

Conservation of the endangered freshwater mussel *Solenia carinata* (Bivalvia, Unionidae) in China

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Abstract

Despite the diversity and economic and ecological value of freshwater mussels, relatively little is known about their biology (especially for species outside of Europe and North America). *Solenia carinata* is an endangered freshwater mussel, is endemic to China and is now only distributed in Poyang Lake basin. However, its conservation status is not clear. Thus, for this study, surveys were conducted at 41 sites along the lower reaches of the Ganjiang River to study the conservation status of *S. carinatus*. The results showed that *S. carinata* had a restricted distribution and extremely low density. In addition, the habitat sediments where *S. carinata* was located were mainly composed of silt (particle size <0.0625 mm). RDA analysis showed that the density of *S. carinata* was correlated to dissolved oxygen, temperature, turbidity and chlorophyll-a. Microsatellite analysis showed that *S. carinata* had a low genetic diversity (mean H_O : 0.419; mean H_E : 0.484; mean PIC: 0.430). At the same time, we firstly report the glochidia of *S. carinata* and describe its morphological characteristic. Surprisingly, its reproduction period and morphological characteristics were different from that of others freshwater mussels. Therefore, this study clarified the resource conditions, endangered status and threat factors for *S. carinata* and it provided a theoretical basis for the conservation and management of its resources.

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Keywords

Solenia carinata, endangered, conservation, glochidia, microsatellite

Introduction

Freshwater mussels are large benthic animals and important components of the freshwater biotic community, they can be important filter feeders and play an important role in aquatic ecosystems (Vaughn and Hakenkamp 2001, Bogan 2008, Geist 2010, Lopes-Lima et al. 2017b). However, over the past few years, the overall situation of freshwater mussels is really alarming (Williams et al. 1993, Lydeard et al. 2004, Strayer et al. 2004, Geist 2010). It is reported that, in a total of 297 freshwater mussel species in North America, there were 19 presumed extinct in 1993; by 2008, the number of extinct freshwater mussels reached 37 (Bogan 1993, 2008, Christian and Harris 2008). The IUCN Red List of Endangered Species listed 247 species of freshwater mussels above the near the threatened level (IUCN 2017). To date, conservation efforts for freshwater mussels are almost non-existent in East and Southeast Asia. The IUCN conservation status assessment for unionoid species could not be completed due to a lack of information on their status, threats and other conservation-related issues (IUCN 2017). National Red Lists for Unionida are available for only four of the 17 countries (i.e. Vietnam, South Korea, Japan and Russia) where they are found. None of the 228 unionoid species in the region is protected by international legislation (Zieritz et al. 2018). Therefore, freshwater mussels are considered one of the most endangered groups of animals in the world, which has caused great concern as they provide the following services (inter alia): providing food resources to higher trophic levels, cleaning the water, controlling of the amount and composition of suspended particles, cycling nutrients and providing of habitat for other organisms (Strayer et al. 2004, Aldridge et al. 2007, Bogan 2008, Vaughn 2012).

The middle and lower reaches of the Yangtze River in China are diverse and abundant in freshwater mussels and they are also hotspots for biodiversity in East Asia. However, more than 80% of freshwater mussels in the region are considered near threatened or threatened (Wu 1998, Prozorova et al. 2005, Shu et al. 2009, Xiong et al. 2012; Zieritz et al. 2018). Poyang Lake is surrounded on three sides by mountains and is fed by five large rivers (Ganjiang, Fuhe, Xiuhe, Xinjiang and Raohe), forming a complex and highly interconnected river–lake–wetland system in a monsoon-dominated sub-tropical climate (Zhang et al. 2013). Poyang Lake basin has the typical natural ecosystem characteristics of the Yangtze River basin, its natural environment is healthy and freshwater mussel species are extremely rich. However, approximately 75% of freshwater mussels are endemic to China and the population of freshwater mussels continues to decline (Wu et al. 2000b, Xiong et al. 2012). Threats to freshwater mussels include extreme climate changes, sand excavation, pollution, dam construction and overharvesting, but human disturbance is the major factor affecting freshwater

mussels (Wu 1998, Zhang et al. 2013, Lopes-Lima et al. 2017b, Zieritz et al. 2018). These factors have resulted in a decrease in freshwater mussel diversity and a change in their community structure (Wu 1998, Zhang et al. 2013, Zieritz et al. 2018).

S. carinata is an endemic unionoid species in China and currently, it is only distributed in Poyang Lake basin (Wu 1998, Lopes-Lima et al. 2017a). In 1877, Heude found fossil fragments in the lower reaches of the Yangtze River and thought they were a new species; however, since that time, there were no more relevant reports on this species until 1991, when Liu and Wu found live specimens of *S. carinata* in Wucheng Town, Poyang Lake region and gave an additional description (Liu and Wu 1991). In addition, the fragmentary fossil specimens, found in the quaternary of the conglomerate layer of the Yellow River bank, indicated that *S. carinata* once lived in northern China (Savazzi and Yao 2010). *S. carinata* preferred to inhabit silt or hard clay, with its narrow front end and strong axe helping it dig down into the sediment to resist erosion from flood waters (Liu and Wu 1991, Wu 1998, Savazzi and Yao 2010). Due to their delicious taste and high nutritional value, like other *Solenia* species, *S. carinata* was also targeted by local people for food (Liu and Wu 1991). However, over the past few years, due to environmental degradation and overfishing, the numbers of *S. carinata* have fallen sharply.

There are few studies on *S. carinatus* and specifically, there is little basic information on the species. The purpose of this study is to survey the distribution, density, age structure and habitat characteristics of *S. carinata* in the lower reaches of the Ganjiang River and to clarify its status, endangered condition and threat factors to provide the basis for its protection. In addition, we also hope that the government departments of China can strengthen the protection for freshwater mussel diversity using the information from this study.

