Early Life History of the endemic leech *Motobdella montezuma* in Montezuma Well, Arizona

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Image 1. The swallet of Montezuma Well.

Abstract

Montezuma Well, a collapsed travertine spring, is known for its unusually high concentrations of dissolved CO_2, alkalinity and arsenic. There are four species of leeches that inhabit the Montezuma Well, including one endemic species, *Motobdella montezuma*. Although much is known about the predator and prey interactions of this leech, the early life history has not been well documented. Between 2007 and 2010, we documented the numbers of juveniles and cocoons through monthly quadrat sampling of the shallow outflow "swallet" of the Montezuma Well and estimated the mean crowding of these life stages. Cocoon abundance surged in early fall, with juvenile abundance increasing a month after the cocoon peak. There was also spatial crowding of cocoons and juveniles during the peak cocoon and juvenile abundance. Our results provide the first documentation of early life history of *Motobdella montezuma*.

Introduction

Montezuma Well (the Well) is a collapsed travertine spring mound, located in the Verde Valley of north-central Arizona, which is a part of the Montezuma Castle National Monument park service system. The Well is over 100 m wide and approximately 17 m deep and is surrounded by a travertine wall that rises 20 m above the water's surface (O'Brien, Blinn & van Riper III 2002). There is an area called the "swallet", which is a shallow outlet (depth <1m) in the northeast corner that drains water through a series of caves and into Wet Beaver Creek, the most (if only) readily accessible section as the rest of the shore drops steeply into the pelagic zone (Cole & Barry 1973).

The Well is continuously replenished by warm spring water that enters through three deep (>16m) fissures. It is also characterized by its unique chemical properties such as high concentrations of arsenic, dissolved CO_2 and alkalinity (O'Brien, Blinn & van Riper III 2002). Foust et al. (2004) showed that the arsenic is naturally occurring with average levels of 100 µg/L. Dissolved carbon dioxide concentrations are also high with levels commonly greater than 450mg/L. This elevated level is the driving factor behind the absence of fish in the Well (O'Brien, Foust, Ketterer & Blinn 2003). The alkalinity present in the Well is also high in concentrations, with levels of greater than 600 mg/L $CaCO_3$. The average pH of the Well is 6.4 and the water temperature remains within 19-24 degrees Celsius throughout the year (Cole & Barry 1973).

These chemical qualities that exclude fish and isolate Montezuma Well from nearby Wet Beaver Creek are exactly what makes this closed ecosystem so suitable for predaceous leeches. With no resident fish populations, the Well's large invertebrate community has filled those ecological niches, which normally would have been filled by fish. This community includes the four known species of leeches that inhabit Montezuma Well including the endemic pelagic predator of our study: *Motobdella montezuma* (Davies, Singhal & Blinn 1985), *Helobdella triserialis*, *Helobdella elongata*, and *Helobdella stagnalis*, the most common leech in the swallet (Blinn 1989).

Although the early life history of the endemic leech, *Motobdella montezuma*, has not been well documented, extensive studies have been done relating to its behavior and morphology. A behavior of particular interest is that of its predator-prey interactions with the endemic amphipod, *Hyalella montezuma*. This amphipod, due to the fishless environment, has evolved an unusual free-swimming behavior where it migrates to the surface of the water to feed on phytoplankton at dusk. After this initial climb, their primary predator, *Motobdella montezuma*, senses the water waves and follows the amphipods through the water column from the deep sediment of the Well that they inhabit during the day to feed. *M. montezuma* then returns to the pelagic zone after the amphipods retreat into the near shore vegetation (O'Brien, Blinn & van Riper III 2002). The morphology of *M. montezuma* can be characterized by three pairs of eyes (the second and third pairs smaller in size), densely pigmented dorsal stripes, 14-18 small papillae on each annulus that contain the sensory organs, and six pairs of large testicular follicles (3-5 per bunch). *M. montezuma* generally do not exceed 71mm in total length (Davies, Singhal & Blinn 1985).

Even though little is known about their early life history in general, it is known that *M. montezuma* secrete their cocoons through their clitellum after the fertilization of their eggs. This species prefers to place its cocoons at depths below 5m on the stems of the endemic pondweed, *Potamogeton illinoensis*, in order to avoid heavy predation from surface insects (Davies et al. 1987). The cocoon hardens once it has formed and the young develop within until they are ready to hatch. Our study investigated the occurrence of cocoons and juveniles present in the swallet of the Well, which is less than 1m in depth, to contribute further analysis of early life history of *M. montezuma*.

