A COMPARISON OF FEEDING BEHAVIOR WITH VARYING TYPES OF PREY THE PONTO-CASPIAN INVADER, *HEMIMYSIS ANOMALA*

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August 22, 2014

Abstract

Hemimysis anomala*, a native of the Ponto-Caspian region, has been discovered in the Laurentian Great Lakes in recent years. As such, it is important to gain an understanding of the behavior of this organism in order to predict the effects it could have on new ecosystems. In this experiment, small and large* Daphnia sp., Artemia *nauplii, & a mixture of wild-caught calanoid copepods were used in a basic feeding experiment under visual observation for comparisons of behavior and preference with different prey. It was found that male* Hemimysis *attacked and captured their prey more frequently than females, while no other behavioral differences were found between the two sexes. Additionally,* Hemimysis *encountered small* Daphnia *more frequently on average, but were more likely to attack and consume calanoid copepods. This preference for calanoid copepods above the other prey types used could be somewhat of an indication of the impacted native communities of the Laurentian Great Lakes.*

Introduction

Since the globalization of the world economy, oceangoing vessels have travelled across the waters of the world, delivering goods to areas near and far. These ships, however, also deliver an unwanted entity: foreign organisms that have the potential to invade ecosystems. Such is the case of the Laurentian Great Lakes, an area that has been riddled with foreign invaders. Of these invaders, one of the most recent is the crustacean zooplankter *Hemimysis anomala*, first discovered in these North American waters in 2006 (Pothoven et al. 2007).

Originally from the Caspian Sea, this alien species has already been an invader in Dutch waters since 1997 and has since destroyed native populations of multiple trophic levels (Ketelaars et al. 1999). In the newly invaded Great Lakes ecosystem, *Hemimysis* is predicted to have a devastating effect yet again (Ricciardi et al. 2011).

In order to fully understand the impact an invasive organism will have on a foreign ecosystem, it is important to understand its behavior and ecology, and such is the case with *Hemimysis anomala* as well. It is well understood that these invaders feed at night (Brown 2011), have had exceptionally high clearance rates reported upon multiple prey items (Branson 2013), and exhibit swarming behaviors (Lafountaine et al. 2012).

Hemimysis has also been reported to exhibit filter-feeding as well as raptorial predation in the wild (Ketelaars et al. 1999). Utilizing two very different methods of feeding makes *Hemimysis* characteristic of the order Mysida, as mysids are typically omnivorous (Mauchline et al. 1980). As with most aquatic organisms, water temperature is a major determinant of feeding activity in *Hemimysis*, and it has been shown that temperature has a large impact on feeding rates (Sun et al. 2013). Additionally, both of the techniques used by *Hemimysis* are not uncommonly observed individually among other crustaceans. A third method of feeding, described only by Branson, called hoarding entailed the predator collecting multiple prey items with non-immediate consumption.

Activity by *Hemimysis anomala* can be broken down into “nonaggressive” and “aggressive” behavior. Nonaggressive behavior refers to actions that do not contribute to the consumption of any sort of prey item, and in this experiment the main nonaggressive behavior addressed is that of passive encounters with prey. Passive encounters consisted of whenever a prey was within striking range or physical contact with the predator without any sort of

aggression exhibited by the predator. Aggressive behavior is broken down into five categories: attacks, misses, captures, escapes, and ingestions. Initial acts of aggression are attacks, and if the prey item is caught by the predator, a capture occurs. However, if the attack fails, it is considered a miss. Once it is captured, if a prey is consumed by the predator, ingestion occurs, but if the prey gets away it is an escape.

Given the difference in prey selection between adult and juvenile *Hemimysis anomala* (Pothoven et al. 2007), it is reasonable to predict that differences will also be present between these two groups in regards to their feeding activity on the same type of prey. As well as a difference between age groups on the same prey, previous experiments have shown *Hemimysis* adults prefer particular prey over others (Borcherding et al. 2006) and as such there will likely be a difference in feeding behavior by adults on a variety of prey types. Additionally, provided the size differences in adult male and female *Hemimysis* (Pothoven 2007), and the role of organism size in behavior (Werner & Gilliam 1984), a difference in feeding behavior between male and female adults is also predicted.