Materials and methods

Study area

The Ganjiang River (116°01'–116°22'E, 25°57'–29°11'N) is the largest river running north to south in Jiangxi Province, China, flowing into Poyang Lake and it is one of the important tributaries (7th largest) of the Yangtze River. The Ganjiang River covers a total catchment area of 82809 km² and its main channel is 823 km. In addition, it has a complex river system and is in the mid-subtropical humid monsoon climate zone. Precipitation is abundant with an average of 1580.8 mm/year.

Sampling method

A total of 41 sampling sites were established in three major tributaries of the lower reaches of the Ganjiang River (the main distribution area of *S. carinata*) from September 2016 to March 2017 (Figure 1). A triangular mussel harrow (550 mm wide,

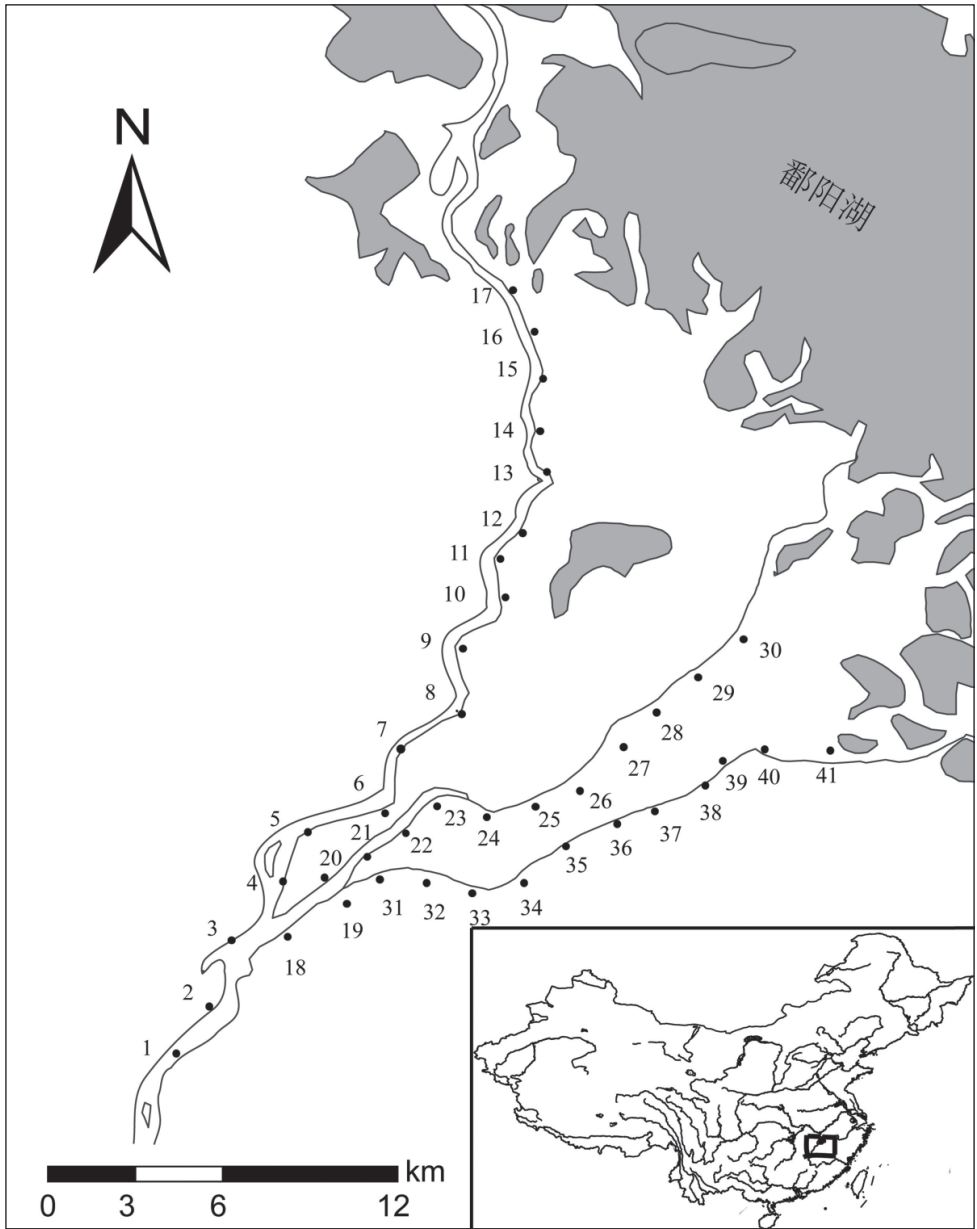


Figure 1. Sampling map of *S. carinata*.

20 mm mesh, rake tooth spacing 15 mm) was used for quantitative sampling. The harrow was dragged slowly by hand in the shallows and the sandy beach of the river (0.5–2.0 m of depth) that had a total area of 2.2 m². The distance between each site was approximately 2 km and each sampling site had 5 quadrats (distance between each quadrat was 100 m). Shell length, shell width and shell height (mm) of live mussels

was measured with a dial caliper (± 0.1 mm). Live mussels and attached dreissenids were weighed separately (± 0.1 g). The specimens were counted and then returned to their original habitat. In addition, when we found glochidia, the whole animal was preserved in 75% alcohol and was taken back to the laboratory.

Habitat characteristic analysis

Temperature (T), turbidity (TURB), salinity (Sal), pH and dissolved oxygen (DO) were measured by a water quality meter (AQUAREAD, AP-800) and Chlorophyll-a (Chl-a) was measured by a chlorophyll meter (HL-168C06, made in China).

