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Mantle displays of freshwater mussels elicit attacks from fish

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SUMMARY

1. Gravid females of some North American freshwater mussel species (Bivalvia: Unionidae) display highly modified mantle margins and other reproductive structures which mimic small fish, terrestrial insects, or aquatic macro-invertebrates. We report the responses of fish to these lures, based on the results of laboratory encounters between the following pairs of displaying mussels and fishes: *Lampsilis cardium* and *Micropterus coosae*; *L. perovalis* and *M. coosae*; and *Villosa nebulosa* and *Percina nigrofasciata*. In all three encounters, the lures elicited attacks from fish.
2. Encounters between *Lampsilis* spp. and *M. coosae* resulted in gill infestations of the fish by larval mussels, which are obligate parasites on fish. An encounter between *V. nebulosa* and *P. nigrofasciata* did not result in infestation.
3. The use of these lures to attract fish may greatly increase the chances of parasite/host encounters and may also reduce the chances of infestation of unsuitable hosts.

Keywords: glochidia, host-fish, mantle displays, mussels, parasites, Unionidae

Introduction

Most species of freshwater mussels in the family Unionidae have a unique life history in which the larvae (glochidia) are obligate parasites on the gills or fins of fish. The host specificity of the glochidia varies among mussel species, ranging from generalists which can use a wide variety of fish in many families to specialists which can use only a few closely related fish species. Therefore, female mussels are faced with the problem of ensuring that their propagules come in contact not only with a fish, but with a suitable host species.

Although glochidia may encounter fish passively in stream drift (Dartnall & Walkey, 1979; Neves & Widlak, 1988), many mussel species show adaptations which probably increase the chances of glochidia contacting a suitable host fish. The most conspicuous and well known of these adaptations are the modified mantle flaps, mantle papillae and large glochidial packages (superconglutinates) which are displayed by gravid females in the genera *Lampsilis* and *Villosa* (Kraemer, 1970; Haag, Butler & Hartfield, 1995; Haag, Warren & Shillingsford, 1999). These structures mimic small fish or large invertebrates, the major food items of basses (*Micropterus* spp.) (Carlander, 1977), which are the primary hosts for many species of *Lampsilis* and *Villosa* (Haag *et al.* 1999). It has been hypothesized that these structures serve as lures to attract host fish to the vicinity of the gravid mussel, whereby the fish may be infected with glochidia (e.g. Kat, 1984).

Despite the resemblance of these structures to food items of host fish, there is disagreement over their function and surprisingly little direct evidence supports their potential role in attracting fish. There are no detailed reports of encounters between fish and mantle flaps or superconglutinates. Wilson & Clark (1912; in Coker *et al.*, 1921) described darters and minnows being attracted to the waving mantle flaps of *Lampsilis cardium* (Rafinesque), but these fish are not known hosts for this species. Others have suggested that mantle flaps serve to aerate the glochidia brooded in the gills (Ortmann, 1911; in

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Coker *et al.*, 1921) or to fan sperm into the female siphons (Utterback, 1931). Kraemer (1970) provided extensive morphological and behavioural observations on mantle flaps in *Lampsilis*, but concluded that the flaps probably did not serve to attract host fish and asserted that the primary function is to suspend the glochidia in the water column upon release from the female. Currently, the most widely accepted and perhaps most plausible explanation remains that of a lure for host fish. However, the response of fish to mantle flaps and superconglutinates remains largely unknown.

Many North American mussel species are critically endangered, and remnant populations often exhibit reduced or non-existent recruitment (Williams *et al.*, 1993). The causes for this decline in recruitment are not understood, but it has been suggested that their complex life history renders freshwater mussels particularly susceptible to habitat alteration and concomitant changes in associated fish communities (Williams *et al.*, 1993). Until strategies for infection of host fish are better understood, it is difficult to assess possible explanations for widespread mussel declines.

The infestation of glochidia upon a suitable host fish is one of the most critical transitions in mussel life history, and has important implications for mussel ecology and conservation. Advances in these areas have been hampered by a fundamental lack of knowledge about how mussels infect host fish with glochidia. The purpose of the present study was to report the first observations and detailed descriptions of physical encounters between fish and gravid mussels which actively display mantle flaps and superconglutinates. We discuss the significance of these observations in the context of mussel reproductive ecology.

