

Life history and propagation of the endangered dromedary pearlymussel (*Dromus dromas*) (Bivalvia:Unionidae)

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Abstract. The reproduction, demography, and propagation of the endangered dromedary pearlymussel (*Dromus dromas*) (Lea, 1834) were studied in the Clinch and Powell rivers, Tennessee. Viable populations of the dromedary pearlymussel now occur only in the Clinch and Powell rivers; the species has been extirpated from the remaining portions of its range in the Cumberland and Tennessee river drainages. Females are long-term winter brooders, and they are gravid from October to June. Glochidia are contained in conglutinates that are red to white and resemble freshwater leeches or flatworms. Conglutinates are 20 to 40 mm long and are released through the excurrent aperture. Estimates of fecundity based on 7 gravid females collected from the Clinch River were 55,110 to 253,050 glochidia/mussel. The ages of 66 valves of *D. dromas* were determined by thin-sectioning and ranged from 3 to 25 y. Annual growth averaged 5 mm/y until age 10 and decreased to ~1.2 mm/y thereafter. Nineteen fish species were tested for suitability as hosts for glochidia. Ten were confirmed as hosts through induced infestations of glochidia: black sculpin (*Cottus baileyi*), greenside darter (*Etheostoma bleennioides*), fantail darter (*Etheostoma flabellare*), snubnose darter (*Etheostoma simoteronum*), tangerine darter (*Percina aurantiaca*), blotchside logperch (*Percina burtoni*), logperch (*Percina caprodes*), channel darter (*Percina copelandi*), gilt darter (*Percina evides*), and Roanoke darter (*Percina roanoka*). Juveniles produced from these hosts were cultured in dishes held in nonrecirculating aquaculture systems containing fine sediment (<105 µm) and were fed the green alga *Nannochloropsis oculata* every 2 d. Survival of 2810 newly metamorphosed juveniles was 836 (29.7%) after 1 to 2 wk.

Key words: dromedary pearlymussel, *Dromus dromas*, life history, fish hosts, reproduction, propagation.

The dromedary pearlymussel (*Dromus dromas*) (Lea, 1834) inhabits shoals of coarse gravel and sand in medium to large rivers of the Cumberland Plateau and Southern Appalachian Mountains (Ortmann 1918). Historically, this lampsiline species was widespread and abun-

dant throughout the Cumberland and Tennessee river drainages (Parmalee and Bogan 1998). It once occurred in the Tennessee River system from its major headwater tributary streams (e.g., Powell, Clinch, Holston, and French Broad rivers) downstream to Mussel Shoals in northern Alabama (Ortmann 1918, 1925, USFWS 1983), and in the Cumberland River system from its major headwater tributary streams (e.g., Big South Fork Cumberland River) downstream to below Clarksville, Tennessee (Wilson and Clark

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1914). *Dromus dromas* was one of the most abundant species in aboriginal shell middens along the Tennessee and Cumberland rivers (Morrison 1942, Parmalee et al. 1980, 1982).

Dromus dromas was listed as endangered by the US Fish and Wildlife Service in 1976 (Terwilliger 1991) because of severe declines in populations during the 20th century. Dams, channel dredging, sand and gravel mining, coal mining, and sewage wastes have caused drastic declines in populations of *D. dromas* throughout its range. Presently, populations in the main Tennessee and Cumberland rivers are gone or not reproducing. A few old specimens were taken in the Tennessee River in Chickamauga Reservoir between 1978 and 1983 (Ahlstedt and McDonough 1995–1996). Likewise, a few old specimens were seen in the Cumberland River near Rome Landing, Tennessee, between 1976 and 1994 (Parmalee et al. 1980, USFWS 1983, Ahlstedt 1995–1996, Hubbs 1995). However, reproducing populations of *D. dromas* occur only in the upper Clinch and Powell rivers in Tennessee and Virginia above Norris Reservoir (USFWS 1983, Terwilliger 1991, Ahlstedt and Tuberville 1997). Currently, only ~158 river km in the Clinch River and ~79 river km in the Powell River, <10% of the historic range of *D. dromas*, contain reproducing populations (USFWS 1983; R. Biggins, US Fish and Wildlife Service, unpublished data). Populations are disjunct, and range reductions may still be occurring even within the Clinch and Powell rivers (Ahlstedt and Jenkinson 1987, Ahlstedt 1991, Ahlstedt and Tuberville 1997).

Dromus dromas was reported as a long-term brooder by Ortmann (1912); however, knowledge of most other life-history aspects is lacking. The purposes of our study were to provide needed biological data specified in the federal recovery plan for this species (USFWS 1983), and to gather information on current distribution, life history, and propagation to apply to its restoration and recovery.

Methods

Fecundity from conglutinates

The fecundity of 7 female *D. dromas* collected from the Clinch River was estimated by flushing conglutinates from each female with a hypodermic needle filled with water. Glochidia were collected from 4 to 6 conglutinates/mussel. Glo-

chidia were teased from each conglutinate using the tip of a hypodermic needle and counted to determine the number of glochidia/conglutinate. This technique damaged some glochidia but released 1000s unharmed. Fifty to 100 glochidia usually could not be removed from a conglutinate, so these glochidia were counted or estimated visually, and the number was added to the total for each conglutinate examined. The mean number of glochidia/conglutinate was multiplied by the total number of conglutinates/female mussel to estimate fecundity.