Materials & Methods

Hemimysis Collection & Maintenance

Hemimysis were captured from Muskegon Lake (Muskegon County, Michigan) via tows through visible swarms with a 30 cm diameter, 64µm mesh zooplankton net. Individuals that were obtained via the net were then be placed into 4L plastic containers filled with sampling site water. In order to prevent thermal shock and maintain a relatively consistent water temperature,

the 4L containers were set within a cooler during transportation back to Central Michigan University. Upon arrival to CMU, the specimen containers were allowed to reach room temperature and will subsequently be transferred into a 4 L tank filled with aerated and de-chlorinated tap water for long-term maintenance. The *Hemimysis* individuals were fed Omega One Natural Protein Formula Super Color fish flakes prior to experimental starving.

Prey Culture and Maintenance

Four types of prey were utilized for our experiments. These organisms included small (<500µm) and large (>500µm) *Daphnia sp.*, a mixture of calanoid copepods, and newly hatched *Artemia* nauplii. *Artemia* nauplii were utilized due to the difficulty of capturing, isolating, and maintaining wild copepod nauplii. The *Artemia* nauplii were hatched from freeze-dried eggs in a 25°C, aerated, 25% solution of salt and water (Sorgeloos et al. 2001). Once the nauplii hatched, they were transferred to a 4 L container and fed lab-cultured *Dunaliella.* The calanoid copepods were collected from ponds on Central Michigan University's campus via surface tows with a 30 cm diameter, 64 µm mesh net. In order to separate the desired zooplankton from the tow as a whole, the tow was emptied into a petri dish and desired copepods were separated into group-specific 4 L containers via pipette. Cultured *Daphnia* were fed a maintenance diet of lab-cultured *Scenedesmus*.

Feeding Trial Parameters

All observations will be performed at Central Michigan University with side lighting via a fiber optic lamp while the overhead lights are turned off in order to reduce glare and increase visibility of prey items. Relatively low levels of light have been shown to provide no difference

in behavior between light and dark feeding of *Hemimysis* (Halpin et al. 2013). During all experiments, *Hemimysis* activity will take place in a 22x15x5cm black plastic tray filled with 500 ml of dechlorinated tap water. Prey density in the plastic tray was 40/L for every prey type used. Each experimental trial would last for ten minutes.

Hemimysis activity was recorded during each trial using a mechanical counter to record six feeding-related actions performed by the predator. These actions consisted of "Passive Encounters," "Attacks," "Misses," "Captures," "Ingestions," and "Escapes." When acts of aggression were observed, they were denoted Attacks, and if they prey item was caught by the predator, a Capture would be recorded, while if the Attack failed, it would be labelled as a Miss. Once captured, if a prey was consumed by the predator, that was considered an Ingestion, but if the prey managed to get away from the predator after being captured, it would have Escaped.

For each of the four prey types, ten separate trials were run with both female and male adult (> 5 mm) *Hemimysis* individuals. Due to difficulty maintaining live juvenile *Hemimysis* (< 5 mm long), only seven trials were run using juveniles, all of which involved feeding of newly hatched *Artemia* nauplii. All data was analyzed statistically using Kruskal-Wallis tests on Minitab statistical software.

Results

A total of 40 *Hemimysis anomala* individuals were used in the feeding trials, which totaled 82 separate feedings. Of these trials, 35 were run with males (10 trials with *Artemia* nauplii, 10 with large *Daphnia*, 10 with calanoid copepod mixtures, 5 with small *Daphnia*).

Only five trials were run with small *Daphnia* and adult male *Hemimysis* due to the relatively short lifespan of the mysids in captivity. Forty feeding trials were run with female *Hemimysis* individuals, with ten trials performed for each experimental prey types. As mentioned previously, only seven trials were performed with juveniles due to difficulty in maintaining their health; all seven trials using juveniles utilized *Artemia* nauplii as prey.