The habitat sediment samples of *S. carinata* were collected by a special pipe, emptied into sealable plastic bags and taken back to the laboratory for sediment particle size analysis. Samples were oven-dried at 105 °C for 24 hours (Gordon et al. 2004) and then sieved through 3 sieves with mesh sizes of 4 mm, 2 mm and 0.0625 mm with hand shaking for 30 minutes. Percentages of pebbles (>4 mm), granules (2–4 mm), sand (0.0625–2 mm) and silt (<0.0625 mm) in the sediment samples were determined (Wentworth 1922).

Analysis of age structure

Analysis of age structure used mussel shells that were obtained by hand-collection from the shallows and the sandy beach of the river. The method of thin section was used for the analysis of age structure (Hua et al. 2001). First, we cut from the top of the shell to the ventral margin along the maximum vector direction of the growth line and we made the inner section at right angles to the growth line. At the same time, we made a second cut on half of the shell and then chose the thin layer of shells which were bonded to a glass slide by a colourless transparent epoxy glue (Hua et al. 2001). Finally, the prepared inner sections were burnished until they were smooth and polished (Hua et al. 2001). Moreover, they were observed with a TCA-10 model stereomicroscope (3664 × 2748 pixel resolution) and then we recorded the number of growth lines (Figure 2).

Morphological observation of glochidia

Mature glochidia were collected from the demibranches of *S. carinata* gravid females and fixed in 75% ethanol for investigation under light microscopy. Measurements of shell length, shell height and hinge length of 30 glochidia were taken under a light microscope equipped with a calibrated ocular micrometer.

For SEM, specimens were washed with 0.65% saline water, then fixed for 12 h in glutaraldehyde (4 °C), macerated with 0.1 mol/l phosphate buffer (pH = 7.2) three



Figure 2. The internal section of *S. carinata*.

times, dehydrated in a graded ethanol series (30 min each at 30%, 50%, 70%, 90%, 95% and three times in 100%) and transferred to isoamyl acetate for 30 min. They were critical-point dried with liquid CO₂ and coated with gold. Observations were made with a scanning electron microscope (S-570) and photographs were taken.

Redundancy analysis

Redundancy analysis (RDA), a multivariate direct gradient analysis technique, was used to evaluate the variations in density in relation to environmental variables. Detrended correspondence analyses indicated that the *S. carinata* dataset had a short gradient length, indicating that the linear model of RDA was more appropriate than canonical correspondence analysis (CCA; ter Braak and Verdonschot 1995). Similar to our regression analysis, we performed RDA to assess the correlations between *S. carinata* densities; we then performed RDA to determine how the physicochemical parameters correlated to *S. carinata* density. All variables were entered in the analysis after a forward selection procedure to show their importance in explaining the total variability in *S. carinata* density. The significance ($P < 0.05$) of the RDA gradient was assessed by Monte Carlo permutation tests and their importance was measured by the eigen values of the first 2 axes (ter Braak and Verdonschot 1995). All *S. carinata* density and environment data were $\log_{10(x+1)}$ transformed to meet the assumptions of multivariate normality and to moderate the influence of extreme data. All the ordinations were performed using CANOCO 4.5 (ter Braak and Verdonschot 1995).

Microsatellite analysis of genetic diversity

A total of 27 specimens of *S. carinata* were collected in the lower part of the Ganjiang River. The specimens were taken to the laboratory where tissues were dissected and preserved at -80 °C. DNA was extracted from the foot tissue for genetic analysis using the TIANamp Marine Animals DNA Kit (TianGen). We used 19 primer sets (scastt1, scastt2, scastt3, scastt4, scastt5, scastt6, scastt7, scastt8, scastt9, scastt10, scastt11, scastt12, scastt19, scastt21, scastt22, scastt23, scastt24, scastt27 and scastt33) devel-

oped by Sun et al. (2016) for PCR amplification of microsatellite loci. Amplification conditions are described in Sun et al. (2016). Amplification products were analysed on an ABI 3730 automated sequencer and scored using GENEMAPPER v. 3.7 (Applied Biosystems) with a ROX-labelled size standard.

The number of alleles (N_A), observed heterozygosity (H_O) and expected heterozygosity (H_E) and tests for deviation from Hardy-Weinberg Equilibrium (HWE) were calculated by POPGENE v. 1.32 (Yeh et al. 2000). CERVUS v. 3.03 (Kalinowski et al. 2007) was used to calculate the polymorphism information content (PIC).

Results

The distribution of *S. carinata*

The distribution of *S. carinata* in the Ganjiang River is shown in Figure 3. *S. carinata* was presented in five of the 41 surveyed sites (12.2%, sites 11, 12, 14, 27 and 29). Historical information on the distribution of *S. carinata* described this species as colonising the lower stream of the Ganjiang River, Xiuhe River, Fuhe River, Xinjiang River and Poyang Lake (Figure 3).

The mean density of *S. carinata*

There were significant differences in the mean densities of *S. carinata* amongst the discovery sites ($p < 0.05$) (Figure 4). The mean density of *S. carinata* was the highest at site 27 (1.54 ± 1.35 ind./m²) and the mean densities of the other four sites were 0.55 ± 0.81 ind./m² (site 14), 0.18 ± 0.41 ind./m² (site 29), 0.09 ± 0.20 ind./m² (site 11) and 0.09 ± 0.20 ind./m² (site 12), respectively.

Age structure of *S. carinata*

The age range of *S. carinata* was 2–8 years and the number of 5-year-old individuals was the greatest, accounting for 29.6% of the total number of individuals. The number of 2-year-old individuals was the next highest, accounting for 22.2% of the total individuals and the number of 8-year-old individuals was the lowest, accounting for 3.7% of the total number of individuals (Table 1).