Age and growth

Live *D. dromas*, shells from freshly dead individuals, and relic shells were collected from various Clinch River km (CRKM) locations between Horton Ford (CRKM 321) and Swan Island (CRKM 277), Hancock County, Tennessee. Shells of various lengths were collected to best represent the size-class structure of the population in the river. The lengths of 81 live *D. dromas* were measured between 1998 and 2001. Nongravid individuals were returned immediately to sites of collection, but gravid individuals were kept for subsequent extraction of glochidia.

Relic and freshly dead shells were aged if the periostracum and external growth rings were intact. Thin sections of 66 shells were prepared following procedures described by Clark (1980) and Neves and Moyer (1988), using a Buehler Isomet low-speed saw unit with a diamond-impregnated blade (Buehler, Evanston, Illinois). Shells were cut from the center of the umbo to the ventral margin. Cut valves were glued (2-Ton Clear Epoxy, Illinois Tool Works, Devcon, Massachusetts) to petrographic microslides (27 × 46 mm), vacuum-sealed into a petrographic chuck, attached to the cutting arm of the saw, and sectioned at a thickness of 280 μm (Neves and Moyer 1988). Thin sections of shells were examined under 4 × magnification. Internal growth lines were considered true annuli if they were continuous from the umbo region to the outer surface of the shell. It was assumed, based on the work of Neves and Moyer (1988) in the rivers of southwest Virginia, that one annulus was formed each year. Lengths for 1- and 2-year old individuals were obtained by back-calculating length-at-age based on internal annuli of 5 older shells (Bruenderman and Neves 1993) be-

cause shells <3 y old were difficult to collect along the river.

Collection of gravid female mussels

Gravid females of *D. dromas* were obtained by snorkeling. Individuals were examined for gravidity by opening the valves slowly by hand to look for conglomerates. Female mussels typically were collected in autumn and winter when water levels in the Clinch River were low. The mussels were held in flow-through raceways containing pea-sized gravel at Buller Fish Hatchery, near Marion, Virginia. Water from the South Fork Holston River was pumped into the raceways to provide suitable holding conditions in the late winter and early spring. Female mussels were moved into temperature-controlled, water-recirculating systems at the Freshwater Mollusk Conservation Center at Virginia Polytechnic Institute and State University once water temperatures in the raceways reached 15 to 18°C in the spring. Glochidia also were removed from gravid females in the late autumn and early winter for the purpose of conducting fish-host trials. Water temperatures were set initially at ~5 to 12°C to mimic temperatures in the South Fork Holston River and then gradually increased to 21 to 23°C over 1 to 2 d. Females were held at 21 to 23°C for 1 to 2 wk to allow the brooding glochidia of female mussels to finish maturation in the laboratory. Females began releasing conglomerates after ~1 wk. Maturity of the glochidia was tested by exposing them to a dilute salt solution (Zale and Neves 1982). Glochidia were deemed mature when >20 to 30% exhibited rapid, sometimes multiple, snapping responses when exposed to salt. The desired number of conglomerates (30–50) for propagation of juvenile mussels was flushed from the gills of the female once the glochidia were conditioned (i.e., mature).

Fish hosts

Nineteen species of fish were tested as potential hosts for glochidia. Most of the fish were collected from the upper North Fork Holston River, ~1 km above Saltville, Virginia. Banded sculpins (*Cottus carolinae*) were collected from the Middle Fork Holston River at Atkins, Smyth County, Virginia, and mottled sculpins (*Cottus bairdi*) were collected from Sinking Creek at

Newport, Giles County, Virginia. Gilt darters (*Percina evides*) were collected from the Powell River, Lee County, Virginia, at a site (State Route 833 bridge) where *D. dromas* was rare. All other fish collection sites had few mussels and no *D. dromas*. Fish were collected using low voltage (200 V) electroshocking in late October while in good physiological condition to increase survival of captive *P. evides* held in the recirculating tank systems (see below). Common and scientific names follow Robins et al. (1991) for fishes and Turgeon et al. (1998) for mussels.

Methods for infesting fish with mussel glochidia generally followed those of Zale and Neves (1982). A plastic container 29 × 19 × 12 cm deep was used to hold fish during infestation. Fifteen to 25 fish of various species were held simultaneously in ~0.5 L of water, and glochidia from 30 to 50 conglomerates were added to the container. The water in the container was agitated by using 2 air stones, 1 on each side of the container, for 1 h. After infestation, fish were separated by species and placed in 38-L aquaria without substrate.

Percina evides required more space and water flow to survive, and this species was held in a 1136-L, water-recirculating tank system. This tank system consisted of four 208-L aquaria, 2 filled with 8 to 10 cm of pea-sized gravel and 2 with no substrate. Infested *P. evides* were held in the aquaria with gravel until glochidial metamorphosis (~3 wk at 20°C), and the fish were transferred to aquaria without substrate to facilitate collection of juveniles when juveniles were ready to excyst.

Fish were held at low densities (1–8/tank in 38-L aquaria and 30–60/tank in 208-L aquaria), depending on the species, sizes, and numbers of fish. The bottoms of the aquaria were siphoned every 2 to 3 d until juvenile mussels were first collected and every 1 to 2 d thereafter. About 10 to 15 L of conditioned water were added to the 38-L aquaria after siphoning to maintain water levels. Juveniles were counted and placed in a culture dish with sediment and algae for rearing. Collection of mobile juveniles confirmed that the glochidia were capable of transforming on the fish species and that the fish species was a potential host of *D. dromas*.