When comparing the number of observed passive encounters between the different prey types, a Kruskal-Wallis test revealed that there is a significant difference in how often each prey item was encountered ($P = 0.019$). When looking over the means relating to observed passive encounters, it is evident that small *Daphnia* were encountered less than the other prey types (Fig. 1). Small *Daphnia* mean passive encounters were 24.3 less than those of large *Daphnia,* 18.7 less than the calanoid copepods, and 25.1 less than *Artemia* nauplii. There is not, however, a difference in passive encounters between male and female *Hemimysis* ($P = 0.308$).

There was a very significant difference in the number of attacks on the different prey types ($P < 0.001$), with calanoid copepods attacked more frequently than the other prey items (6.9 more on average than large *Daphnia,* 11.2 more than small *Daphnia,* and 11.65 more than *Artemia* nauplii), as seen in Figure 2. Additionally, there is significance in the difference between the attacks performed by males and females on all prey types ($P = 0.026$), with males averaging one more attack per trial than females. However there is not any significance in the proportion of attacks that became captures between males and females ($P = 0.160$), while there was significance in the number of captures ($P = 0.004$), with males giving the impression of capturing more prey (Fig. 3) and converting attacks to captures more frequently (Fig. 4).

When analyzing the proportion of ingestions that occurred (Fig. 5), there was a significant difference between the prey types ($P = 0.001$), however it appears that the only prey

type that deviates from the rest in this regard is the large *Daphnia*, with the rest of the prey being ingested about the same proportion of the time when captured. Additionally, the proportion of ingestions that resulted from captures did not yield a statistically different value between males and females (P = 0.106).

For analysis of the differences in feeding between the adult and juvenile *Hemimysis* on *Artemia* nauplii, male and female adult *Hemimysis* values were grouped into one category in order to make this comparison. There was no evidence of any significant difference between adults and juveniles for observed passive encounters (P = 0.072). Additionally, there were no differences in attacks (P = 0.069), captures (P = 0.304), proportion of captured attacks (P = 0.252), ingestions (P = 0.977), or percentage of ingested captured prey (P = 0.079), as seen in Figure 6.

Three methods of prey capture were observed to be performed by *Hemimysis anomala* during the experimental trial. Each individual typically utilized one method of capture consistently throughout each trial and was repeated use of that method on all prey types utilized. The first capture method consisted of a typical full-body lunge towards the prey. The second method was more subtle, with the *Hemimysis* individual slowly approaching the prey and nonchalantly grabbing the prey without any dramatic movement of any appendages. Additionally, there was another capture method in which the predator aligned its body more or less perpendicular to the bottom of the tray with its head nearly in contact with the bottom. From there, the *Hemimysis* would use its swimming appendages to create a slow current that gradually dragged prey towards it until the predator could make a minor lunge to capture its prey. Throughout all trials, there was no observed hoarding as described by Branson. The most common method was the first mentioned, with a vast majority of the *Hemimysis* utilizing it.

Discussion

The data show that observed attacks differed between the different prey types, and when looking at the prey types it is possible to speculate as to why this is. Large *Daphnia*, which would be easy for *Hemimysis* to detect, proves a difficult prey to handle in observation, and as such they have the lowest rate of being captured after being attacked among the used prey types. The small *Daphnia*, however, can be speculated to be more difficult to detect given their slow swimming speed, but easier to handle once they are captured due to their smaller size. The same assumptions can be made of the *Artemia* nauplii, although their slightly quicker swimming speed allowed them to be more evasive of the predators in regards to being attacked as well as being captured. It can be inferred that as a result of these same factors that the juvenile *Hemimysis* exhibited statistically the same feeding behavior on the young *Artemia*. The calanoid copepods proved easy for *Hemimysis* to detect and attack, most likely due to their mid-range size and the attraction they create for themselves by swimming more than the Daphnids. Of all the prey types, the large *Daphnia* appeared to be the most difficult to consume merely because of their size, while the rest of the prey were of a size range much more manageable for *Hemimysis.* The low rate of ingestion of the large *Daphnia* found here is similar to what has been reported in literature (Ketelaars et al. 1999).