Basic Research

Methods

Each month from November 2007 to December 2010 (except in January 2008, July 2008, and May 2009), we surveyed the population of the endemic leech, *Motobdella montezuma,* in the swallet of Montezuma Well, AZ using a 1 m^2 portable quadrat divided evenly into 25, 20 cm^2 squares. Sampling was accomplished within 18 quadrat positions (although actual amount of positions varied because of areas that could not be sampled due to maneuverability). At each quadrat position within the swallet, we randomly selected five squares without replacement, and identified and counted all of the *M. montezuma* found, noting the life stage of each leech observed (either cocoon, juvenile or adult). After counting five quadrants in one position, we moved the portable transect directly horizontal to the just censored area and repeated the above counting method. Once a row was completed, we moved down toward the opening of the swallet, and counted a new row (Beresic-Perrins 2010).

In order to determine the distribution over space and time of *Motobdella montezuma*, we estimated their mean crowding through quadrat sampling of the swallet of Montezuma Well. Mean crowding can be defined as the "mean number per individual of other individuals in the same quadrat" (Lloyd 1967). Basically, mean crowding estimates the degree that leech individuals are clumped in a particular area, usually due to resources such as food or reproductive mates. Wade (1995) showed that mean crowding could be expressed as $m^* = m + [(V_m/m) - 1$, where m^* represent mean crowding, *m* is the mean density per quadrat and V_m is the variance around *m.* According to Wade (1995), patchiness, P = m*/m, equals the ratio of mean crowding, m*, to mean density, m, and shows how the degree of crowding per individual compares with the degree of crowding per patch. In this study, we measured the relative concentration of *Motobdella montezuma* cocoons and juveniles spatially clumped in a quadrat. If patchiness (*P*) is equal to one, this indicates the individuals of the population are randomly distributed. If *P* is less than one, then the individuals are over dispersed and if it is greater than one, the individuals are considered spatially clumped (Beresic-Perrins 2010; Lloyd 1967; Shuster & Wade 2003).

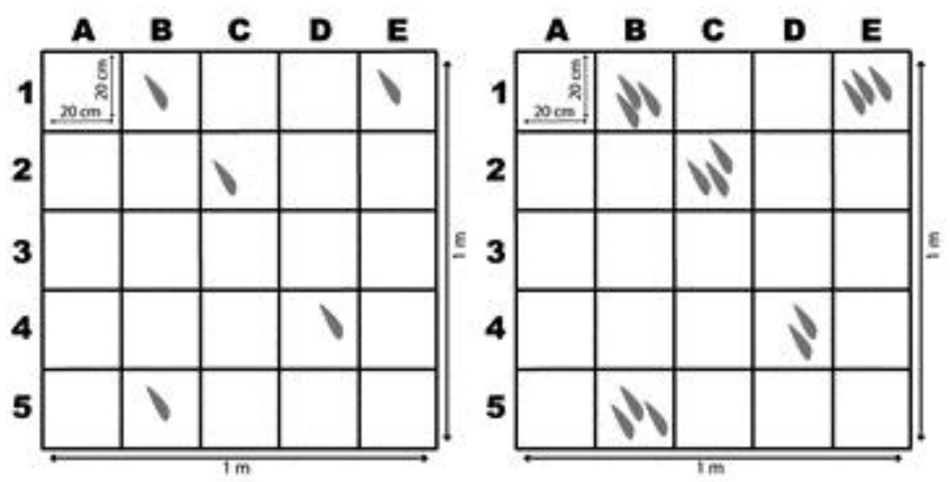

Fig. 1 "Diagram of the portable transect used for the leech surveys. If the leeches were evenly distributed with a patchiness value equal to 0, then a leech would be found sparsely distributed in the quadrats as illustrated in the first diagram. If the leeches were spatially clumped and had a patchiness value greater than 0, more than one leech would be found in a quadrat as illustrated in the second diagram" (Beresic-Perrins 2010).

Image 2. (above) Student counting and identifying leech specimens on rock substrate in the swallet of the Well.

Image 3. (below) Becky and students using the portable transect for leech surveys in the swallet of the Well.