Culture of juveniles

Some juvenile mussels were cultured in small (6 × 6 × 5 cm) Tupperware® containers using



FIG. 1. Conglutinates of the dromedary pearlymussel (*Dromus dromas*). Pink and white conglutinates (~2.5 cm long) contain mature glochidia along the outer margins.

a nonrecirculating culture system (Jones and Neves 2002). A total of 2049 juvenile mussels were placed in 14 containers (mean = 146 juveniles/container; range = 37–257). The containers were filled with 50 mL of conditioned water (a 1:1 mix of dechlorinated tap water and well water) and 50 mL of conditioned water containing the green alga *Nannochloropsis oculata* (1×10^5 to 1×10^6 cells/mL). Hardness ranged from 250 to 350 mg/L CaCO_3 . In addition, ~0.5 mL of fine sediment (particle size $<105 \mu\text{m}$) was pipetted into each dish. Sediment and sand were autoclaved to kill predators such as flatworms and dipteran larvae prior to placement in culture dishes. The water, algae, and sediment were exchanged every 2 d, and water temperatures were maintained at ambient levels (20–21°C). Juvenile mussels were counted and measured to determine survival and growth at 2 wk.

Data analysis

All analyses were conducted using SAS (version 8.2, SAS Institute, Cary, North Carolina). A von Bertalanffy growth equation was computed from length-at-age data and was fit by nonlinear

procedures to derive the parameters of the growth equation.

Results

Fecundity and release of conglutinates

Glochidia of *D. dromas* were contained in red, pink, or white conglutinates shaped like leeches (Fig. 1). The yolk of the embryos was usually red at the beginning of the brooding season (autumn), giving the conglutinates their color. Some females contained white conglutinates in the autumn and spring, but this color was less common. The red color faded and conglutinates typically became pink as embryos matured into glochidia during the spring. This color change could be seen at the margin of a conglutinate where glochidia were mature and transparent, making this section opaque (Fig. 1).

Females were gravid from October through May (Table 1) and contained 33 to 151 conglutinates/female (Table 2). Conglutinates were contained only in the water-tubes of the outer gills, and conglutinates typically were observed in all of the water-tubes from the anterior to the posterior portion of the gill. In the autumn, some females were only partially gravid, i.e.,

TABLE 1. Reproductive condition of *Dromus dromas* collected from the Clinch River between 1998 and 2002.

| | October– November | December– January | February– March | April–May | June–July | August– September |
|----------------|----------------------|----------------------|--------------------|-----------|-----------|----------------------|
| No. examined | 80 | 37 | 60 | 150 | 203 | 211 |
| No. gravid (%) | 28 (35) | 15 (41) | 17 (28) | 41 (27) | 2 (1) | 1 (0.5) |

only a portion (typically the anterior ½) of their gills were filled with conglomerates. Conglomerates were 20 to 50 mm long and 4 to 7 mm wide. An estimated 50 to 70% of conglomerate contents were eggs or embryos in most of the females examined. These eggs or embryos may serve a role in keeping the conglomerate together and making it alluring to fish hosts. Fecundity ranged from 55,110 to 253,050 glochidia/mussel (Table 2).

Between 1998 and 2002, >100 gravid *D. dromas* were observed in the field and laboratory, and >50% more embryos developed into glochidia in female mussels collected late in the brooding season than in female mussels collected early in the brooding season, suggesting that high water temperatures facilitated the maturation process. Mature glochidia were observed only in conglomerates extracted from female mussels collected in April and May.

Females in captivity released conglomerates from the excurrent aperture. Conglomerates were released first into the suprabranchial cavity before exiting through the excurrent aperture. Conglomerates were released 1 at a time at a rate of 3 to 4/d; the basal end exited first. The earliest date of expulsion of conglomerates from the marsupium of females in the river was late Feb-

ruary and early March. However, many females collected in late April and May had few conglomerates remaining, suggesting that most females released their conglomerates from late March to late April, when water temperatures in the Clinch River were 15 to 20°C. Females held at Buller Fish Hatchery released conglomerates at similar water temperatures. Some of the mussels held at the hatchery released their entire glochidial brood (typically ~150 conglomerates) over a period of 2 wk.

In November 2002, a gravid female mussel was moved from Buller Hatchery to the Freshwater Mollusk Conservation Center, where it was held in a temperature-controlled (21°C) water recirculating system. This female was held in the system for 2 wk to allow the glochidia to finish maturation. The female began releasing conglomerates in ~4 to 5 d. The initial conglomerates contained immature glochidia, whereas conglomerates released after 10 to 12 d appeared mature. Glochidia from these later conglomerates were used for host fish trials. The juveniles produced from these trials were actively pedaling and appeared robust. Previous host fish trials using glochidia removed from females in autumn and winter of 1998/1999 produced juveniles that did not survive. The glochidia were removed from these female mussels within days after collection from the river; therefore, it is unlikely maturation was complete.