The low number of observed attacks on both large and small *Daphnia* can lead one to believe that the severe drops in *Daphnia* and *Bosmina* populations reported by Ketelaars et al. were the result of competition for algae, rather than the direct effect of predation upon the cladocerans by the mysids. The major effect of the indirect influence *Hemimysis*, as well as its

apparently less severe direct impact, has on cladocerans can be superimposed on the Laurentian Great Lakes ecosystems. In this scenario, the combination of the indirect effect of *Hemimysis* combined with the direct Daphnid suppression by invasive *Bythotrephes* (Manca & Ruggiu 1998) can be forecasted to have a dire effect on the native zooplankton communities of the affected lakes.

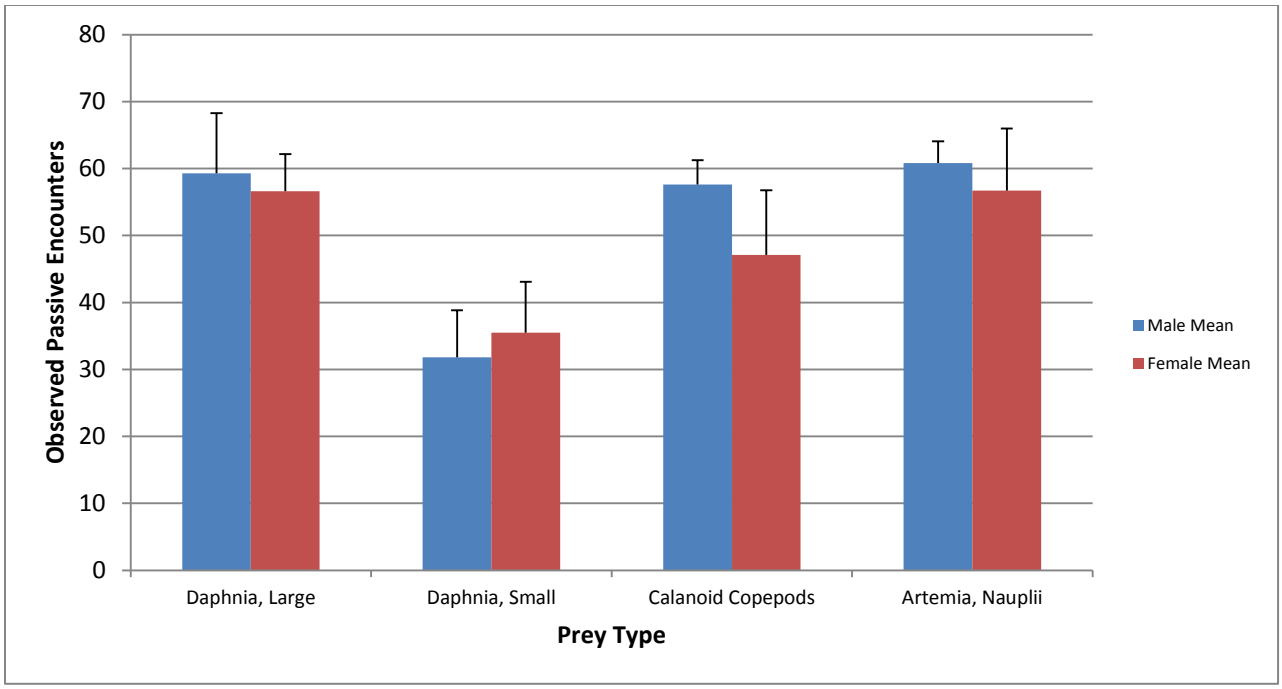

Figure 1. Mean number of Passive Encounters between male and female *Hemimysis anomala* for each experimental prey type used.