Habitat characteristic of *S. carinata*

The habitat sediments of *S. carinata* were mainly composed of silt and the silt content at site 11 was 90.83%, followed by the silt content at site 27 being 84.59% (Table 2). The

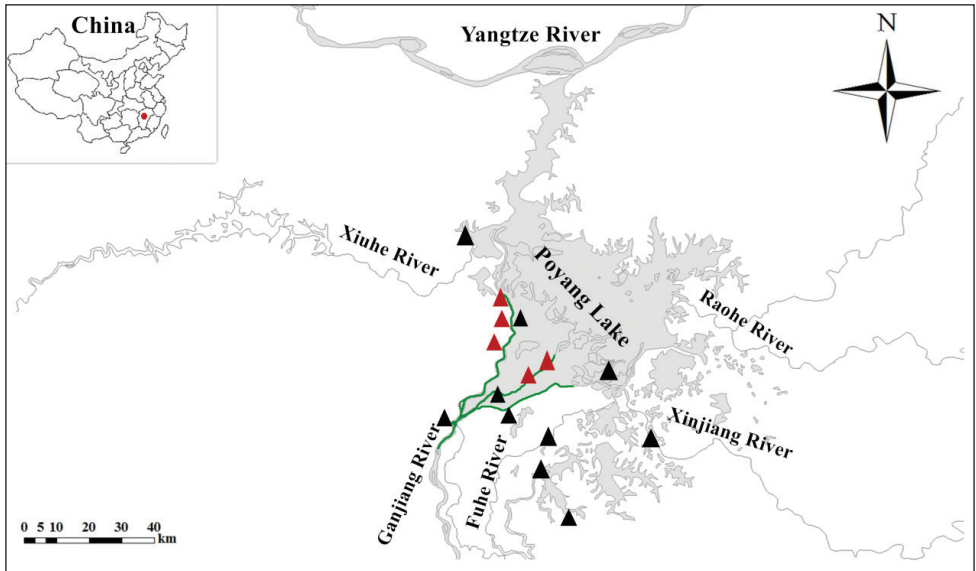


Figure 3. The distribution of *S. carinata* (Red triangle: In the study; Black triangle: Historical research; Green line: In the survey area; Historical data sources: Liu and Wu. 1991; Wu et al. 1994; Wu et al. 2000b; Liu et al. 2008; Xiong et al. 2011; Xiao et al. 2012; Zhang et al. 2013; Huang et al. 2013).

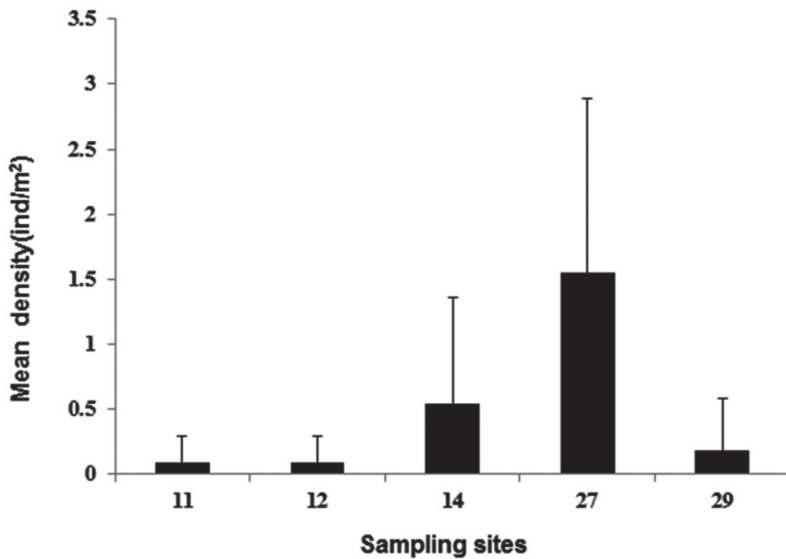


Figure 4. The mean density of *S. carinata* along the 5 sites with mussels in the Ganjiang River.

temperature range of all sites was 11.20–29.10 °C; the pH range of all sites was 6.68–7.10; the dissolved oxygen range of all sites was 3.46–11.10 mg/l; the salinity range of all sites was 0.02–0.10 mg/l; the turbidity range of all sites was 7.20–107.00 mg/l; the

Table 1. Shell shape data and age structure of *S. carinata*.

Number of samples	Shell length(mm)	Shell height(mm)	Shell width(mm)	Body-weight(g)	Age
1	126.4	25.6	16.8	75.6	2
2	137.2	27.6	20.1	80.1	2
3	145.3	30.4	22.4	81.7	2
4	145.5	30.6	22.6	82.1	2
5	157.6	32.4	24.3	101.1	2
6	164.3	34.4	25.5	109.5	2
7	171.1	26.9	35.4	113.5	3
8	178.0	37.4	28.3	122.1	3
9	195.3	39.4	30.2	144.2	4
10	198.4	40.4	30.9	151.3	4
11	205.6	40.8	30.9	174.5	4
12	210.4	41.2	32.1	180.7	5
13	210.7	40.2	31.4	181.6	5
14	217.6	40.9	27.1	193.5	5
15	218.7	42.1	31.7	200.5	5
16	220.4	42.2	31.9	205.4	5
17	221.3	42.2	28.9	208.5	5
18	222.2	41.4	32.6	209.5	5
19	222.6	42.2	31.9	211.4	5
20	225.8	41.8	32.9	217.9	6
21	230.0	46.2	33.5	234.8	6
22	230.5	46.4	34.1	236.3	6
23	238.8	48.2	35.1	289.8	7
24	240.0	48.4	35.3	307.1	7
25	244.3	48.8	35.8	317.9	7
26	245.0	48.9	36.1	320.1	7
27	253.1	50.1	40.1	340.9	8

Table 2. Characteristics of sediment particle size of *S. carinata*

Particle size	Site 11	Site 12	Site 14	Site 27	Site 29
Silt (%)	90.83	18.52	75.44	84.59	23.08
Sand (%)	9.17	61.23	23.01	15.41	76.48
Granules (%)	0	8.21	1.55	0	0.44
Pebbles (%)	0	12.04	0	0	0

Note: Grading standards for particle sizes are from Wentworth (1922).

chlorophyll-a range of all sites was 5.75–13.54 mg/l (Table 3). The results of RDA analysis showed that the mean density of *S. carinata* was positively correlated with dissolved oxygen, temperature, turbidity and chlorophyll-a (Figure 5).