Materials and methods

We report encounters for the following mussel/fish pairs: *L. cardium* and *Micropterus coosae* (Hubbs & Bailey); *L. perovalis* (Conrad) and *M. coosae*; and *Villosa nebulosa* (Conrad) and *Percina nigrofasciata* (Agassiz). These encounters included species representing three types of displays: mantle flaps, *L. cardium*; superconglutinates, *L. perovalis*; and mantle papillae, *V. nebulosa*. Combinations of fish and mussels were chosen to pair gravid females with known or suspected hosts based on available host information. At the time of the present study, the hosts for *L. cardium* and *L. perovalis* were known to be basses (*Micropterus* spp.) (Waller *et al.*, 1985; Haag & Warren, 1997, respectively). Hosts for *V. nebulosa* were not known and were surmised incorrectly to be darters (i.e. *Etheostoma* spp. and/or *Percina* spp.; later work showed hosts for this species to be basses as well) (Haag & Warren, 1997).

Gravid mussels were collected and brought into the laboratory during periods of display activity in the wild. *Lampsilis perovalis* and *V. nebulosa* were collected in March 1996 from Flannagin Creek, Lawrence County, AL, U.S.A. *Lampsilis cardium* was collected in August 1996 from the Little Tallahatchie River, Panola County, MS, U.S.A. Prior to use in encounters, mussels were maintained in aerated aquaria at 9 °C to prevent discharge of glochidia. Fish for encounters were collected by electriofishing. *Micropterus coosae* and *P. nigrofasciata* were collected from Hubbard Creek, Lawrence County, AL, U.S.A., and Clear Creek, Winston County, AL, U.S.A. Before being used in encounters, fish were maintained at 21 °C and acclimated to laboratory conditions until they fed readily on minnows and earthworms (*M. coosae*) or frozen bloodworms (*P. nigrofasciata*).

Encounters were staged at 21 °C in a 151.2-L aquarium which was separated into two compartments by a removable plastic divider. One compartment held gravel and sand substrate to a depth of about 12 cm, and the other compartment had a false black plexiglass bottom level with the top of the substrate. For each encounter, a single gravid female was placed in the substrate and allowed to assume a natural posture. For the encounter involving *Lampsilis perovalis*, a power head was installed in the aquarium to provide current necessary to fully extend the superconglutinate. When the female began a sustained display, the fish was placed in the opposite chamber and allowed to acclimatize for 30 min. Prior to encounters, each fish was deprived of food for 2 days, anaesthetized (tricaine methanesulphonate, MS-222) and inspected under a dissecting microscope for pre-existing glochidial infestations. None of the fish were infested with glochidia prior to the encounters. After 30 min, the partition was removed and the encounter was videotaped for about 1 h. Videotapes were later reviewed and significant features of the encounters were timed using a digital stopwatch. Prior to release, female mussels brood

glochidia in the gill water tubes. After each encounter, female mussels were examined to estimate the number of gill water tubes which were evacuated during the encounter by holding the valves slightly open and examining the gills. This method gave an estimate of the proportion of total fecundity which was released during a single encounter. Fish were anaesthetized and inspected for glochidial infestation after encounters with displaying mussels.

Results

Lampsilis cardium

The display of *L. cardium* consisted of a sustained, regular flapping of the mantle margins; the mean interval between flaps was 0.47 s (SE = 0.01 s, $n = 7$). The individual was oriented with about one-third of the shell above the substrate. The mantle flaps were pigmented with a distinctive eyespot and longitudinal stripe (Fig. 1), similar to previous descriptions of this species (Kraemer, 1970). Gravid gills were displayed conspicuously above the shell margin (Fig. 1).

