kept in a 12/12 dark-light cycle maintained as before in room temperature conditions. The male crickets will be divided in two groups. One group will never be exposed to females, while the other group is kept in a bin along with females. The effect of age will be controlled for by using crickets of the same age in all studies (5-10 days after the imaginal molt). The sample size for this experiment will be 20-30 crickets. As before, I will release two crickets at a time into the large sand arena and allow 5 minutes for them to adjust to the new environment. The calls will be presented in the same order as the previous experiment for 3 minutes each and the crickets will be given 2 minutes between each call to rest. I will again be recording the paths of the crickets by tracing them onto a transparency. The paths will only be recorded if they ultimately lead to the center and they will be judged as positive phonotaxis only if the path varies less than 60 degrees from the initial course towards the center.

I then analyzed the data for the first experiment using a Friedman rank test which is a one-way analysis of variance designed to determine whether any syllable periods are responded to at a significantly higher rate. The data for the second experiment was also analyzed using a Friedman rank test and the chi-square analysis.

Results:

Table 1 below shows the phonotactic responses of seven male crickets model calls of differing syllable periods. The tests were done every other day and the shaded squares indicate 60% or more of the crickets responding positively to the calling song. After performing the Friedman's rank test on the data from the first part of the study, no significant differences were found in syllable periods 30, 40, 60, 70, or 80ms. However, in syllable periods 50ms and 90ms at least two coordinate pairs were significantly different when compared in the test. Cricket responses for these two groups ranged widely. In the 50ms trial the responses ranged from one cricket to seven crickets and in the 90ms trial the responses ranged from one cricket to eight crickets. The results for the Friedman rank tests that demonstrated significance were: 50ms Asymptotic significance = 0.034 and Chi-Square value= 41.845 and for 90ms Asymptotic significance = 0.011Chi-Square=46.691. For the socialization experiment, the results can be seen in Table 2 below. The data was analyzed with a Friedman rank test and a chi-square test. The results of which can also be found in Table 2. The cricket responses are shown visually on the graph in Figure 4.