TABLE 2. Shell length of gravid female mussels and fecundity estimates for *Dromus dromas* collected from the Clinch River ($n = 7$). See text for details.

| Date examined | Shell length (mm) | Con- glu- tina- tes | Glochi- dia/ conglu- tinate | Glochidia/ mussel |
|----------------|-------------------|------------------------------|--------------------------------------|----------------------|
| 5 January 1999 | 62.0 | 151 | 1687 | 253,050 |
| 3 May 1999 | 53.2 | 123 | 980 | 120,540 |
| 4 April 2000 | 64.1 | 52 ^a | 1892 | 98,384 |
| 4 April 2000 | 54.5 | 33 ^a | 1670 | 55,110 |
| 19 April 2001 | 51.0 | 93 | 519 | 48,267 |
| 26 April 2001 | 61.0 | 128 | 929 | 118,912 |
| 27 April 2001 | 56.0 | 97 | 644 | 64,408 |

^a Only ~50% of water tubes contained conglomerates

Age and growth

Relic shells and shells from recently dead individuals ranged in age from 3 to 25 y. Mean lengths (L) for each age class (t) were fitted to the von Bertalanffy growth equation, $L_t = 70.1 \text{ mm} \times (1 - e^{-0.123(t-0.111)})$, and used to predict values for length ($R^2 = 0.97$). Growth averaged 5.0 mm/y through age 10 y and decreased to ~1.8 mm/y thereafter (Table 3).

Lengths of shells from dead and live individuals were normally distributed. The mean length of shells from dead individuals was 51.3

TABLE 3. Observed and predicted shell lengths at internal annuli of *Dromus dromas* from the Clinch River, 1999 to 2000. – = no data.

| Internal annulus | No. of individuals | Observed length (mm) | | Predicted length (mm) | Growth increment (mm) |
|------------------|--------------------|----------------------|-----------|-----------------------|-----------------------|
| | | Mean | Range | | |
| 0 | 1 | 0.26 | 0.26 | -0.96 | 0 |
| 1 | 5 | 7.5 | 4.6–9.6 | 9.0 | 9.0 |
| 2 | 5 | 13.0 | 8.3–16.1 | 16.1 | 7.1 |
| 3 | 5 | 28.1 | 20.0–34.5 | 22.3 | 6.2 |
| 4 | 1 | 29.6 | 29.6 | 27.9 | 5.6 |
| 5 | 5 | 33.62 | 26.8–39.0 | 32.8 | 4.9 |
| 6 | 6 | 34.2 | 30.1–37.9 | 37.1 | 4.3 |
| 7 | 4 | 41.5 | 36.9–49.3 | 40.9 | 3.8 |
| 8 | 6 | 44.5 | 44.4–49.3 | 44.3 | 3.4 |
| 9 | 5 | 45.9 | 42.5–49.4 | 47.3 | 3.0 |
| 10 | 3 | 49.3 | 46.3–54.8 | 50.0 | 2.7 |
| 11 | 6 | 56.3 | 48.2–67.2 | 52.3 | 2.3 |
| 12 | 1 | 46.9 | 46.9 | 54.4 | 2.1 |
| 14 | 3 | 59.3 | 57.6–62.3 | 57.8 | 1.6 |
| 15 | 3 | 59.5 | 55.1–65.3 | 59.2 | 1.4 |
| 16 | 4 | 60.9 | 58.8–64.3 | 60.5 | 1.3 |
| 17 | 1 | 60.0 | 60.0 | 61.6 | 1.1 |
| 18 | 3 | 61.2 | 58.6–64.0 | 62.6 | 1.0 |
| 19 | 4 | 63.4 | 61.0–66.5 | 63.5 | 0.9 |
| 20 | 5 | 67.2 | 64.5–75.6 | 64.3 | 0.8 |
| 21 | 0 | – | – | 64.7 | 0.4 |
| 22 | 0 | – | – | 65.3 | 0.6 |
| 23 | 0 | – | – | 65.9 | 0.6 |
| 24 | 0 | – | – | 66.4 | 0.5 |
| 25 | 2 | 75.0 | 71.5–78.4 | 66.8 | 0.4 |

mm (SE = 1.6, median = 53.0 mm, range = 26.8–78.4 mm), whereas the mean length of shells of live individuals was 51.1 mm (SE = 1.4, median = 51.0 mm, range = 28.9–83.0 mm). Lengths of shells from dead individuals ($n = 66$) and live individuals ($n = 81$) from the Clinch River were not significantly different ($p > 0.05$). The mean age of dead shells was nearly 12 y (based on data presented in Table 3). The smallest gravid individuals ranged from 38 to 42 mm, indicating that most individuals were mature by 6 to 9 y of age.

Fish hosts

Nine species of darter (Percidae) and 1 species of sculpin (Cottidae) were identified as hosts from induced infestations of glochidia (Table 4). Nine of the species were native to the Tennessee River system (Jenkins and Burkhead 1993) and

sympatric with *D. dromas* in all or part of its range. The Roanoke darter (*Percina roanoka*) was native to the Roanoke River system (Jenkins and Burkhead 1993) and was the only suitable fish host identified from another drainage.

Culture of juveniles

In 1999, 2049 viable juveniles were collected from fish-host experiments and cultured in non-recirculating containers. The average 1-d-old juvenile was 256 μm long, 128 μm high, and 102 μm wide ($n = 70$). The excystment of juvenile mussels varied in timing and duration (17–70 d) among fish host species identified in our study (Table 4). Survival of these juvenile mussels at 2 wk was 29.7%, and mean length was 308 μm . Juveniles were released into the Clinch River in spring 1999.