About 40 min after the beginning of the encounter, a redeye bass, *M. coosae*, approached slowly to within about 10 cm of the displaying mussel and paused for 8 s (Fig. 2). The fish then made a short, aborted attack, during which no physical contact was made with the mussel. However, the substrate was disturbed and the mussel began to slowly retract the mantle margins and gravid gills. No release of glochidia was apparent during this encounter. After this, the fish paused for ≈ 1 s then vigorously attacked and made physical contact with the mussel (Fig. 2). It was not possible to ascertain whether the fish struck the mantle margins or the gravid gill. At the moment of contact, the mussel quickly withdrew the gravid gills and mantle margins into the shell. As the gills were being withdrawn, a large cloud composed of individual glochidia and small aggregations of glochidia was released (Fig. 2). No complete conglutinates were released. Glochidia were not released from the excurrent siphon (Fig. 2), but apparently from ruptures or other openings in the distal ends of the gills.

After contact, the fish abruptly turned and fled; this attack lasted about 0.25 s. For several minutes after the encounter, the fish displayed an irritated behaviour, flaring its gills and repeatedly opening and closing its mouth, shaking its head violently, and rubbing the side of its head on the bottom of the aquarium. During the encounter, the mussel released the contents of approximately four gill water tubes. Examination of the fish revealed an infestation of at least eight glochidia.

Lampsilis perovalis

The display of *L. perovalis* consisted of a pair of superconglutinates (glochidial packets, each containing the entire contents of a single gravid gill) tethered to the female by a mucous strand emanating from the excurrent siphon (see Haag *et al.*, 1995). Superconglutinates were pigmented with a distinctive eyespot and lateral bands, and showed darting motion in the current as observed in the wild (Haag *et al.*, 1995). Approximately 70 min after initiation of the encounter, a redeye bass made a feigned pass at one of the superconglutinates, then retreated to the bottom of the aquarium for 30 s. The fish then rapidly approached the same superconglutinate and sucked it into its mouth with a rapid expansion of the buccal cavity. Almost immediately, the fish ejected the superconglutinate from its mouth and retreated to the bottom of the aquarium. For several minutes after the encounter, the fish displayed an irritated behaviour similar to that described for the redeye bass/*L. cardium* encounter. The superconglutinate remained intact after the attack and showed no visible damage from the encounter. After the attack, the fish remained on the bottom of the aquarium and showed no interest in either superconglutinate lure. After the encounter, the fish was infested with at least four glochidia.

Villosa nebulosa

During encounters, *V. nebulosa* exhibited only a partial display of mantle margins. The modified mantle margins of this species consist of long, tentacle-like papillae. During full display, the animal is exposed completely above the substrate, gravid gills are visible above or at the shell margin, and papillae are extended fully and pulsated rhythmically (Haag & Warren, 1997). The individual used in this encounter was exposed fully above the substrate, but gravid gills were not visible, papillae were not extended fully, and papillae showed no motion other than that produced by water currents in the aquarium. About 35 min after beginning the encounter, a blackbanded darter, *P.*

Fig. 1 Gravid female *Lampsilis cardium* in the laboratory displaying mantle flaps. The cream-coloured structure with horizontal striations in the centre of the photograph is the gravid left gill and this is flanked by the mantle flaps. The area between each striation represents one gill water tube. The mottled tissue below and adjacent to the mantle flaps are the unmodified mantle margins. The conical structure at the top is the excurrent siphon. The incurrent siphon is immediately below the excurrent siphon, and immediately above the mantle flaps and gravid gill. The large dark structure on the left side of the photograph is the left valve. The individual was collected from the Little Tallahatchie River, Panola County, MS, U.S.A., in August 1996 (bar = 1 cm).

nigrofasciata, swam to within 3 cm of the mussel and paused on the bottom for 16 s. The fish then turned its head toward the mussel and made an attack on the exposed, retracted ends of the papillae. This caused the mussel to close slightly, but no glochidia appeared to have been released. About 12 min later, the mussel had reopened slightly, and the fish again swam to the vicinity of the mussel, paused on the bottom for 7 s, then made a violent attack on the papillae that dislodged the mussel from its position on the substrate and caused it to completely withdraw the papillae and to close the shell. We saw no glochidia released during this encounter. After the attack, the fish did not exhibit any irritated behaviours such as those described for redeye bass encounters with *Lampsilis* spp. After the encounter, the mussel had no empty water tubes in the gravid portion of the gill, suggesting that few or no glochidia had been released, and the fish was not infected with glochidia.