Table 1

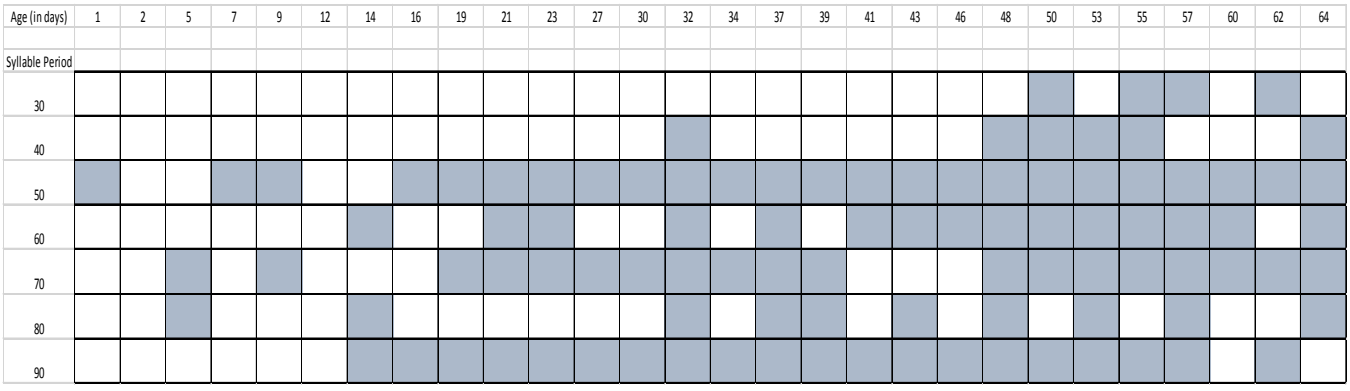


Figure 1

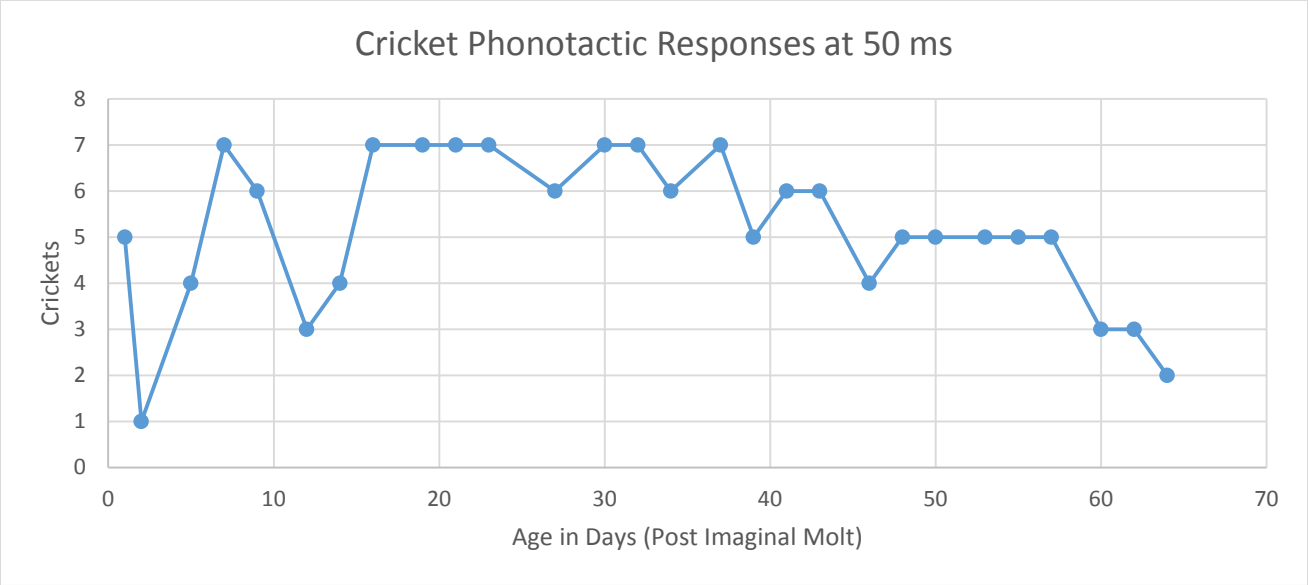


Figure 2