Table 3. Environmental factors of *S. carinata* (T: temperature; DO: dissolved oxygen; Sal: salinity; TURB: turbidity; Chl-a: Chlorophyll-a).

Factors	Site 11	Site 12	Site 14	Site 27	Site 29	All sites	
						Mean ±SE	Range
T	20.3	20.4	20.5	19.3	19.2	20.3 ± 7.1	11.2–29.1
pH	6.74	6.72	6.72	7.03	7.04	6.94 ± 0.16	6.68–7.10
DO	7.18	7.13	7.22	8.01	8.19	7.60 ± 1.20	6.46–11.10
Sal	0.05	0.05	0.05	0.04	0.04	0.05 ± 0.01	0.02–0.10
TURB	47.7	26.4	36.8	57.2	57.8	49.4 ± 22.9	7.2–107.0
Chl-a	6.14	6.91	6.44	12.15	13.54	8.40 ± 2.00	5.75–13.54

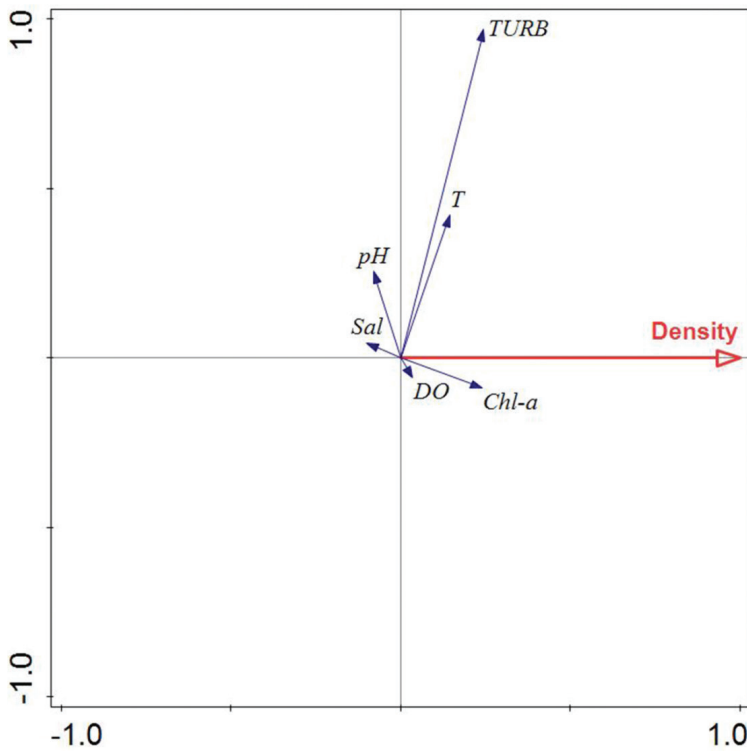


Figure 5. Analysis of correlation between environmental factors and mean density of *S. carinata* (T: temperature; DO: dissolved oxygen; Sal: salinity; TURB: turbidity; Chl-a: Chlorophyll-a).

The morphology of glochidia of *S. carinata*

Mature glochidia of *S. carinata* were from December to February of the following year. *S. carinata* had marsupium in all 4 demibranches, the hinge length of glochidia was $31.8 \pm 2.9 \mu\text{m}$ and the length and height of the shell of $58.9 \pm 1.8 \mu\text{m}$ and $51.6 \pm$

2.2 μm , respectively. Moreover, its glochidia were classified as small according to Davis et al. (1981) ($G_{ln} = \text{shell length} \times \text{shell height}$).

The results of the morphology of glochidia showed that they were ovate subtriangular, nearly symmetric and moderately inflated (Figures 6A, B). The hook styli-form was bluntly rounded at the distal end, the microstylets were sparsely and evenly distributed on the hook and the micropoints were distributed densely on the ventral margin of valves, but did not extend to the hinges (Figure 6C). The exterior surface of the shells were densely but shallowly pitted (Figure 6D). The sensory hairs had tufts in one pair, located near the ventral margin of the valve (Figure 6E). Long larvae threads (a diameter of 0.5 μm) wrapped around each other in the shell and protruded out of the shell (Figure 6F).

Genetic diversity of *S. carinata*

A total of 27 individuals of *S. carinata* were successfully genotyped for all 19 microsatellite loci. The results showed that 63 alleles were detected amongst the three sampling locations. The number of alleles (N_A) at each sampling location ranged from 2 to 6 and the mean number of alleles (N_A) was 3.32; the observed heterozygosity (H_o) ranged from 0.143 to 0.796, the mean H_o was 0.419; the expected heterozygosity (H_e) ranged from 0.155 to 0.767, the mean H_e was 0.484; the PIC ranged from 0.124 to 0.708 and the mean PIC was 0.430. After the Bonferroni correction for multiple comparisons, no loci were deviations from HWE (Table 4).