Discussion

These observations confirm that modified mantle margins and superconglutinates of *Lampsilis* and *Villosa* act as lures which elicit attacks from fish.

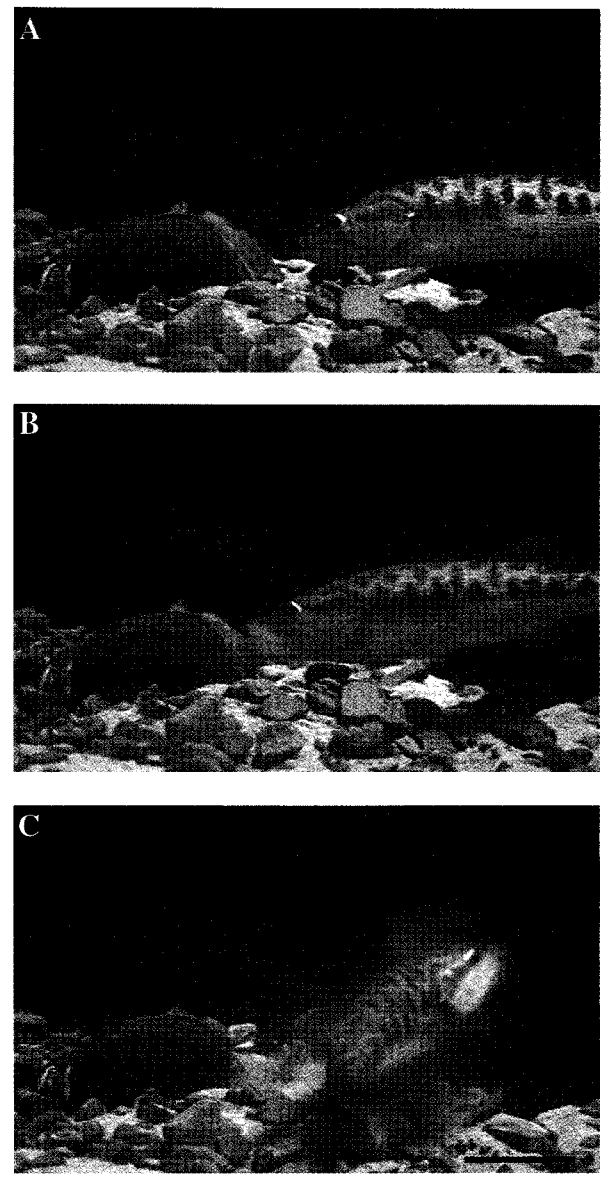

Fig. 2 Sequence of video stills from an encounter in the laboratory between a gravid female *Lampsilis cardium* (left) and a redeye bass (right). The left mantle flap is visible on the right margin of the shell with the gravid left gill immediately above. The conical excurrent siphon is visible at the top of the shell. In the bottom frame, the mantle flaps have been withdrawn after the attack, but the gravid gill is still visible. The large white mass immediately to the right of the gill is an aggregation of glochidia, and the white clouds are composed of free glochidia and sand suspended during the attack (bar = 4.5 cm).

During encounters with suitable host fish, these attacks resulted in the attachment of glochidia onto the gills of the fish. We hypothesize that these highly specialized structures function to increase the likelihood of glochidia encountering a fish. Other hypothesized functions are dubious and lack supporting evidence. The confirmation of these structures as lures raises questions regarding modes of host infection for mussels.

