Glochidia morphology of *Uniandra contradens* Lea, 1838 from Vietnam

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Morphology of bilaterally asymmetrical (unequivalve, unequilateral) glochidia of the freshwater bivalve *Uniandra contradens* Lea, 1838 from the Binh Thien Lake, southern Vietnam, was studied by scanning electron microscopy. Unique characteristics of the outer valve microsculpture is described. Literature data on the south-east Asian freshwater mussels with asymmetrical glochidia are reviewed.

**Key words:** Bivalvia, glochidium, morphology, bilaterally asymmetrical, *Uniandra contradens*, Vietnam.

Family Unionidae (commonly referred to as naiads or freshwater mussels) differs in comparison to other freshwater bivalves (e.g. Sphaeriidae and Cyrenidae) because of their unique life history with a short, obligate parasitic stage (glochidium). Glochidia

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and its various morphological types have been important characters in freshwater mussel systematics for over a century. The glochidia of the south-east Asian freshwater mussels can be divided into two groups: (1) bilaterally symmetrical (equivaleve, equilateral) glochidia with two hooked (for example, *Lanceolaria* Conrad, 1853) or hookless valves (*Pilsbryoconcha* Simpson, 1900, *Hyriopsis* Conrad, 1853, *Chamberlainia* Simpson, 1900, *Scabies* Haas, 1911) and (2) bilaterally asymmetrical (unequivaleve, unequilateral) glochidia with a large marginal appendage at only one of the two larval valves [Ortmann, 1916; Panha, Eongprakornkeaw, 1995; Deein et al., 2008; Pfeiffer, Graf, 2015]. Bilaterally asymmetrical glochidia have been reported from five Asian genera: *Pseudodon* Gould, 1844, *Physunio* Simpson, 1900, *Solenia* Conrad, 1869 (subfamily Gonideinae), *Trapezoideus* Simpson, 1900, *Uniandra* Hass, 1912 (=*Contradens* Haas, 1913) (subfamily Rectidentinae) [Ortmann, 1916; Panha, Eongprakornkeaw, 1995; Deein et al., 2008].

It is necessary to mention that Haas [1969] listed *Unio contradens* Lea, 1838 from Java as the type species of genus *Contradens* and recognized nine species in the genus. He also considered *Unio inaequalis* Rochebrune, 1882 as the type species of another genus, *Uniandra*. Later Brandt [1974] regarded *U. inaequalis* as a junior synonym of *U. contradens*; so, he listed *Contradens* as a synonym of *Uniandra* and *U. contradens* as the type species of *Uniandra*. Heard [1977] also used the genus name *Uniandra*. According to Brandt [1974], the genus name *Uniandra* is used in this paper instead of *Contradens*; classification of subfamilies is given after Whelan et al. [2011] and Huang et al. [2013]. Previously, *U. contradens* together with many other Asian bivalves were included into the family Amblemidae [Panha, Eongprakornkeaw, 1995].

The species of the genus *Uniandra* (with recognised synonyms) are recorded from Indonesia (Java, Kalimantan, Sumatra), Malaysia, Thailand, Cambodia, Laos and Vietnam. In the southern Vietnam, Binh Thien Lake (named as the Peaceful Lake of Heaven) is one of the naturally largest freshwater open lake of the Mekong Delta (Fig. 1). It is located in An Phu District, An Giang Province, linking three communes such as Khanh Binh, Nhon Hoi and Quoc Thai (10°54′46″–10°55′53″ N, 105°03′35″–105°05′49″ E). Nowaday, this lake only adjoins Binh Di River but there is no longer exchange of water with the Hau River (Fig. 2).

Wetland area of the lake in flooding season is about 800 hectares but only 220 hectares in the dry season. It is 4 km long and 500 m wide and relatively low in the northern bank, with a high potential sedimentation. Average depth of the lake is 4 m, deepest area in the middle of the lake is about 6.3 m. During the flooding time, its depth can increase to 8–10 m. Its sediment is mainly silty clay. However, in the time of Mekong delta flooding, most of rivers in the area are muddy with alluvium, but the water of the Binh Thien Lake still remains clean and blue with high biological diversity. A lot of bamboo cover around the lake while many mimosa trees, vegetable shy, water hyacinth bushes and also lotus develop on the surface and shallow parts. In the north part of the lake, foxtail weeds and algae are also abundant.
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**Fig. 1.** A schematic map of the Mekong Delta region in Vietnam with location of the Binh Thien Lake.

**Fig. 2.** View of the Binh Thien Lake.
Although the Mekong delta (including some parts of wetland area of the protected Binh Thien Lake) has been substantially modified by humans, primarily to support rice-based and vegetables agriculture, it still supports a diverse biota where freshwater bivalves play an important role. *U. contradens* (Fig. 3) is a significant part of the Binh Thien Lake ecosystem, so a detailed study of the bivalve lifestyle including data on reproduction and morphology of glochidia is of big interest. Until our study, there were no data on glochidia of *U. contradens* from Vietnam and only glochidia of *U. contradens* from Thailand were investigated [Panha, Eongprakornkeaw, 1995].

The present study is a morphological survey of the glochidial stage of *U. contradens* from Binh Thien Lake (the Mekong River basin), Vietnam, made by light and scanning electron microscopy (SEM).