Discussion

Taxonomic status and release of conglutinates

Simpson (1896, 1900) and Ortmann (1910a, 1912) reported that conglutinates of *D. dromas* were contained only in the lower posterior portion of the gill, a character Simpson termed "eschatigenae". This characteristic was used later by Heard and Guckert (1971) to indicate the unique systematic position of the species within the Unionidae. These early observations probably were based on gravid female mussels that were collected early in the brooding season when the water-tubes of the outer gills were only partially filled with conglutinates. These observations probably were an artifact of the timing of collections of mussels by early 19th and 20th century naturalists, who typically collected in late summer and early autumn when water levels were low and the river shoals were accessible. Conglutinates are contained in all of the water tubes of the outer gill when females are completely gravid, and individuals that are partially gravid in the early autumn can give the impression of having the eschatigenae condition.

Dromus dromas is one of the few mussel species in the subfamily Lampsilinae that produces modified conglutinates that are released through the suprabranchial cavity. Ortmann (1909, 1910b, 1911) considered the release of glochidia through the suprabranchial cavity and

TABLE 4. Results of induced infestations on possible fish hosts for the glochidia of *Dromus dromas*. Days to transformation was the first day and last day when juveniles excysted from a fish host. The peak (in parentheses) was when most juveniles excysted. Temperature is the mean water temperature in the aquaria where fish were held. * = successful host species. – = no data.

| Fish species | No. fish tested | No. fish alive | Time (d) | Juveniles recovered | Days to transformation | Temperature (°C) |
|--|-----------------|----------------|----------|---------------------|------------------------|------------------|
| Centrarchidae | | | | | | |
| <i>Micropterus dolomieu</i> ^b | 2 | 2 | 65 | 0 | – | 19.0 |
| Cottidae | | | | | | |
| <i>Cottus carolinae</i> ^a | 3 | 3 | 48 | 0 | – | 19.0 |
| * <i>Cottus baileyi</i> ^a | 6 | 5 | 48 | 7 | 31–48 | 20.5 |
| Cyprinidae | | | | | | |
| <i>Rhynchithys atratulus</i> ^b | 3 | 3 | 65 | 0 | – | 19.0 |
| Ictaluridae | | | | | | |
| <i>Amieurus nebulosa</i> ^b | 2 | 2 | 65 | 0 | – | 19.0 |
| <i>Noturus insignis</i> ^c | 2 | 2 | 65 | 0 | – | 19.0 |
| Percidae | | | | | | |
| * <i>Etheostoma blennioides</i> ^a | 5 | 3 | 53 | 3 | 43–48 | 19.0 |
| * <i>E. blennioides</i> ^d | 1 | 1 | 31 | 77 | 17, (18–22), 31 | 23.0 |
| * <i>Etheostoma flabellare</i> ^a | 4 | 3 | 53 | 8 | 21, (28), 48 | 19.0 |
| * <i>E. flabellare</i> ^c | 50 | 40 | 52 | 259 | 33, (40–46), 48 | 19.0 |
| * <i>E. flabellare</i> ^d | 20 | 7 | 32 | 945 | 17, (18–22), 31 | 23.0 |
| * <i>E. flabellare</i> ^g | 300 | 20 | 35 | 15 | 17, (22–25), 37 | 21.0 |
| <i>Etheostoma rufilineatum</i> ^a | 5 | 4 | 53 | 0 | – | 19.0 |
| * <i>Etheostoma simoterum</i> ^a | 6 | 4 | 53 | 12 | 21, (46), 48 | 19.0 |
| * <i>E. simoterum</i> ^c | 20 | 17 | 50 | 70 | 31, (40–45), 48 | 19.0 |
| <i>Etheostoma zonale</i> ^a | 3 | 3 | 53 | 0 | – | 19.0 |
| <i>E. zonale</i> ^c | 6 | 5 | 53 | 0 | – | 19.0 |
| <i>Perca flavescens</i> ^b | 4 | 4 | 65 | 0 | – | 19.0 |
| * <i>Percina aurantiaca</i> ^a | 2 | 1 | 48 | 68 | 21, (31), 43 | 20.5 |
| * <i>Percina burtoni</i> ^a | 2 | 2 | 48 | 121 | 21, (30–32), 34 | 20.5 |
| * <i>Percina caprodes</i> ^a | 8 | 3 | 48 | 38 | 21, (34–39), 48 | 20.5 |
| * <i>P. caprodes</i> ^d | 3 | 2 | 49 | 108 | 21, (37–39), 47 | 21.0 |
| * <i>P. caprodes</i> ^e | 5 | 4 | 47 | 143 | 19, (32–35), 45 | 21.0 |
| * <i>Percina copelandi</i> ^c | 2 | 2 | 47 | 5 | 28, (33), 45 | 21.0 |
| * <i>Percina evides</i> ^c | 20 | 1 | 58 | 49 | 48, (51–53), 55 | 18.0 |
| * <i>P. evides</i> ^h | 50 | 48 | 74 | 3489 | 37, (50–59), 69 | 21.0 |
| * <i>P. evides</i> ⁱ | 50 | 45 | 74 | 1655 | 30, (40–59), 70 | 21.0 |
| * <i>Percina roanoka</i> ^a | 3 | 3 | 46 | 4 | 46 | 19.0 |
| * <i>P. roanoka</i> ^c | 65 | 54 | 50 | 713 | 32, (38–42), 48 | 18.0 |
| * <i>P. roanoka</i> ^f | 53 | 47 | 44 | 245 | 16, (29–34), 42 | 21.0 |
| Salmonidae | | | | | | |
| <i>Oncorhynchus mykiss</i> ^b | 1 | 1 | 65 | 0 | – | 19.0 |

