

## Two simple methods for observation of shapes in podocypid ostracod valves

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### Abstract

Two simple methods have been developed to study the shapes of podocypid ostracod valves. One is a new observation method by using of a rotation-inclination stage. Using this method, it is possible to observe the shapes of various directions of ostracod valves under the light microscope. The other is a modified method of producing internal mold by using of a petropoxy resin. The modified method can carry out a short time and steady mold finer structure by comparison with the previous method.

Key words: podocypid ostracod, rotation-inclination stage, internal mold

### Introduction

Podocypid ostracod is generally small (about 0.5-1.0mm) laterally compressed Crustacea. The chitinous soft body is enclosed between two valves. Those valves have numerous surface features that closely relate with soft parts of organs. For example, pore canals, muscle scars and ocular sinus indicate the presence of sensory hairs, muscles and lateral eyes respectively. Species with strongly calcified valves are readily fossilized, therefore, those characters on valves provide valuable information for paleontological reconstructions.

The morphological observation methods in ostracods are similar to ones in other micro fossils. However, the laterally compressed bivalve carapace and the presence of many structures inherent in ostracod valves have necessarily led to the development and employment of certain special methods. Several simple and rapid methods of studying the carapace morphology have been developed by several workers. Trievel (1947) invented the heating, staining and moistening with glycerine method which is changed into an opaque valve to easily observe surface features on transparent valves under the light microscope.

Morkhoven (1962) introduced some good paints for staining ostracod valves. Those paints are easily removed again by washing the specimen with water, therefore, those

specimen can be reexamined enclosed features such as radial pore canals, ocular sinus and line of concrescence under transmitted light.

Scanning electron microscopy reveals the details of surface features. However, it is only applicable to surface topography and the coating specimen can no longer be studied in transmitted light. To observe the enclosed features using SEM, Kontrovitz (1982) developed excellent method of producing internal molds.

Both observations by optical and electronic microscope are important for studying carapace morphology. Some other technical methods for observation of ostracod valve were proposed by several workers (Honjo, 1963; Empson, 1982). These methods, however, need special equipment. To observe the shape of ostracod valve, I devised two simple methods.

### Materials and methods

(I) For observation from various directions of ostracod valve, I made a rotation-inclination stage as Fig. 1a. This stage can rotate itself on the microscope stage, furthermore, the innermost disk (ID) and the middle ring (MR) can be inclined at an orthogonal axis each other. Outer diameter of the figured stage was made to fit the diameter of the stage of the light microscope (OLYMPUS B061)(Fig. 1b).

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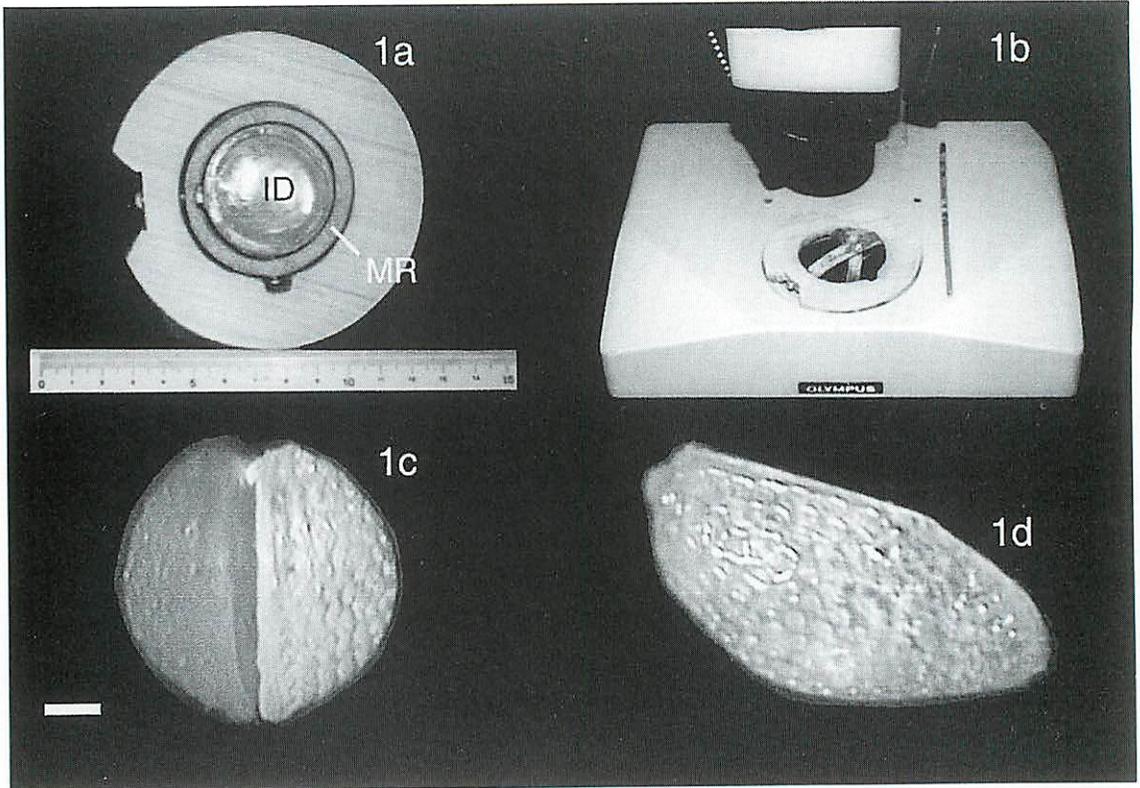


Fig. 1a. Rotation-inclination stage. ID= innermost disk. MR = middle ring. Fig. 1b. Optical microscope (OLYMPUS B061) equipped with the rotation-inclination stage. Figs. 1c, 1d. Posterior view (1c) and anterodorsal view (1d) of male right valve of *Loxoconcha japonica* Ishizaki. The blurring around the margin of the valve of each photograph is due to the depth-of-focus limitation of the microscope. In Fig. 1c, reflective image appears on the left. Scale bar = 0.1 mm for 1c, 1d.