Results

The results of this study were quantified in three graphs, one measuring the total distribution of each life stage (including adults) over the course of the study (Fig. 2), one measuring the abundance of cocoons and juveniles through monthly survey counts (Fig. 3) and the last measuring the patchiness of cocoons and juveniles during each month (Fig. 4). We found that a majority of *M. montezuma* present in the swallet during our quadrat sampling, between 2007 and 2010, were either cocoons or juveniles but not adults. Cocoons were the majority of the population, with approximately 64% of all *M. montezuma* surveyed with juveniles following with approximately 36% of the distribution. Only one adult out of the 560 leeches surveyed was documented in the swallet of the Well throughout the duration of the study. We also found that there was a surge in cocoon abundance in the months of early fall, with juvenile abundance increasing approximately a month after the cocoon peak. Our patchiness data indicated spatial crowding (where patchiness is greater than one) of cocoons and juveniles during these months of peak cocoon and juvenile abundance.

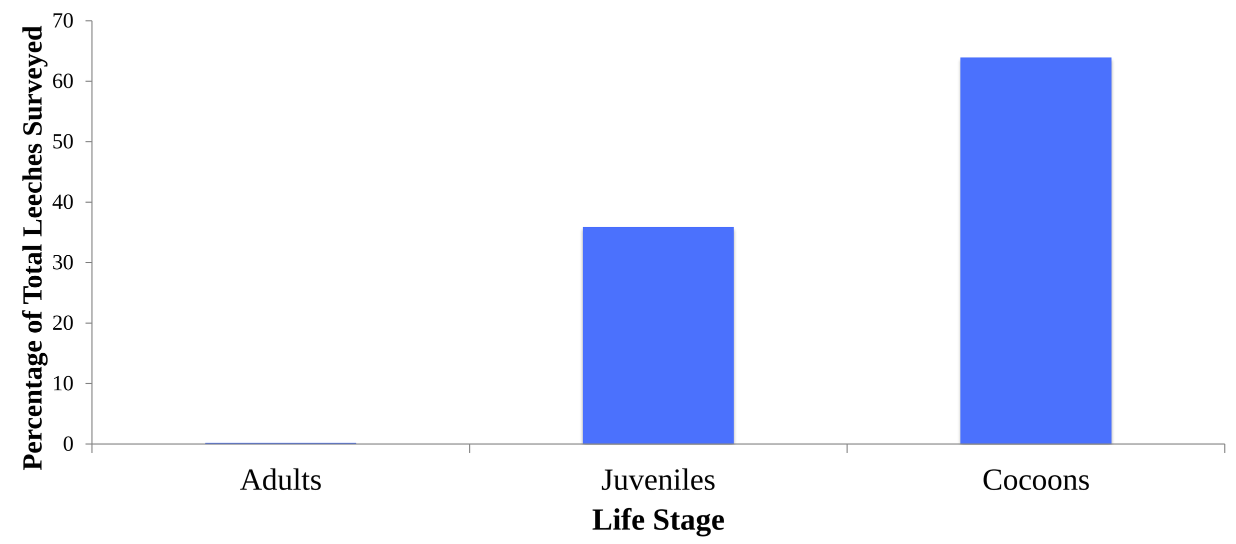

Fig. 2 The distribution of *M. montezuma* cocoons, juveniles, and adults during the months of quadrat sampling.