^a Fish were infested together for 1 h with glochidia from 123 conglutinates on 18 November 1998. Glochidia were mature enough to close their valves during a salt test. However, the closing response for many of these glochidia was slow, possibly indicating they were not very mature

^b Fish were infested together for 1 h with glochidia from 50 conglutinates on 29 January 1999

^c Fish were infested together for 1 h with glochidia from 35 conglutinates on 4 February 1999

^d Fish were infested together for 1 h with glochidia from 30 conglutinates on 29 May 1999

^e Fish were infested together for 1 h with glochidia from 43 conglutinates on 31 May 1999

^f Fish were infested together for 1 h with glochidia from 52 conglutinates on 2 June 1999

^g Fish were infested together for 1 h with glochidia from 50 conglutinates on 20 May 2001

^h Fish were infested together for 1 h with glochidia from 47 conglutinates on 12 November 2002

ⁱ Fish were infested together for 1 h with glochidia from 42 conglutinates on 5 May 2003

excurrent aperture primitive and more typical of amblemine and anodontine than lampsiline mussels. Female mussels of most lampsiline species release glochidia through pores along the ventral margin of the gill, have a modified mantle lure to attract fish hosts, and are bradytictic or long-term brooders. *Dromus dromas* is bradytictic, but females release conglutinates over a relatively short period of time once glochidia are mature, a characteristic similar to many tachytictic or short-term brooding species. *Dromus dromas* also lacks a mantle lure, and has a thicker shell than typical of the Lampsilinae. In addition, the shells of the dromedary are not typically sexually dimorphic; occasionally the shells of some females appear more inflated than those of males, but the difference is subtle. Lack of sexually dimorphic shells is a characteristic of most amblemine mussel species. Therefore, the dromedary has some traits characteristic of both amblemine and lampsiline species. Similar traits have been reported for other lampsilines, the fanshell *Cyprogenia stegaria* (Jones and Neves 2002) and *Ptychobranthus* spp. Thus, these genera, and others, may represent a distinct group within the subfamily Lampsilinae. To our knowledge, the shape of the glochidium is unique among the Unionidae, supporting the monotypic status of the genus. Our measurements of newly excysted juveniles agree with dimensions of glochidia reported by Hoggarth (1999).

Age and growth

The original listing of this species (USFWS 1983) as endangered was based, in part, on the apparent absence of recruitment for this species in the Clinch River. However, our study showed that the mean shell length of live *D. dromas* in the Clinch River was 51.1 mm, corresponding to an age of 10 to 12 y old. This length was obtained from only that portion of the population that could be sampled easily, i.e., individuals >3 to 4 y old. The true mean age could have been determined only if smaller, subsurface specimens also had been sampled. Therefore, the true mean age of the population presumably is younger than 12 y. The current population appears to consist mostly of young and middle-age cohorts because the species is known to live to at least 25 y in the river. Many specimens found in the river between 1998 and 2002 were of small

and medium sizes, and in excellent condition, and these facts suggest that the population is reproducing.

Fish hosts

The darters *Percina aurantiaca*, *P. burtoni*, *P. evides*, *Etheostoma blennioides*, and *E. flabellare* were excellent fish hosts because more glochidia transformed to juveniles on these species than on other fish species. *Percina evides* and *E. blennioides* are widespread, abundant, and easy to collect, and they do well in captivity if held in large aquaria with adequate flow. However, *P. aurantiaca* and *P. burtoni* are challenging to collect because they are uncommon or difficult to capture in seine nets. Furthermore, *P. burtoni* is a species of special concern in Virginia and, in our experience, *E. flabellare* has been difficult to keep alive in captivity during the spring (e.g., last entry for *E. flabellare*, Table 4). The high mortality of *E. flabellare* probably was the result of collecting fish during their spring spawning period, when the fish were physiologically stressed because the females were gravid and males aggressive and combative. Thus, use of *P. aurantiaca*, *P. burtoni*, and *E. flabellare* for propagation of *D. dromas* is not currently practical. The remaining 5 hosts identified in this study (*Cottus baileyi*, *E. simoterum*, *P. caprodes*, *P. copelandi*, and *P. roanoka*) were marginally suitable hosts on which few juvenile mussels transformed.

Time to transformation of glochidia varied among fish-host trials for unknown reasons; however, observed variation in transformation rates may have been regulated by mean water temperature and maturity of glochidia (Table 4). Glochidial infestations induced in the autumn and winter were successful in identifying host fish; however, many glochidial valves and premature juveniles were sloughed from host fish 2 to 3 wk after infestation. The juveniles produced from these host fish in autumn and winter were of poor quality; pedal locomotion was slow, and many never moved at all. All of these juveniles died within 1 wk of our efforts to culture them, indicating the glochidia were immature at the time of their removal from the female mussel.