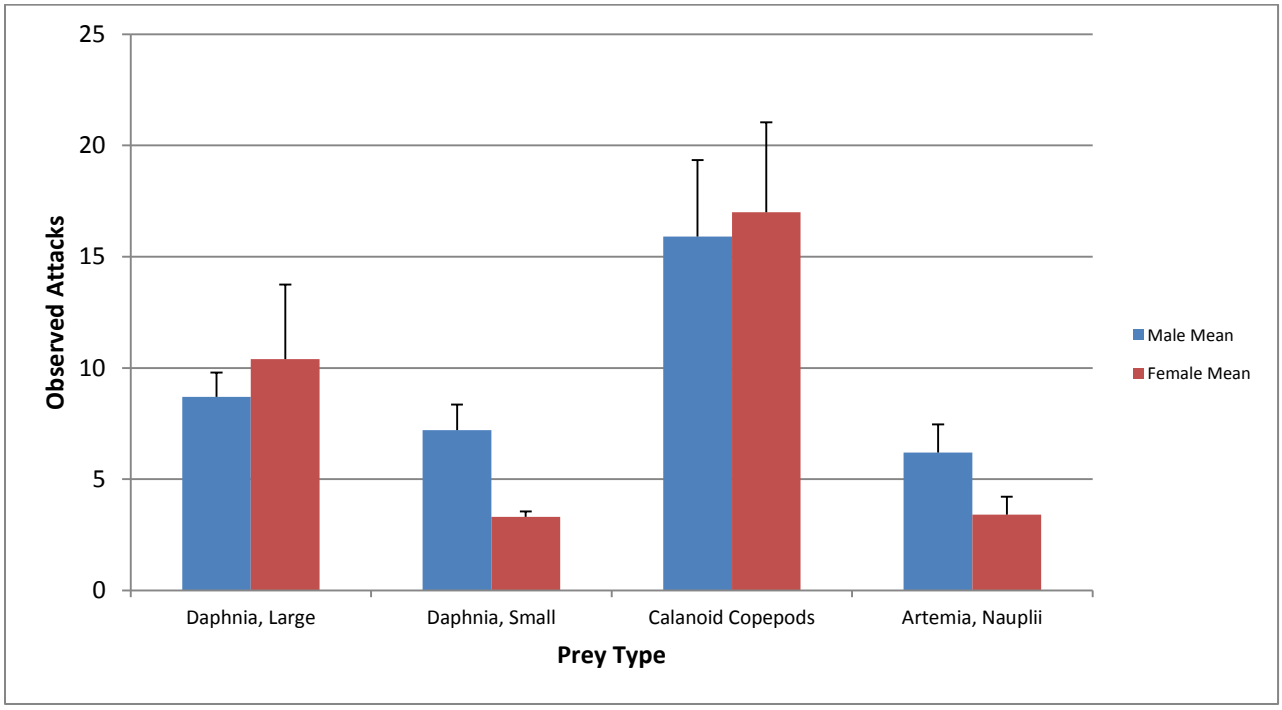

Figure 2. Mean number of Attacks performed by male and female *Hemimysis anomala* upon each experimental prey type used.