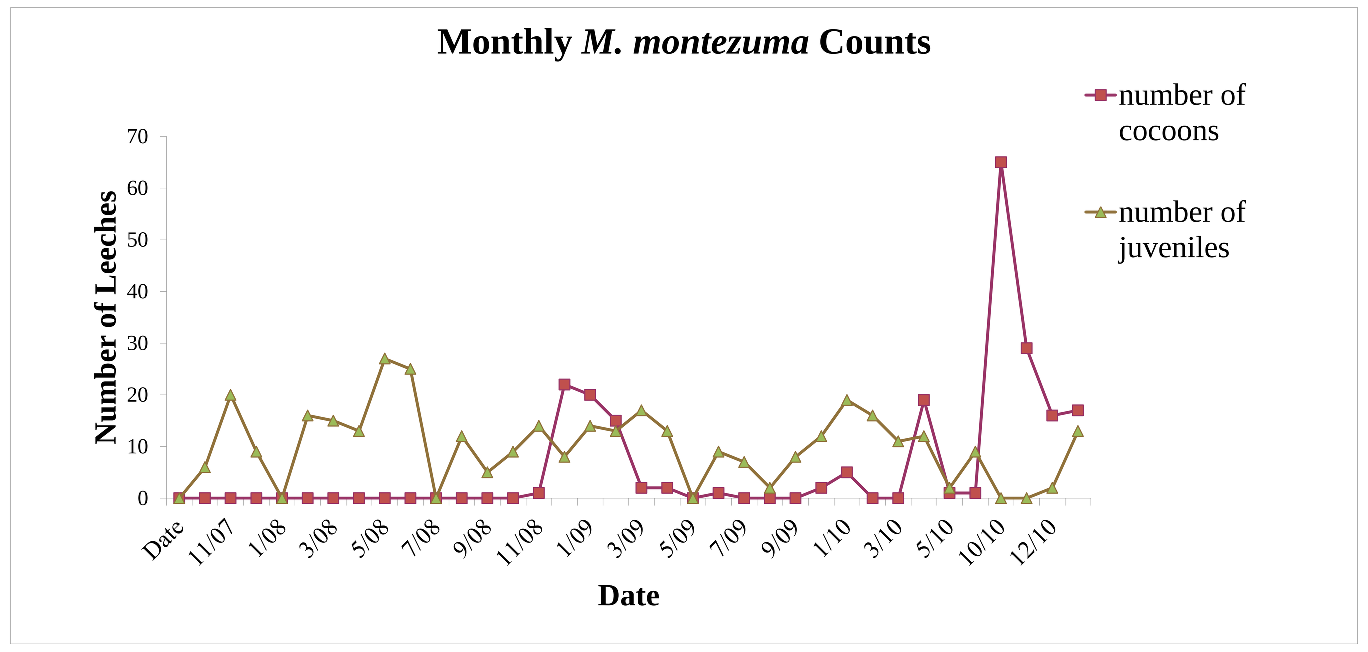

Fig. 3 Monthly counts of *Motobdella montezuma* cocoons and juveniles present in the swallet of the Montezuma Well.

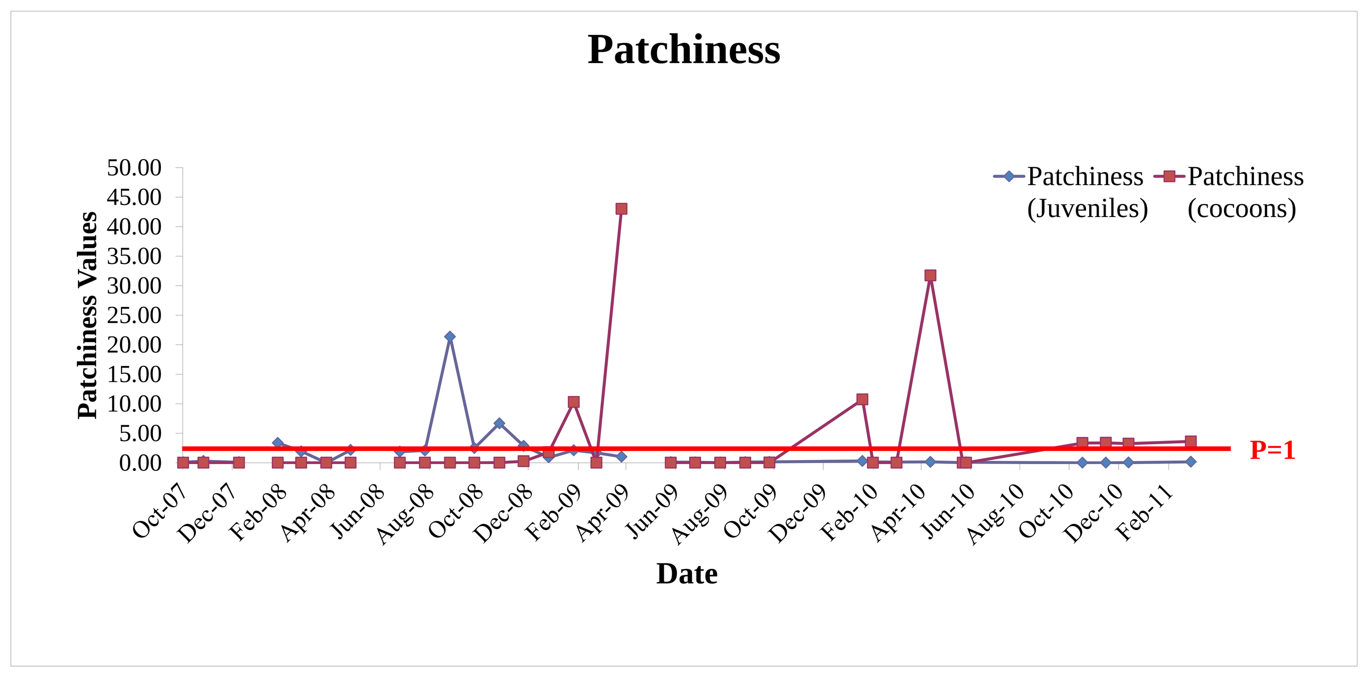

Fig. 4 Patchiness graph, showing spatial crowding of cocoons and juveniles where (p) is greater than one. Any value above the red line indicates clustering.