Maturity of glochidia and culture of juveniles

Developing methods to obtain mature glochidia is a significant challenge in the effort to

culture *D. dromas* and other endangered mussel species effectively. Ideally, gravid female *D. dromas* should be collected for propagation in April and early May when the glochidial brood of most females is mature and would be released naturally. Most conglutinates were released in early spring in the Clinch River. Collection of gravid female mussels was difficult during this time of the year in the springs of 2000 through 2003 because the release period coincided with rising river water levels and temperatures. Therefore, some gravid female mussels should be collected while glochidia are still immature in late February and early March, when water levels are usually lower, and should be held at a hatchery until water temperatures exceed 15 to 18°C. Females should be observed until they release their first conglutinates in the hatchery, and then held for 1 to 2 wk at 21 to 23°C in a temperature-controlled tank system to allow the glochidia to mature. Maturity of glochidia for infesting host fish could be determined as conglutinates are released, but conditioning the glochidial brood is an empirical process in need of research to establish rigorous protocols.

Many of the first 50 to 100 juveniles that excysted from a host did not appear to be completely developed. The valves of these juveniles were more compressed than those of later juveniles, the valves were closed, no pedal-feeding was observed, and these juvenile mussels experienced high mortality. Fully developed juveniles were elongate, half-moon to bean shaped, slightly agape, light brown to gray, and were capable of active pedal-feeding.

The static, nonrecirculating culture system was effective for sustaining *D. dromas* juveniles for the first 2 wk. We postulate that this system was effective because it significantly increased food availability while juvenile mussels were pedal-feeding. This culture method ensured that dense concentrations of algae and organic material from the sediment settled into the substratum with juvenile mussels. After 1 d of culture in these conditions, algae were visible in the guts of juveniles. However, this method was less effective for culture of older juveniles. Growth rate of individuals older than 1 mo declined or stagnated, and mortality increased, possibly because juvenile mussels became more reliant on filter feeding as they aged (Yeager et al. 1994).

The keys to obtaining good results with this

culture method are to: 1) keep juvenile mussels at cool temperatures (~21°C); 2) change algae, water, and sediment every 2 to 3 d; and 3) feed the juveniles algae of sufficient quantity (1×10^5 to 1×10^6 cells/mL) and quality (e.g., *N. oculata*, *N. oleoabundans*, or other nutritious algae species <10 µm) (Jones and Neves 2002). These techniques also help facilitate control of predators (e.g., flatworms), bacteria, and fungi, all of which can decrease survival and growth of juvenile mussels during culture.

This method is labor-intensive and can be a substantial daily investment of time if many containers of juvenile mussels are propagated and cultured (Jones and Neves 2002). Rearing juvenile *D. dromas* in nonrecirculating containers is an effective short-term culture method for this species; however, research is needed to increase the effectiveness of recirculating and flow-through hatchery aquaculture systems so that juveniles can be grown to greater sizes in captivity.

Acknowledgements

This study was funded by Tennessee Wildlife Resources Agency, Nashville, Tennessee, and US Fish and Wildlife Service, Asheville, North Carolina. The Virginia Cooperative Fish and Wildlife Research Unit is supported jointly by the US Geological Survey, Virginia Department of Game and Inland Fisheries, Wildlife Management Institute, and Virginia Polytechnic Institute and State University.

Literature Cited

- AHLSTEDT, S. A. 1991. Twentieth century changes in the freshwater mussel fauna of the Clinch River (Tennessee and Virginia). *Walkerana* 5:73-122.
- AHLSTEDT, S. A. 1995-1996. Biological assessment of proposed United States Corps of Engineers navigation dredging and disposal on freshwater mussels at sites downstream from Cordell Hull Dam, Cumberland River Miles 304.3-308.9. *Walkerana* 8:177-185.
- AHLSTEDT, S. A., AND J. J. JENKINSON. 1987. A mussel die-off in the Powell River, Virginia and Tennessee, in 1983. Pages 21-28 in R. J. Neves (editor). *Proceedings of the Workshop on Die-offs of Freshwater Mussels in the United States*. Virginia Polytechnic Institute and State University, Blacksburg, Virginia.
- AHLSTEDT, S. A., AND T. McDONOUGH. 1995-1996.