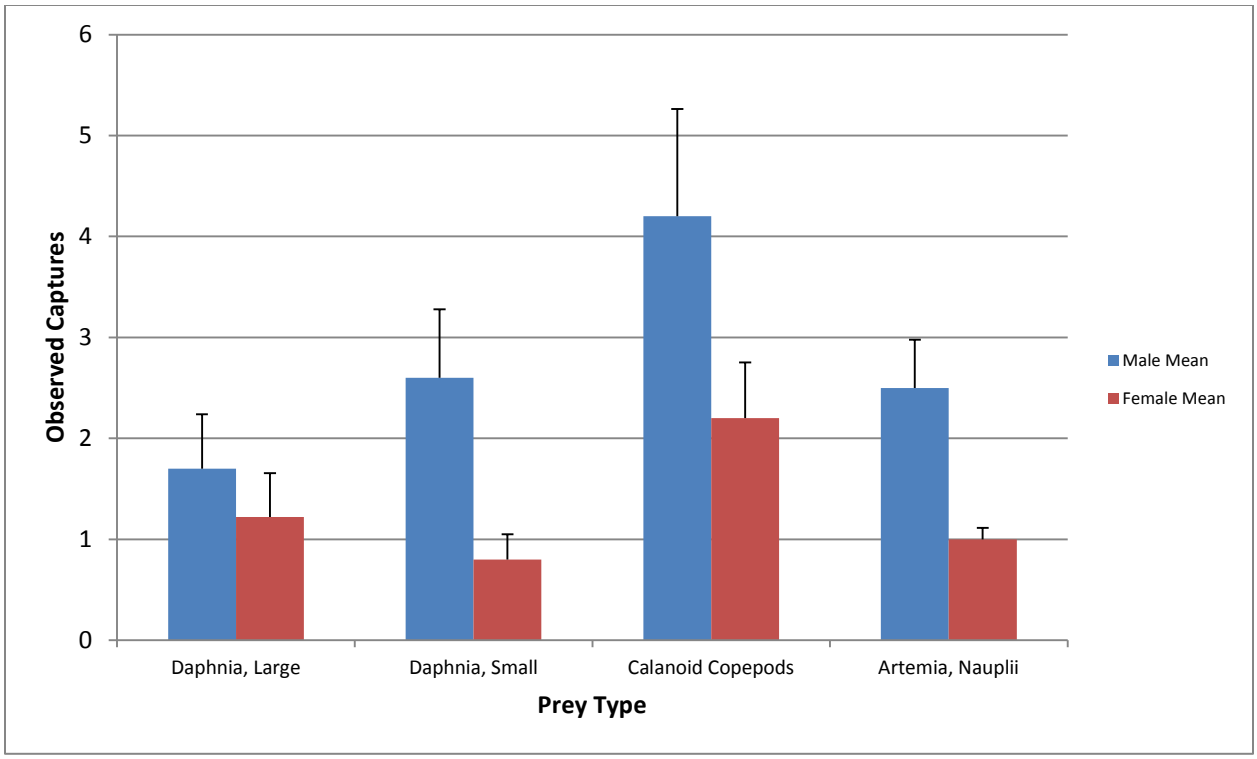

Figure 3. Mean number of Captures performed by male and female *Hemimysis anomala* upon each experimental prey type used.