Discussion

Solenia are mainly distributed in East Asia, Thailand and India's Assam (Liu and Wu 1991, Wu 1998, Deekin et al. 2003, Lopes-Lima et al. 2017a). Heude (1874–1885) reported that the genus *Solenia* had 11 species and live specimens were mostly distributed in the south of the Huaihe River, specifically in the middle and lower reaches of the Yangtze River. Moreover, fossil specimens were widely distributed in the Quasi Algor basin of Xinjiang Province, Jilin Province, Hebei Province and Shanxi Province (Heude 1874–1885, Savazzi and Yao 2010, Xu et al. 2003). The fossil specimens of *S. carinata* were once reported as occurring in the quaternary of the conglomerate layer in the Yellow River bank, but now live specimens are only distributed in Poyang Lake basin (Liu and Wu 1991, Savazzi and Yao 2010). *S. carinata* do not move throughout their lives because they always insert themselves into the silt. Therefore, they were highly selective in their habitat choice, especially in the sediment. Moreover, they preferred to inhabit the silt and stable mussel beds. The decrease in the distribution range of *S. carinata* was likely caused by human disturbance, long-term environmental changes and habitat destruction (Wu 1998, Xiong et al. 2012, Zhang et al. 2013, Zieritz et al. 2018). In this study, *S. carinata* only occurred in 5 of the 41 surveyed sites in the lower

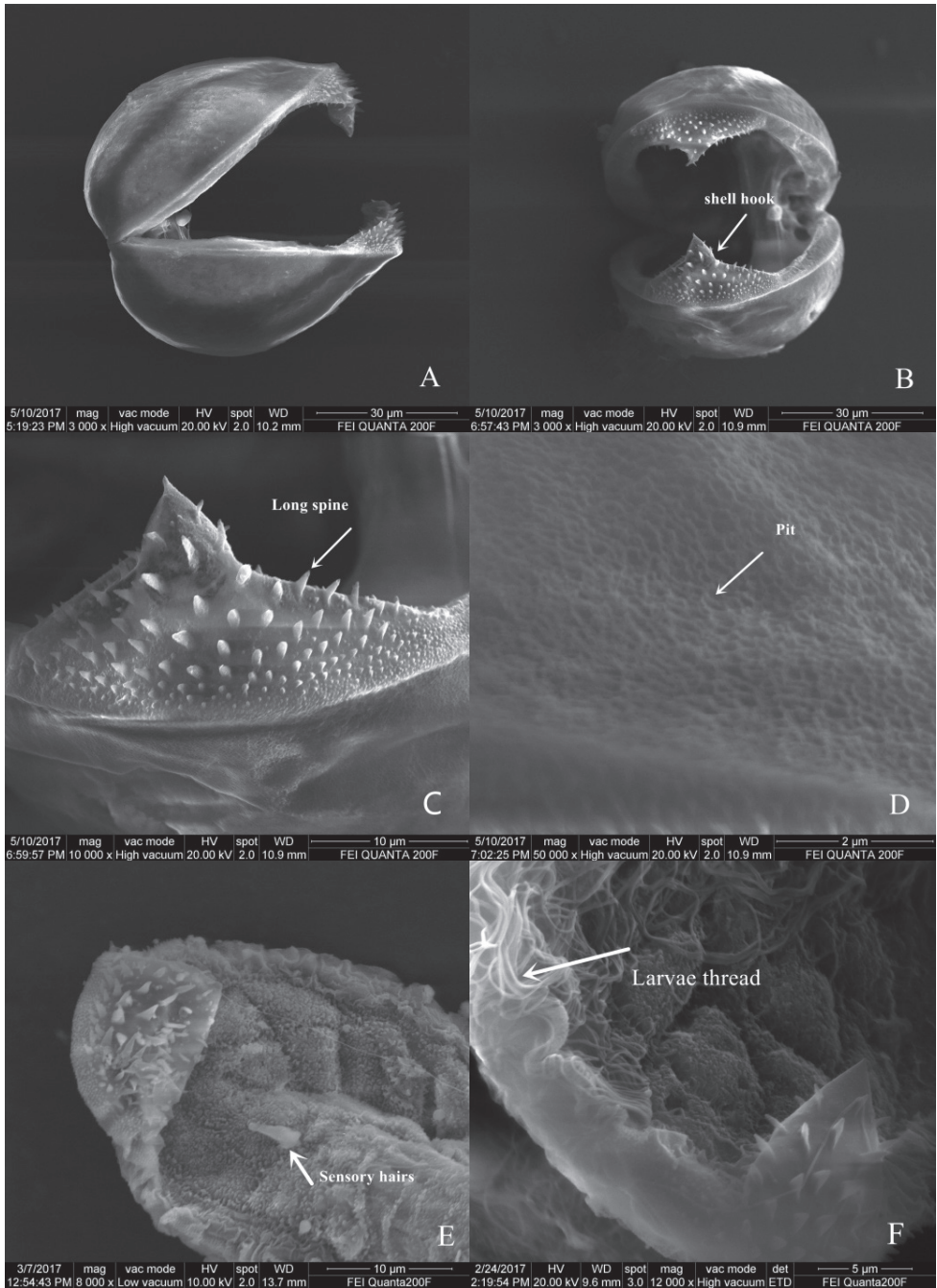


Figure 6. The morphology of glochidia of *S. carinata* (A lateral B ventral valve C hook with large spines D the exterior valve sculpture E sensory hair tufts F the larvae thread).

Table 4. Population genetic parameters in three populations of *Solenia carinata*. N_A : the number of alleles, H_O : observed heterozygosity; H_E : expected heterozygosity.