- Summary of pre-operational monitoring of the mussel fauna in the upper Chickamauga Reservoir (Tennessee River) in the vicinity of TVA's Watts Bar Nuclear Plant. *Walkerana* 8:107-126.
- AHLSTEDT, S. A., AND J. D. TUBERVILLE. 1997. Quantitative reassessment of the freshwater mussel fauna in the Clinch and Powell rivers, Tennessee and Virginia. Pages 72-97 in K. S. Cummings, A. C. Buchanan, C. A. Mayer, and T. J. Naimo (editors). Conservation and management of freshwater mussels II. Upper Mississippi River Conservation Committee, Rock Island, Illinois.
- BRUENDERMAN, S. A., AND R. J. NEVES. 1993. Life history of the endangered fine-rayed pigtoe (*Fusconia cuneolus*) (Bivalvia: Unionidae) in the Clinch River, Virginia. *American Malacological Bulletin* 10:83-91.
- CLARK, G. R. 1980. Study of molluscan shell structure and growth lines using thin sections. Pages 603-606 in D. C. Rhoads and R. A. Lutz (editors). Skeletal growth in aquatic organisms. Plenum Press, New York.
- HEARD, W. H., AND R. H. GUCKERT. 1971. A re-evaluation of the recent Unionacea (Pelecypoda) of North America. *Malacologia* 10:333-355.
- HOGGARTH, M. A. 1999. Descriptions of some of the glochidia of the Unionidae (Mollusca: Bivalvia). *Malacologia* 41:1-118.
- HUBBS, D. 1995. 1994 statewide commercial mussel report. Report No. 95-30. Tennessee Wildlife Resources Agency, Nashville, Tennessee.
- JENKINS, R. E., AND N. M. BURKHEAD. 1993. Freshwater fishes of Virginia. American Fisheries Society, Bethesda, Maryland.
- JONES, J. W., AND R. J. NEVES. 2002. Life history and propagation of the endangered fanshell pearly mussel, *Cyprogenia stegaria* Rafinesque (Bivalvia: Unionidae). *Journal of the North American Benthological Society* 21:76-88.
- MORRISON, J. P. E. 1942. Preliminary report on mollusks found in the shell mounds of the Pickwick Landing Basin in the Tennessee River valley. Bureau of American Ethnology Bulletin 129:339-392.
- NEVES, R. J., AND S. N. MOYER. 1988. Evaluation of techniques for age determination of freshwater mussels (Unionidae). *American Malacological Bulletin* 6:179-188.
- ORTMANN, A. E. 1909. The breeding season of Unionidae in Pennsylvania. *Nautilus* 22:91-103.
- ORTMANN, A. E. 1910a. A new system of the Unionidae. *Nautilus* 23:114-120.
- ORTMANN, A. E. 1910b. The discharge of the glochidia in the Unionidae. *Nautilus* 24:94-95.
- ORTMANN, A. E. 1911. A monograph of the najades of Pennsylvania, part I and II. *Memoirs of the Carnegie Museum* 4:279-342.
- ORTMANN, A. E. 1912. Notes upon the families and genera of the najades. *Annals of the Carnegie Museum* 8:222-366.
- ORTMANN, A. E. 1918. The nayades (freshwater mussels) of the upper Tennessee drainage with notes on synonymy and distribution. *Proceedings of the American Philosophical Society* 57:521-626.
- ORTMANN, A. E. 1925. The naiad fauna of the Tennessee River system below Walden Gorge. *American Midland Naturalist* 9:321-372.
- PARMALEE, P. W., AND A. E. BOGAN. 1998. The freshwater mussels of Tennessee. University of Tennessee Press, Knoxville, Tennessee.
- PARMALEE, P. W., W. E. KLIPPEL, AND A. E. BOGAN. 1980. Notes on the prehistoric and present status of the naiad fauna of the middle Cumberland River, Smith County, Tennessee. *Nautilus* 94:93-105.
- PARMALEE, P. W., W. E. KLIPPEL, AND A. E. BOGAN. 1982. Aboriginal and modern freshwater mussel assemblages (Pelecypoda: Unionidae) from the Chickamauga Reservoir, Tennessee. *Brimleyana* 8:75-90.
- ROBINS, C. R., R. M. BAILEY, C. E. BOND, J. R. BROOKER, E. A. LACHNER, R. N. LEA, AND W. B. SCOTT. 1991. Common and scientific names of fishes from the United States and Canada. 5th edition. Special Publication 20. American Fisheries Society, Bethesda, Maryland.
- SIMPSON, C. T. 1896. The classification and geographical distribution of the pearly fresh-water mussels. *Proceedings of the United States National Museum* 18:295-343.
- SIMPSON, C. T. 1900. Synopsis of the naiades, or pearly fresh-water mussels. *Proceedings of the United States National Museum* 22:501-1044.
- TERWILLIGER, K. 1991. Virginia's endangered species. McDonald and Woodward Publishing Company, Blacksburg, Virginia.
- TURGEON, D. D., J. F. QUINN, A. E. BOGAN, E. V. COAN, F. G. HOCHBERG, W. G. LYONS, P. M. MIKKELSEN, R. J. NEVES, C. F. E. ROPER, G. ROSENBERG, B. ROTH, A. SCHELTEMA, F. G. THOMPSON, M. VECCHIONE, AND J. WILLIAMS. 1998. Common and scientific names of aquatic invertebrates from the United States and Canada: mollusks. 2nd edition. Special Publication 26. American Fisheries Society, Bethesda, Maryland.
- USFWS (US FISH AND WILDLIFE SERVICE). 1983. Dromedary pearly mussel recovery plan. US Fish and Wildlife Service, Atlanta, Georgia. (Available from: Fish and Wildlife Reference Service, 5430 Grosvenor Lane, Suite 110, Bethesda, Maryland 20814 USA.)
- WILSON, C. B., AND H. W. CLARK. 1914. The mussels of the Cumberland River and its tributaries. Report of the Commissioner of Fisheries for 1912. Document No. 781. US Bureau of Fisheries, Washington, DC.

YEAGER, M. M., D. S. CHERRY, AND R. J. NEVES. 1994. Feeding and burrowing behaviors of juvenile rainbow mussels, *Villosa iris* (Bivalvia:Unionidae). *Journal of the North American Benthological Society* 13:217–222.

ZALE, A. V., AND R. J. NEVES. 1982. Fish hosts of four

species of lampsiline mussels (Mollusca: Unionidae) in Big Moccasin Creek, Virginia. *Canadian Journal of Zoology* 60:2535–2542.

Received: 8 December 2003
Accepted: 10 May 2004