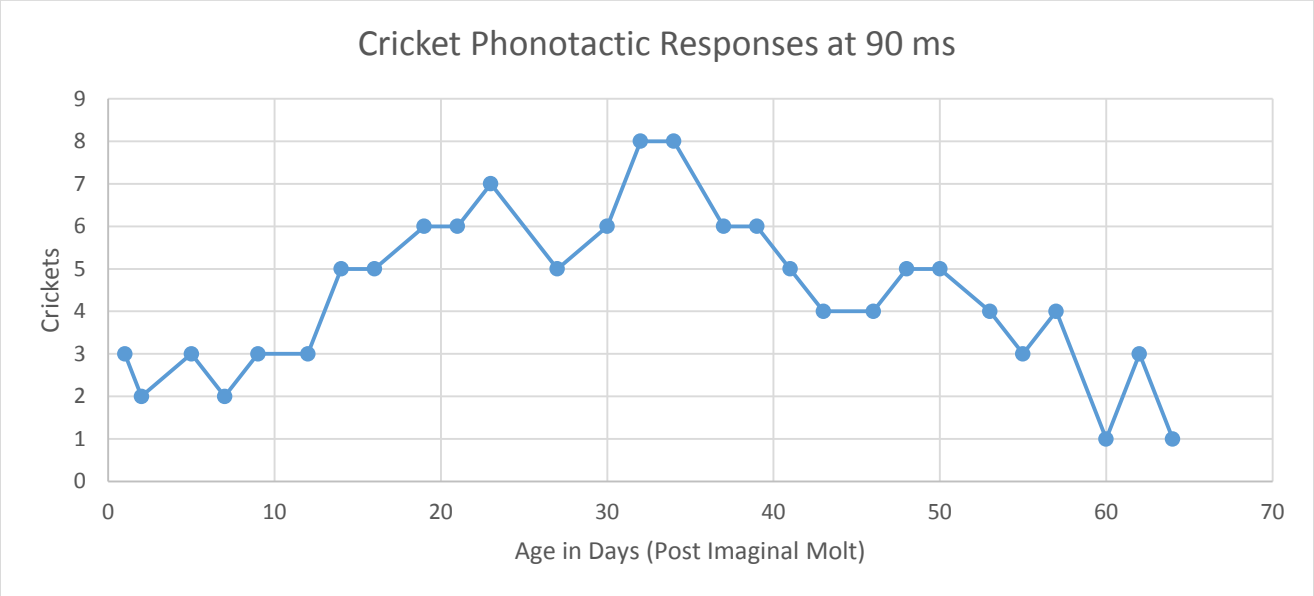


Figure 3

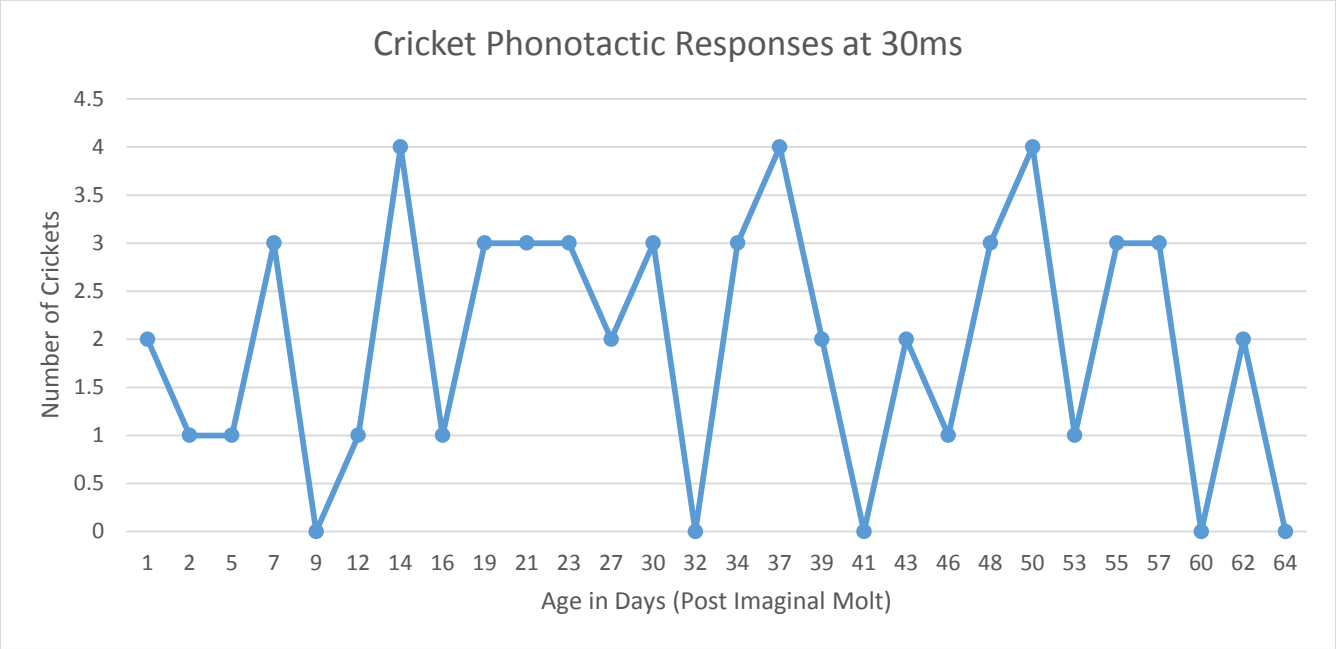
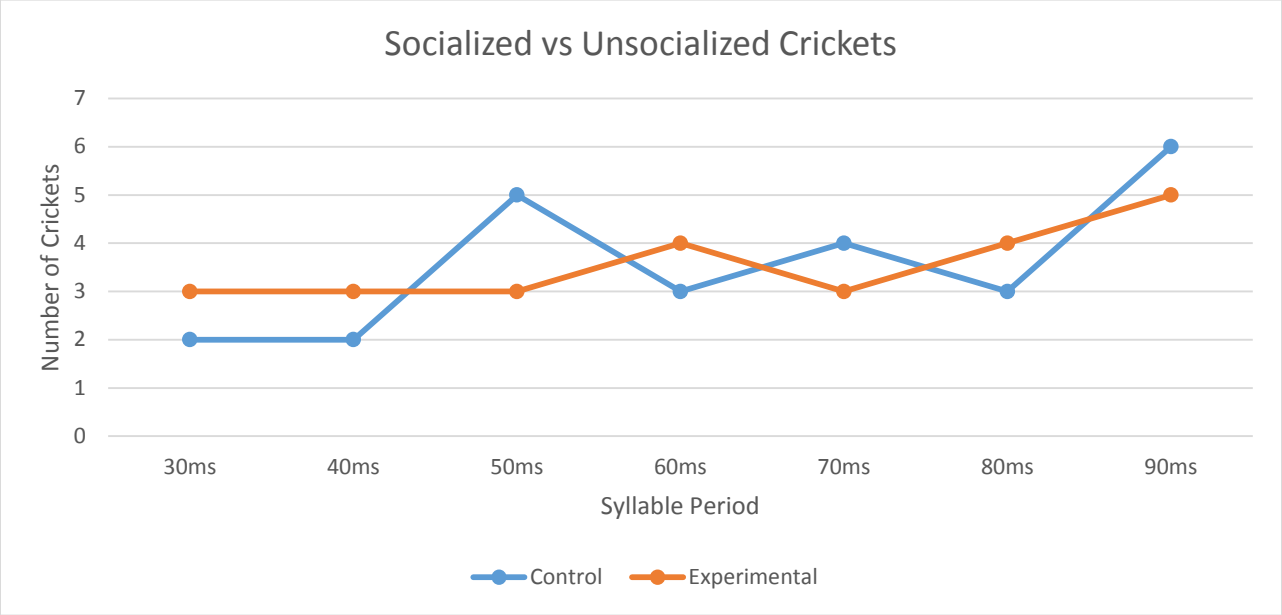