Locus	N_A	H_O	H_E	PIC	F_{is}
scastt1*	4	0.551	0.657	0.614	0.165
scastt2	2	0.434	0.463	0.357	0.041
scastt3*	2	0.153	0.155	0.134	-0.077
scastt4	3	0.341	0.526	0.449	0.401
scastt5*	5	0.659	0.718	0.652	0.048
scastt6	3	0.143	0.321	0.264	0.529
scastt7	2	0.240	0.265	0.213	0.073
scastt8	2	0.210	0.265	0.213	0.073
scastt9	3	0.189	0.290	0.236	0.350
scastt10*	4	0.551	0.767	0.697	0.231
scastt11	4	0.403	0.533	0.449	-0.093
scastt12	3	0.551	0.532	0.432	-0.093
scastt19	4	0.510	0.377	0.592	0.209
scastt21	5	0.617	0.569	0.527	-0.106
scastt22	3	0.474	0.517	0.458	0.102
scastt23*	2	0.353	0.467	0.348	0.123
scastt24	3	0.331	0.521	0.447	0.360
scastt27*	3	0.454	0.489	0.382	0.039
scastt33	6	0.796	0.761	0.708	-0.066

Note: *: significant deviations from Hard-Weinberg equilibrium ($p < 0.05$).

reaches of the Ganjiang River and this result indicated that its distribution range was limited in the region. Compared with *S. oleivora*, although both have similar morphology and life styles, the population size, density and distribution of *S. carinata* were much smaller than that of *S. oleivora* and the ratio of *S. carinata* and *S. oleivora* from the catch of local fishermen was 1:37.5.

Environmental factors, such as sediment, dissolved oxygen and water turbidity, significantly affect the distribution of freshwater mussels (Zhao et al. 2016, Negi and Mamgain 2013). The ability of freshwater mussels to spread was weak and they did not move; thus, they are less tolerant of the environment. In general, muddy areas had more abundant organic matter and freshwater mussels prefer to live in this habitat (Akiyama and Maruyama 2010). *S. carinata* is a relatively large-bodied mussel, remaining almost completely sessile as an adult (more so than most unionids) and it burrows in soft mud and clay sediments near riverbanks (Liu and Wu 1991, Yao and Wu 2001, Liu et al. 2017). We found that the habitat sediments of *S. carinata* were mainly composed of silt and the moderate ratio of mud or sand can form a hard substrate structure, which kept mussel beds stable and protected from the impacts of water flow.

Some studies have shown that environmental factors such as water temperature, DO and turbidity influence benthic community structure. Dissolved oxygen was one

of the important factors which affects the distribution and abundance of macrozoobenthos. For example, a positive correlation was found between the macrozoobenthos diversity and dissolved oxygen at all of the sites during the present investigation which is in accordance with the findings of Joshi et al. (2007) who reported maximum benthic diversity during the winter season when the amount of dissolved oxygen is higher and the temperature is low (Negi and Mangan 2013). Chlorophyll-a was another factor that affected the distribution and abundance of macrozoobenthos. Within a certain range, phytoplankton can provide rich food for macrozoobenthos and promote their reproduction and growth. However, when water had a high concentration of chlorophyll-a, algae density was also higher, which would restrain the biomass and distribution of macrozoobenthos (Stout and William 1985). Turbidity was also an important environment factor by its influence on the distribution of macrozoobenthos (Martin et al. 2008). In this study, we also found that the distribution of *S. carinata* was correlated to dissolved oxygen, water temperature, turbidity and chlorophyll-a. In addition, strikingly, the number of *S. carinata* was greater in water with high turbidity, which indicated that high turbidity of water could provide rich food sources for the reproduction of *S. carinata*.

Freshwater mussels are selective and adaptive to habitats (Vaughn 2012). Some studies called a region or niche of mussels gathered as “mussel bed” (Brainwood et al. 2008, Vaughn 2012). A “mussel bed” was the result of long-term adaptation of mussels to the hydrological situation and characteristics of the river (Vaughn 2012). Diversity and richness varied greatly amongst different microhabitats (Hornbach et al. 2010). Different mussels had different diffusion abilities and changing water level was one of the important factors that affected the survival and dispersion of mussels (Spooner et al. 2012; Jones and Neves 2011; Vaughn 2012). The average water level in the lower reaches of the Ganjiang River was 13.51 m (Wusong elevation), the lowest water level was 9.09 m (January) and the highest water level was 17.82 m (July). There were lots of “mussel beds” in the lower reaches of the Ganjiang River, especially in the low water level zone. When the water level of Nanchang reaches dropped to approximately 8.0 m (Wusong elevation), some “mussel beds” are exposed. The movement ability of mussels varied amongst the different water levels. The life style of *S. carinata* is almost completely sessile as an adult (more so than most unionids), remaining burrowed in soft mud and clay sediments near river banks (Figures 7A, B). Although *S. carinata* usually inhabited the low water level zone (9–10 m), when the water level was below 9m or much more than 10 m, the survival of *S. carinata* would be affected. Therefore, the coupling between the survival of *S. carinata* and the distribution of “mussel beds” and changes in water levels is the results of a long-term adaptation of the mussels to the water level in the river.

The reproduction characteristics of freshwater mussels and the morphology of glochidia were important to its classification and phylogeny (Heard and Gluckert 1970; Wu et al. 1999, Graf and Foighil 2000, Lopes-Lima et al. 2017a). *S. carinata* had a marsupium in all 4 demibranches. The reproduction type of *S. carinata* was similar to that of *Lamprotula*, such as *L. caveata*, *L. cornuum-lunae*, *L. scripta* and *L. lei* and, for this reason, *Lamprotula* and *Solenaia* were previously thought to have a close relation-