An ostracod specimen is mounted and pasted on the innermost disk. The specimen can be observed from various directions by using of this stage (Figs. 1c, 1d).

(II) For observing internal features, I modified the procedure proposed by Kontrovitz (1982). The modified procedure is summarized as follows:

- 1) To remove the organic materials attached to the valve, specimens are etched with sodium hypochlorite for a day. Juvenile specimens and/or thin shelled specimens are etched for several hours.
- 2) The valve is cleaned by a small brush and is placed in a small beaker filled with distilled water, then the beaker is submerged in an ultrasonic bath.
- 3) The valve is naturally dried in the room.
- 4) The specimen is stuck (it is placed with the convex side down) on a slide glass with double-sided adhesive tape.
- 5) The valve is filled with a petropoxy resin. This resin is a viscous fluid, therefore, the specimen is permeated by the resin little by little with a hair.
- 6) The slide glass attached with the specimen is heated in the oven at 130°C for five minutes.
- 7) The slide glass with the specimen is taken out from the oven, then, cooled.
- 8) The valve is removed from the slide glass. Then, the valve is dissolved with 5% hydrochloric acid.
- 9) After the valve is completely dissolved, the internal mold is washed with a small brush and dried in the room.

Compared with the previous study, improvement points of this method are as follows. 1) petropoxy resin is a viscous fluid which differs from the previous solid material (the Lakeside), therefore, a suitable amount for the necessary can regulate little by little with a hair. 2) the resin hardens with five minutes at 130°C, therefore, the time required to take the internal mold is shortened more than ten minutes. 3) for the special property of this resin, we can steady obtain finer structure (< 0.001 mm) than the previous study (> 0.002 mm) (Fig. 2).

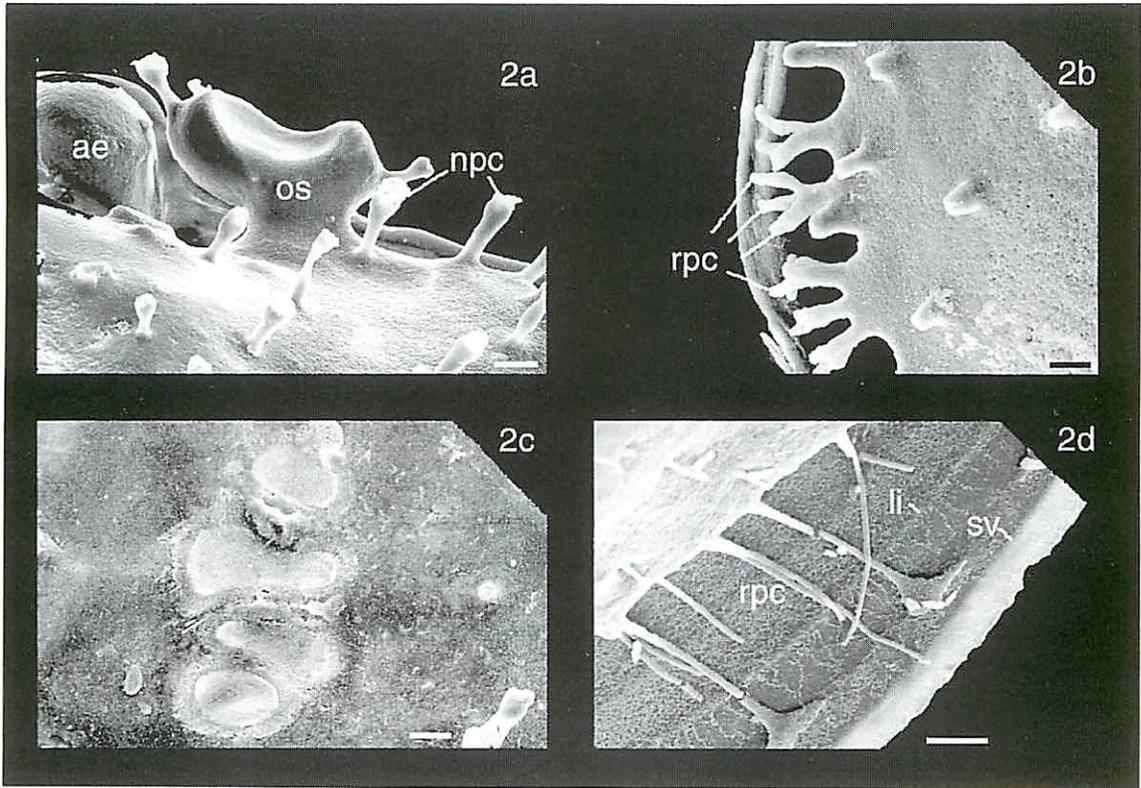


Fig. 2. Internal molds of several specimens. 2a. Anterodorsal part of the female right valve of *Aurila imotoi* Ishizaki. Anterior element of hinge (ae), ocular sinus (os) and normal pore canals (npc) are visible. 2b. Anteroventral part of the female left valve of *Xestoleberis hanaii* Ishizaki. Radial pore canals (rpc) branch off. 2c. Adductor muscle scars of the female left valve of *Bicornucythere bisanensis* (Okubo). 2d. Posteroventral part of the female left valve of *Bythoceratina hanaii* Ishizaki. Radial pore canals (rpc), list (li) and selvage (sv) are visible. Scale bar = 0.01mm

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