Table 2

	30ms	40ms	50ms	60ms	70ms	80ms	90ms
Control N=10	2	2	5	3	4	3	6
Experimental N=10	3	3	3	4	3	4	5
X^2	.27	.27	.83	.22	.22	.22	.20
df	1	1	1	1	1	1	1
p	.61	.61	.36	.64	.64	.64	.65

Figure 4



Conclusions:

The first glance at the results appears to indicate that there is no significant link between age and phonotactic responses in male *Acheta domesticus* for syllable periods other than 50ms and 90ms. However, this could be due to the small sample size used for this experiment. One of the reasons all the graphs seem to decrease drastically near the end of the experiment is not because cricket responsiveness waned, but rather because some of the cohort of crickets began to die off. This can be seen by examining Table 1 to see that while the numbers on the graphs decrease, the amount of times more than 60% of the crickets responded actually increases. The Friedman's rank test shows that there is no statistical significance in any of the trials except for some slight significance in the 50ms and 90ms trials. The Friedman's rank test is a one-way analysis of variance and tests whether or not the crickets respond consistently to one syllable period over another over the aging process. As we can see in Fig. 1 there is a large difference in the number

of crickets responding to the call from the second trial to the fourth trial and again from the sixth trial to the eighth trial. These pronounced differences in the number of responding crickets may indicate periods of increased sensitivity to the calling song at 50ms in the aging process. In a trial not exhibiting significant data such as Fig. 3 which represents the 30ms call there is what appears to be a random pattern of responses with little variation from zero to four crickets responding. Again in Fig. 2 the most pronounced pattern emerges. There are appears to be a trend towards more responsiveness from little response early to a pronounced response near midlife for the crickets. The overall trend for the observed crickets seems to be that as the crickets age the selectivity for which call they respond to decreases which is similar to the pattern observed in female *Acheta domesticus.* This overall pattern is most clearly evident in Table 1 as we can see the amount of shaded in squares increases as the age of the crickets in the study increases. As a suggestion for continued research on this topic, I would recommend beginning an experiment much like my experiment 1, but beginning with crickets of varying ages to help better investigate age as a factor in phonotactic responses. I would also suggest expanding the sample size of the experiment to include more crickets so that statistical analysis can be done with greater efficacy. One final suggestion for further study is to investigate the correlation between repeated trials on male *Acheta domesticus* over their life span and determine whether it effects the pattern of responses. This would be done in order to test whether the multiple testing experiences have any effect on the results of the study and help determine whether age is indeed a factor affecting phonotactic responses.

In the socialization portion of this experiment, the results showed no significant difference between males kept in isolation from female crickets and males that were kept with female crickets. The p values were far higher than the .05 needed to demonstrate significance,

but the Friedman rank test did show that learning was unlikely between trials. One possible short-coming of this experiment is that it only investigated the effect of socialization on crickets on one particular age group (10 days old). More research needs to be done with a larger sample size of male crickets of varying ages and variable periods of socialization with females.

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