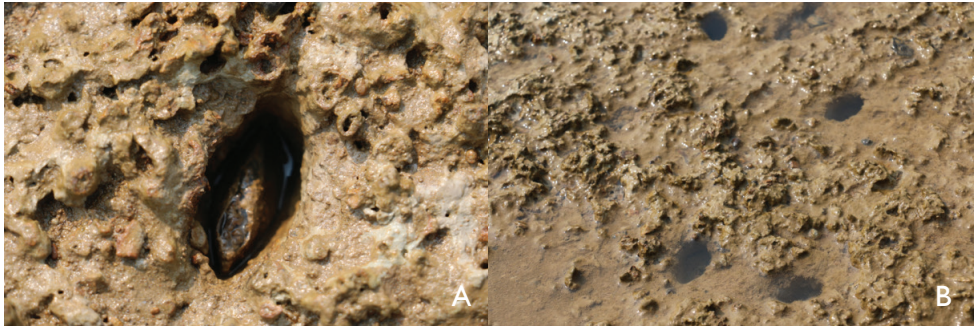


Figure 7. The life style of *S. carinata* (A *S. carinata* used its narrow front end and strong axe to keep digging down to resist erosion from flood waters B *S. carinata* preferred to inhabit silt or hard clay).

ship (Wei et al. 1994, Wu et al. 2000a). However, the glochidia of *S. carinata* were hooked, while those of *Lamprotula* were hookless. It is unfortunate that no information is available on potential host fishes.

Glochidia of *S. carinata* were small-sized and numerous, which could promote population viability based on increasing the number of juveniles and shrinking the morphology size of glochidia. This study indicated that the reproduction period of *S. carinata* was from December to February the following year. In this period, Poyang Lake basin was in its dry season, which resulted in many freshwater mussels and fish being easily harvested. However, the reproduction period of *S. oleivora* was in May and, in this period, the middle and lower reaches of the Yangtze River were in their wet season and fish activity was high, which was beneficial to the parasitism of glochidia. Droughts not only affected the survival of freshwater mussels, but also led to a reduction in host fishes. These factors were likely to be reasons for a sharp decline in the *S. carinata* population.

Higher levels of genetic diversity amongst populations of aquatic organisms could improve evolutionary potential for dealing with habitat change, effects of pathogen infection and other selective forces (Freeland et al. 2011, Liu and Yao 2013, Wu et al. 2013, MacDonald et al. 2017). However, the genetic diversity of *S. carinata* (mean H_O : 0.419; mean H_E : 0.484; mean PIC: 0.430) was lower than *Sinohyriopsis cumingii* (mean heterozygosity ranged from 0.617 to 0.750), *S. oleivora* (mean H_O ranged from 0.501 to 0.620; mean H_E ranged from 0.598 to 0.701) and *L. caveata* (mean H_O : 0.455; mean H_E : 0.835; mean PIC: 0.795) (Luo 2006, Min et al. 2015, Xu 2014), which indicated that *S. carinata* was less adaptive to the environment and populations were prone to decay and extirpate. Narrow-range endemics like *S. carinata*, as well as rare and imperilled species, often have lower genetic diversity and may have experienced genetic bottlenecks (Hamrick and Godt 1989). Genetic bottlenecks in a narrow-range endemic could be the result of the initial founder event that led to speciation in *S. carinata* (Freeland et al. 2011).

Human activities such as dam construction, sand mining, water pollution and overfishing could seriously affect the survival of freshwater mussels (Burlakova et al. 2011, Geist 2011, Lopes-Lima et al. 2017b). Large-scale and unregulated sand mining operations not only damaged the habitats of freshwater mussels, but also resulted in

refloated heavy metals and toxic substances in the water, which could be dangerous for freshwater mussels. Similarly, construction of dams would change the river hydrological characteristics, which affected the colonisation of juveniles and reduced the survival rate of juveniles (Mueller et al. 2011). In addition, dam construction also affected the survival of host fishes. For *S. carinata*, we should not only protect and manage their own river habitats, but also prevent the discharge of pollutants in upper reaches of the river and sustain appropriate flow rates, especially in the dry season (Saunders et al. 2002). Recently, global climate change had resulted in an earlier and prolonged dry season. The water levels in the Ganjiang River have also continued to decline and some reaches of the river are below the drought alarm level. During this period, *S. carinata* have been exposed to drought conditions for a long time, which has led to death and, moreover, it has also been easy for fishermen to capture lots of *S. carinata* by using their tools. Freshwater mussels are highly dependent on host fishes. However, fishermen overfished host fishes due to commercial interests, which could affect the survival and communication of freshwater mussels amongst lakes. Therefore, it is necessary to prevent overfishing to protect freshwater mussel diversity.

Despite the diversity and value of freshwater mussels, relatively little is known about the biology of many of these species (especially species outside of Europe and North America) (Zieritz et al. 2018). Currently, a special conservation area for freshwater mussels has not been established in China. In addition, studies on freshwater mussels are mainly concentrated on resource surveys in some regions (e. g. Yangtze River; Wu 1998, Xiong et al. 2012; Zhang et al. 2013), but there are still many gaps in the information on the conservation status of these species and research efforts are almost non-existent (Lopes-Lima et al. 2014). To a large extent, the lack of protection for freshwater mussel species in China is due to the lack of knowledge on their status, threats and other conservation-related issues. Scientific studies should specifically investigate their threats and known host fish identities in China (Zieritz et al. 2018). The lack of Chinese freshwater mussel species in the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) is particularly surprising in that the commercial use and trade of these animals is most likely more intense in China than anywhere else in the world (Fiske and Shepherd 2007, Ali and Cartier 2013, Ng et al. 2016). Therefore, conservation biology research on freshwater mussels in China is very important. Overall, urgent management measures devoted to the conservation of freshwater mussels are required to: 1) raise ecological protection awareness; 2) prevent commercial capture; 3) strengthen protection efforts for host fishes and habitats of freshwater mussels and 4) prolong the closed fishing season appropriately, which can provide a good ecological environment for the reproduction of freshwater mussels.

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