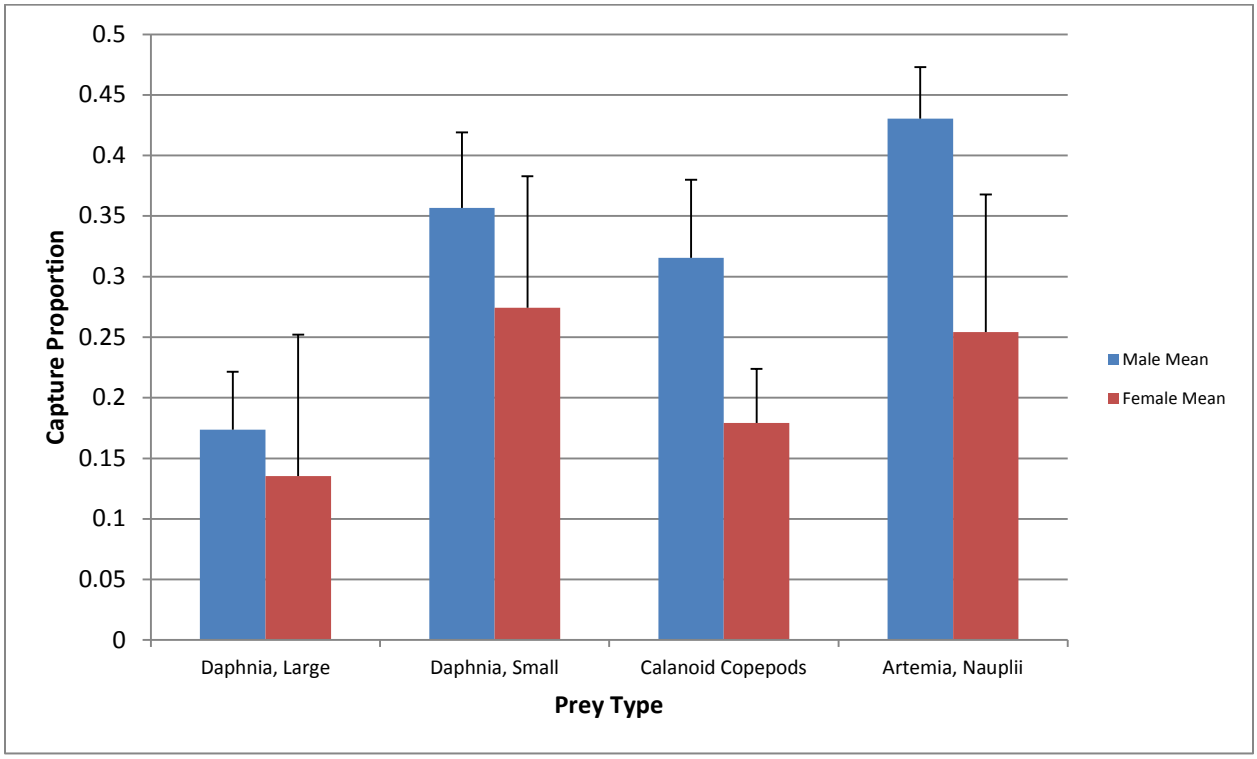

Figure 4. Mean percentage (displayed as proportion) of Attacks that became Captures by male and female *Hemimysis anomala* upon each experimental prey type used.