**Materials and methods**

The mature glochidia were collected from outer demibranchs of gravid female mussels and preserved in 75% ethanol. For SEM they were dehydrated in 95% ethanol followed by a few changes of absolute ethanol. The dried specimens were mounted on the specimen stubs with double sided sticky tap and then coated with gold. Gold was sputtered immediately after drying the samples on the stub to prevent the possibility of deformation of the outer shell layer. The microsculpture of each larval shell was examined at three points: closer to the ventral end, at the center of the valve (the adductor region), and at the ligament. The photographs of glochidia were obtained on the Zeiss EVO 40 scanning microscope at the Biology and Genetic Engineering Center for Collective Use of the IBSS FEB RAS.

Glochidia shell morphology was investigated for the following characters: size, shape, structure of the outer surfaces of the valves. To compare our results with literature data shell shape was analysed by «Glochidial Index» (*Gln*) used after Panha and
Eongprakornkeaw [1995], where \( Gln = \text{shell length} (L) \times \text{shell height} (H) \) (both characters measured in millimeters).

The mollusks collected are stored at the Laboratory of Freshwater Hydrobiology, Institute of Biology and Soil Science, Far Eastern Branch, Russian Academy of Sciences (IBSS FEB RAS), Vladivostok, Russia.

**Results and discussion**

In comparison with the detailed and numerous investigations on bilaterally symmetrical glochidia, the investigations on asymmetric glochidia are still fragmentary and generally devoted to the Thai unionids: *Solenaia khwaenoiensis* Panha et Deein, 2003 [Deein et al., 2008], *U. contradens*, *Physunio superbus* (Lea, 1854), *P. eximius* (Lea, 1856), *Trapezoideus exolescens* (Gould, 1843) [Panha, 1990; Panha, Eongprakornkeaw, 1995]. The study of five mentioned above Thai species were made with scanning electron microscopy; as a result, it was indicated a significant role of the microsculpture of the outer surface of glochidial valves for mussels taxonomy [Panha, Eongprakornkeaw, 1995]. For six investigated species of the genus *Pseudodon* [Kondo, Yamashita, 1980; Kondo, 1987; Graf, Cummings, 2007; Pfeiffer, Graf, 2015], it was shown that *P. cambodjensis* (Petit, 1865) from Cambodia was the only taxon with bilaterally asymmetrical glochidia resolved among the Gonideinae while all other species had hookless larvae.

Glochidia of *U. contradens* are rounded in shape with a large marginal appendage on only one of the two larval valves (Fig. 4). The mean glochidial shell height \( H \) and length \( L \) were 211.5±0.03 and 243.1±0.04 µm, respectively, with \( Gln \) of 0.0505–0.0523. In comparison, glochidia of *U. contradens* from Thailand had \( H=210±0.05 \) µm, \( L=230±0.05 \) µm, with \( Gln=0.0484 \) [Panha, Eongprakornkeaw, 1995].

Among species with bilaterally asymmetric larvae, glochidia of *S. khwaenoiensis* were the biggest, with \( H=220±0.03 \) µm, \( L=280±0.04 \) µm, and \( Gln=0.0612 \) [Deein et al., 2008], while glochidia of *P. superbus* were the smallest: \( H=170±0.03 \) µm, \( L=190±0.03 \) µm, \( Gln=0.00324 \) (see Table).

Ventral rims of each glochidial valve of *U. contradens* were covered by numerous lanceolate microspines and micropoints arranged in more or less complete tight rows. Maximal size of microspines was 1.67 µm (Fig. 5).

External surface of glochidial valves with numerous pores that were 1.77–2.1 µm in diameters. Exterior valve sculpture is coarse (Fig. 6). If we compare our data on valve sculpturing with literature data, the external microsculpture was also coarse for *P. superbus*, *T. exolescens* and *U. contradens* from Thailand, and it was smooth for *P. eximius* [Panha, Eongprakornkeaw, 1995].

There are differences in glochidia brooding among discussed mussels. *U. contradens*, *Uniandra rustica* Sowerby, 1866 (=*C. cambojensis*), *S. khwaenoiensis* are ecto-branchious: only the outer demibranchs are served as marsupia [Ortmann, 1916; Deein
Morphometric characteristics of bilaterally asymmetric glochidia

<table>
<thead>
<tr>
<th>Species</th>
<th>Mean glochidial shell height ($H$, $\mu$m)</th>
<th>Mean glochidial shell length ($L$, $\mu$m)</th>
<th>Glochidial Index ($Gln$)</th>
<th>Reference</th>
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<tbody>
<tr>
<td><em>Uniandra contradens</em> (Vietnam)</td>
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<td>Our data</td>
</tr>
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</tr>
<tr>
<td><em>Physunio superbus</em> (Thailand)</td>
<td>170±0.03</td>
<td>190±0.03</td>
<td>0.00324</td>
<td>Panha, Eongprakornkeaw [1995]</td>
</tr>
</tbody>
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Fig. 4. Glochidia of *Uniandra contradens* from various point of view: $a$ – large marginal appendage; $ms$ – parts of ventral rims with microspines observed from glochidial valves ajar. Scale bar – 50 $\mu$m.
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**Fig. 5.** Ventral rim of glochidial valve: **A** – fragment with microspines; **B** – microspines in large scale. Scale bars – 4 µm (A), 1 µm (B).

**Fig. 6.** External surface of glochidial valves with pores and coarse microsculpturing. Scale bar – 1 µm.

et al., 2008]. In comparison, tetragenous mussels of *Pseudonodon* have all four gills for brooding the larvae [Kondo, 1987]. Information on reproduction of South-East Asian mussels is very sketchy and require further research.

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