

Journal Article Review

Aminergic Control of Social Crayfish Agonistic Encounters

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## Introduction

Do winning experiences increase levels of aggression in *Procambarus Clarkii*, while losing decreases aggressive motivation? According to Yuto Momohara et. al, “A previously winning experience increases the winning probability of the next agonistic encounter, whereas a previous losing experience has the opposite effect.” This experiment explores questions such as: Are physically larger crayfish more likely to win their encounters?, Are serotonin and Octopamine responsible for winner/loser effects?, If Serotonin and Octopamine are blocked, will there be an opposite effect? Momohara et. al hypothesized, “If crayfish have a winning experience, then aggression levels increase, while a losing experience has the opposite effect.” Also, If crayfish are induced with serotonin, then there will be a winning effect, and likewise, If crayfish are induced with Octopamine, then there will be a losing effect (both effects being reversed and having opposite effects by adding Mianserin and Phentolamine, respectively).

## Methods

Adult male *Procambarus Clarkii* Girard, obtained from a commercial supplier in Okayama, Japan, were used as test subjects in this experiment. Each subject ranged from 6-9 cm in body length and weighed between 10 and 29 grams. Pharmacological agents were received from Sigma (in St. Louis, MO, USA). Serotonin and Octopamine were used in determining an agent for increased aggression and decreased aggression. Receptor blockers, Mianserin and Phentolamine, were also used in order to observe opposite effects of previously witnessed behavior with Serotonin and Octopamine. As controls, each subject was kept in a 19x33x15 container filled with 10 cm of water, for 30 days. All were fed equally, once a week. All were

exposed to the same 12 hour light/dark cycle. And all were tested for aggression bouts in a dimly lit lab at a consistent 23 degree celsius. Two crayfish, each being different in size, were placed into a 26x38x24 cm container filled half way with water. Day one, the two subjects were placed in a container together. A plastic barrier was used to separate subjects for 10 minutes and then removed, allowing for interaction. Behavior was recorded via video. Subjects were first tested on how physical size difference induces agonistic bouts. Day two, there was a second pairing of naive large vs naive small, dominant large vs dominant small, and subordinate large vs subordinate small. Later, these baseline recordings were used for comparison purposes when adding Serotonin and Octopamine to crayfish, to induce agonistic bouts. The treatment consisted of biogenic amines, serotonin creatinine sulfate complex (5-HT), octopamine hydrochloride (OA) and precursors, 5-HTP, tyramine hydrochloride (TA) and the receptor antagonists, mianserin hydrochloride, phentolamine hydrochloride, epinastine hydrochloride, serotonin reuptake inhibitor, and fluoxetine hydrochloride.

## Results

In pairings based on size difference alone, the larger crayfish won in 19 of 23 bouts. There was no significance in the outcome of agonistic bouts with subordinates, however. During antagonistic bouts, 23 pairings of naive large crayfish were beat by dominant small crayfish 70% of the time. Effects of serotonin injections in small crayfish resulted in 63% winnings against larger subjects (10 of 16 times). The probabilities of 1  $\mu$  M and 2  $\mu$  M of serotonin injections causing winning in small crayfish significantly increased when compared to saline injected small subjects. A significant decrease in winning probability was observed when 1  $\mu$  M octopamine

was injected in large, naive animals. When Mianserin, a serotonin blocker, was injected, it decreased winnings in former, dominant winners. The effects of octopamine blocker, phentolamine, when administered in 25  $\mu$  M, cancelled the effects of losing, as observed in previous agonistic bouts of the same subjects ( $p=0.018$ ). The winner effect of the small, dominant crayfish was ( $p=1.0$ ). Overall, serotonin induced aggressive bouts, while octopamine inhibited aggressive bouts.

## Discussion

The data collected reflects remarkable resemblances to other studies, which were seeking to answer the same or similar questions. The authors of this paper discuss the results in context with the hypothesis. Each scenario was tested for the null hypothesis and was discussed in detail the outcome of each result. The hypotheses were supported with great data and statistics. Previous research runs parallel with the findings in this study, as we know that size of crayfish and dominance correlate. This experiment provided sufficient evidence and validity for past research of the same nature. Other research has suggested that serotonin creates dominant behavior in crustaceans and octopamine creates submissive behavior, as witnessed in Momohara et al's experiment. It would be interesting to know if these results are analogous in humans. Certain limitations such as statistics not being significant could hinder the reputation of this research. However, most of the data indicated a difference. Time limits could be one factor that may be manipulated next time this particular experiment is tested. One new question may be, "If time of observation is extended in crayfish subjects, will there be a change in levels of aggression?" Momohara et al's results answer the big question by establishing baseline

recordings of aggressive bouts in untampered with crayfish and then comparing those results with new and manipulated data, such as levels of aggression in bouts where serotonin and octopamine were injected into subjects. This project is beneficial to the literature world because it adds to the data already acquired in similar experiments and also provides us with a sense of how our own brains work.

### Bibliography

Momohara Y, Kanai A, Nagayama T (2013) Aminergic Control of Social Status in Crayfish Agonistic Encounters. PLoS ONE 8(9): e74489. doi:10.1371/journal.pone.0074489