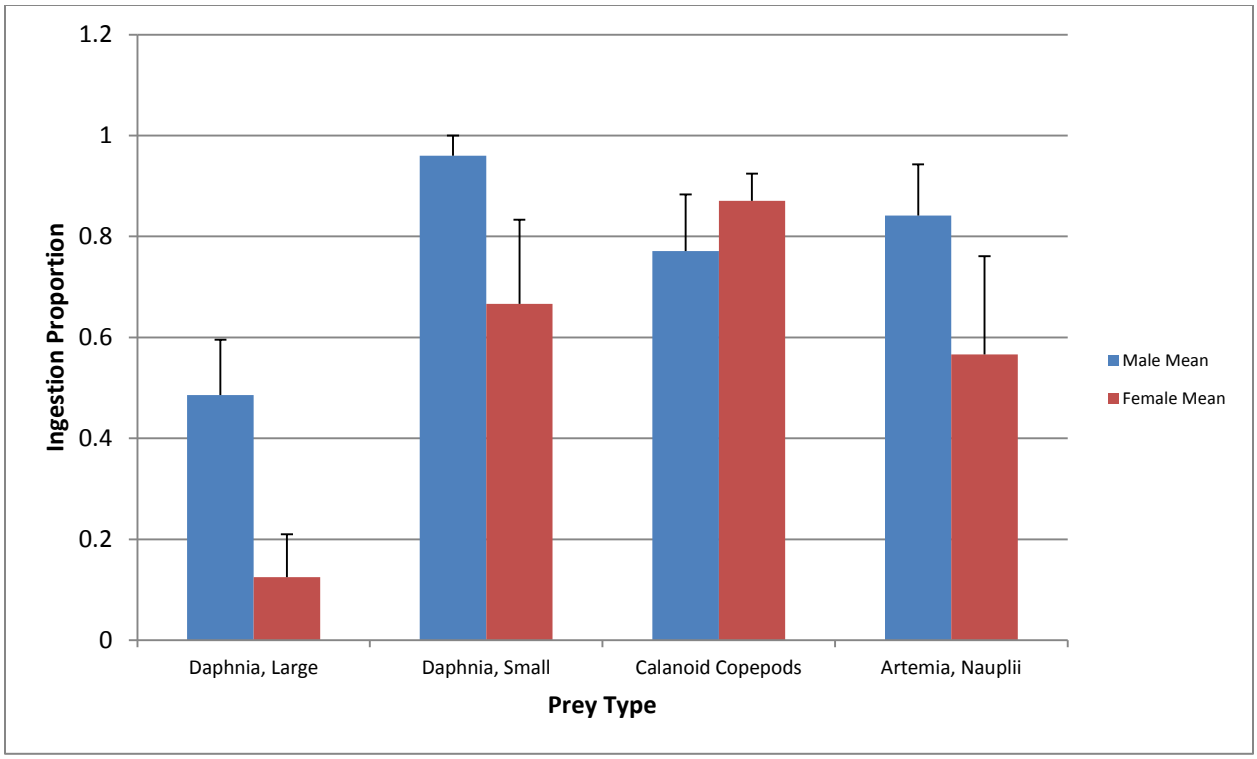

Figure 5. Mean percentage (displayed as proportion) of Captures that became Ingestions by male and female *Hemimysis anomala* upon each experimental prey type used.

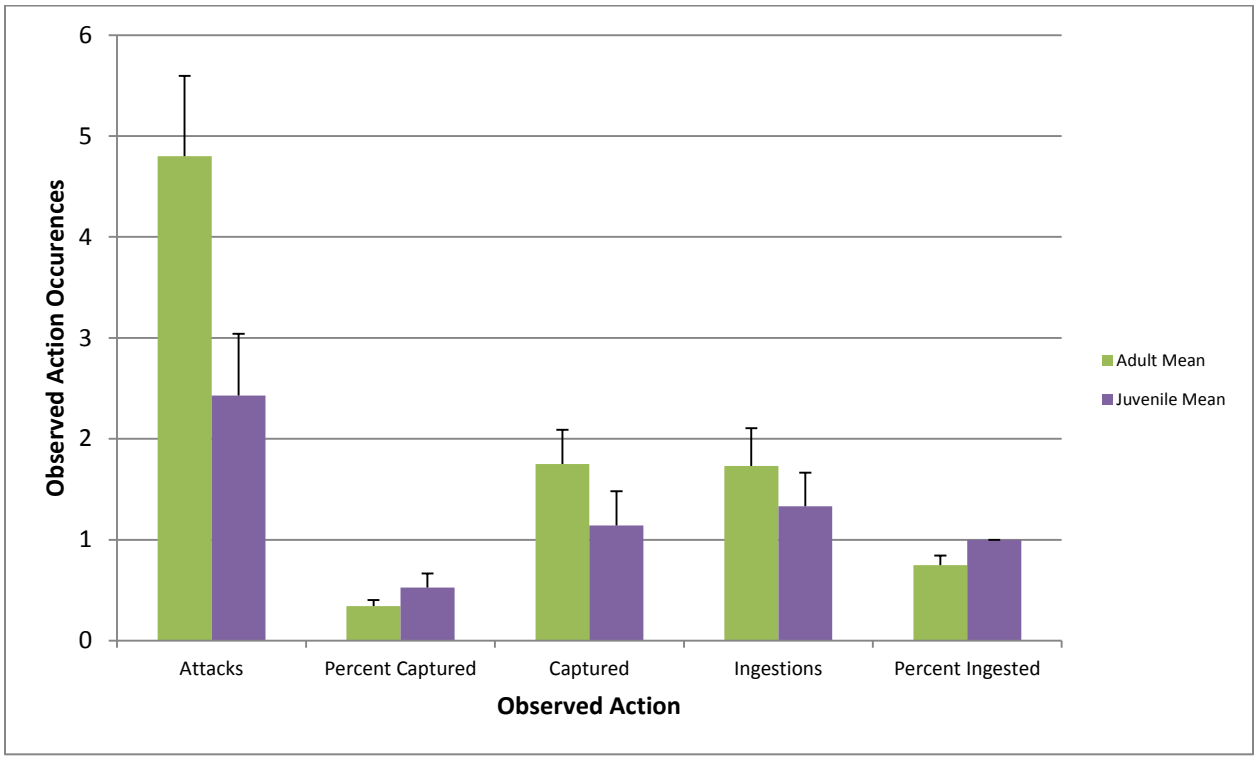

Figure 6. Mean values for recorded observations for adult and juvenile *Hemimysis anomala* with *Artemia* nauplii as prey.

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