Little is known of the timing and duration of glochidial release or the number of fish which may be infected by a single gravid female. Our laboratory observations suggest that gravid females do not release the entire glochidial contents of the gills in any one encounter with a fish. In *Lampsilis*, about sixty water tubes (thirty in each gill) serve as glochidial marsupia (Oesch, 1984; W. R. Haag & M. L. Warren, unpublished data). When attacked by a redeye bass, *L. cardium* released the contents of only four water tubes, representing about 8% of the year's reproductive effort. In the wild, gravid females are often found with some water tubes full and others empty (personal observation). Similarly, the superconglutinates of *L. perovalis* remained intact after being attacked by a fish, and in the wild, superconglutinates are often found tattered, disassociated from the female, and snagged on stream debris where these continue to display fish-like motion (Haag *et al.*, 1995). Furthermore, in the laboratory encounters in the present study and according to data from wild populations (Neves & Widlak, 1988), numbers of glochidia infesting individual host fish are low. These data suggest that female mussels use a glochidial release strategy which allows multiple encounters with potential host fish. Potential advantages to such a strategy include increased opportunities for glochidia to encounter a suitable host as well as the distribution of glochidia among many individual hosts.

Strategies of host attraction and glochidial release in freshwater mussels may have evolved to exploit predator–prey relationships and feeding guilds of host fish (Haag & Warren, 1997), and may preferentially attract suitable host fish species. The wide variety of other apparent host-infestation strategies observed in other mussel genera also may serve to increase chances that glochidia will infest a suitable host. Few, if any species likely rely on chance alone to infect hosts. Confirmation of modified mantle flaps and superconglutinates as lures for host fish establishes that modes of host attraction and glochidial attachment in freshwater mussels are among the most important and elegant stages in the life history of these animals.

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References

Carlander K.D. (1977) *Handbook of Freshwater Fishery Biology*, Vol. S. Iowa State University Press, Ames, IA.

Coker R.E., Shira A.F., Clark H.W. & Howard A.D. (1921) Natural history and propagation of fresh-water mussels. *Bulletin of the U.S. Bureau of Fisheries*, **37**, 76–181.

Dartnall H.J.G. & Walkey M. (1979) Distribution of glochidia of the swan mussel, *Anodonta cygnea* (Mollusca) on the three-spined stickleback *Gasterosteus aculeatus* (Pisces). *Journal of Zoology, London*, **189**, 31–37.

Haag W.R., Butler R.S. & Hartfield P.D. (1995) An extraordinary reproductive strategy in freshwater bivalves: prey mimicry to facilitate larval dispersal. *Freshwater Biology*, **34**, 471–476.

Haag W.R. & Warren M.L., Jr (1997) Host fishes and reproductive biology of 6 freshwater mussel species from the Mobile Basin, USA. *Journal of the North American Benthological Society*, **16**, 576–585.

Haag W.R., Warren M.L., Jr & Shillingsford M. (1999) Host fishes and host-attracting behavior of *Lampsilis altilis* and *Villosa vibex* (Bivalvia: Unionidae). *American Midland Naturalist*, **141**, 149–157.

Kat P. (1984) Parasitism and the Unionacea (Bivalvia). *Biological Review*, **59**, 189–207.

Kraemer L.R. (1970) The mantle flap in three species of *Lampsilis* (Pelecypoda: Unionidae). *Malacologia*, **10**, 225–282.

Neves R.J. & Widlak J.C. (1988) Occurrence of glochidia in stream drift and on fishes of the upper North Fork Holston River, Virginia. *American Midland Naturalist*, **119**, 111–120.

Oesch R.D. (1984) *Missouri Naiades*. Missouri Department of Conservation, Jefferson City, MO.

Ortmann A.E. (1911) A monograph of the Najades of Pennsylvania. *Memoirs of the Carnegie Museum*, **4**, 279–347.

Utterback W.I. (1931) Sex behavior among naiades. *Proceedings of the West Virginia Academy of Science*, **5**, 43–45.

Waller D.L., Holland L.E., Mitchell L.G. & Kammer T.W. (1985) Artificial infestation of largemouth bass and walleye with glochidia of *Lampsilis ventricosa* (Pelecypoda: Unionidae). *Freshwater Invertebrate Biology*, **4**, 152–153.

Williams J.D., Warren M.L., Jr, Cummings K.S., Harris J.L. & Neves R.J. (1993). Conservation and status of freshwater mussels of the United States and Canada. *Fisheries*, **18**, 6–22.

Wilson C.B. & Clark, H.M. (1912) *The mussel Fauna of the Kankakee Basin, U.S.* Bureau of Fisheries Document 758. U.S. Bureau of Fisheries, Washington, DC.

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