Image 4a (left), **4b** (right). Expanded view of Montezuma Well, including the swallet where this study was focused.

Discussion

These results are significant, as they provide the first documentation of the early life history of *Motobdella montezuma*. Our data suggests that this species breeds and secretes their cocoons in the early months of fall, with the juveniles hatching throughout the entirety of the fall season. In addition, only one adult *M. montezuma* was documented throughout the entirety of our study. This supports our assumption that the swallet is being used as a breeding area for these leeches. The implication that the swallet is indeed a breeding area to this species is important not only for the management of the swallet but also for the continuation of future research on the early life history of *M. montezuma*.

There was not as much spatial crowding of the juveniles and cocoons as we expected, based on personal observation of patchiness, but this could be due to the solitary, predatory nature of this species. Our data shows that cocoons had more spatial crowding than juveniles, which suggests that it is more beneficial for the adults to secrete their cocoons onto hard substrates with other cocoons, possibly to increase the likelihood of hatching by selecting a substrate that has been consistently chosen by its conspecifics (the substrate is most likely a good resource for the developing leeches). For juveniles, the lack of spatial crowding suggests that not clumping is more beneficial for *M. montezuma* juveniles, possibly because they are less vulnerable to predation at this point in their development (they are generally larger than the other species of leech that inhabit the Well) and have access to more resources when clumped in fewer numbers. Further research could be conducted on the swallet during the months prior to and of the cocoon and juvenile abundance peak in order to observe the *M. montezuma* adults entering the swallet. This research could be significant because now that the idea of the swallet as a breeding area for this species is supported, more information could be gleaned from observing the adults during the breeding event, as well as the secretion of the cocoons.

Image 6. *Motobdella montezuma* juvenile on a rock in the swallet of the Well.

References

Beresic-Perrins, R.K., 2010. NAU Thesis.

Blinn D.W., Davies R.W., & Dehdashti B. 1987. Specialized pelagic feeding by *Erpobdella Montezuma* (Hirudineae) Holartic Ecol. 10: 235-240.

Blinn D.W. & Davies R.W. 1989. New distribution records for leeches (Hirudinoidea) in Arizona. SW Nat. 34: 431-432.

Cole G.A. & Barry W.T. 1973. Montezuma Well, Arizona, as a habitat. J. AZ Acad. Sci. 8: 7-13.

Compton-O'Brien A.M, Foust, R. D., Ketterer, M. E., & Blinn, D. W. (2003). Total arsenic in a fishless desert spring: Montezuma Well, Arizona. American Chemical Society , 835, 200-209.

Davies R.W., Blinn D.W., Dehdashti B, & Singhal RN 1988. The comparative ecology of three species of Erpobdellidae (Annelida = Hirudinoidea). Archiv. Hydrobiol., 111: 601-614.

Davies R.W., Singhal R.N., & Blinn D.W. 1985. *Erpobdella Montezuma,* a new species of freshwater leech from North America (Arizona: U.S.A). Can. J. Zool. 63: 965-969.

Foust RD, Mohapatra P., Compton-O'Brien A-M, & Reifel J 2004. Groundwater arsenic in the Verde Valley in central Arizona, USA. Appl. Geochem. 19: 251-255.

Lloyd M. 1967. Mean crowding. J. Anim. Ecol. 36:1030.

O'Brien C. & Blinn D.W. 1999. The endemic spring snail *Pyrgulopsis montezumensis* in a high CO_2 environment: Importance of extreme chemical habitats as refugia. Freshwater Biol. 42: 225-234.

O'Brien, C., Blinn, D. W., & van Riper III, C. (2002). Waterfowl, acanthocephalans, and amphipods at Montezuma Well, Arizona - what is the role of a parasite?. Southwest Biological Science Center.

Shuster S.M. & Wade M.J. 2003. Mating systems and strategies. Princeton University Press, New Jersey.

Wade M.J. 1995. The ecology of sexual selection: Mean crowding of females and resource-defence polygyny. Evol. Ecol. 9: 118-124.

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