

Note

Use of crustacean burrows as habitat by the marine snail *Circulus cinguliferus* (Gastropoda: Truncatelloidea: Vitrinellidae)RYUTARO GOTO^{1,*} & TAIGI SATO²¹Seto Marine Biological Laboratory, Field Science Education and Research Center, Kyoto University, 459 Shirahama, Nishimuro, Wakayama 649–2211, Japan²Faculty of Science, University of the Ryukyus, 1 Sembaru, Nishihara, Nakagami, Okinawa 903–0213, Japan

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Abstract: The family Vitrinellidae is a group of tiny marine snails that generally occur in shallow waters of temperate and tropical seas. The biology of most vitrinellid species remains poorly understood. In this study, we report that *Circulus cinguliferus* (A. Adams, 1850) (Vitrinellidae), distributed widely in the warm shallow waters of the Pacific, inhabit crustacean burrows, including those of the mud shrimp *Neaxius acanthus* (Strahlaxiidae) and snapping shrimp *Alpheus rapax* (Alpheidae), in the intertidal and subtidal flats of the Okinawa Islands, southern Japan. They exhibited highly clumped distribution among the host burrows, suggesting that they are attracted by conspecifics. Although the biology of most *Circulus* species remains unknown, *Circulus texanus* (D. R. Moore, 1965) is known to inhabit stomatopod burrows in the western Atlantic. Our findings suggest that such a commensal habit may be more widespread in this genus than previously thought.

Key words: *Alpheus*, symbiosis, commensalism, sexual size dimorphism, yabby pump

The family Vitrinellidae Bush, 1897 (Gastropoda: Truncatelloidea) is a group of tiny marine snails commonly occurring in shallow waters of temperate and tropical seas (Ponder 1994; Bieler & Mikkelsen 1988; Rubio et al. 2011; Hasegawa 2017; Rubio & Rolán 2018). This family was previously considered a synonymy of Tornidae (Bouchet & Rocroi 2005; Rubio et al. 2011; Hasegawa 2017), but was separated from the latter by Bouchet et al. (2017) based on recent molecular phylogenetic analyses by Takano and Kano (2014) (but see Rubio & Rolán 2018; Ponder et al. 2020). Some vitrinellids occur as free-living species on seagrasses and hard substrates (wood and stones) in estuaries, others live underneath deeply embedded rocks, whereas a few occur as commensals associated with burrowing invertebrates (Ponder & de Keyser 1998). However, the microhabitat of most vitrinellid species remains unknown, mainly due to the difficulty in obtaining living specimens (Sasaki 2008).

Circulus cinguliferus (A. Adams, 1850) is a vitrinellid species inhabiting the sandy gravel bottom in the subtidal zone (Hasegawa 2017). This species was originally described as *Cyclostrema cingulifera* based on a specimen collected from

the subtidal zone (ca. 7.3 m depth) in Dumaguete, Philippines, by A. Adams (1850). In Japan, this species is distributed from the Boso Peninsula to the tropical Pacific (Hasegawa 2017); however, the biology of living specimens remains poorly understood. In this study, we report that living *C. cinguliferus* snails inhabit crustacean burrows in the intertidal and subtidal flats of the Okinawa Islands, southern Japan.

We sampled burrowing crustaceans and their burrow symbionts around Okinawa Island (Fig. 1A) by using a yabby pump, during February–June 2020. The contents suctioned from the burrows were sieved through a 1-mm mesh. Consequently, we collected (1) three individuals of *C. cinguliferus* from a burrow of the mud shrimp *Neaxius acanthus* (A. Milne-Edwards, 1879) (Strahlaxiidae) [or perhaps *Alpheus rapax* Fabricius, 1798 (Alpheidae)] in the intertidal gravelly sand bottom with patches of seagrass *Thalassia hemprichii* (Ehrenberg) Ascherson, 1871, in Urasoe, western Okinawa Island (Fig. 1B), on February 7, 2020, (2) more than 10 individuals from a burrow of an unknown host in the subtidal mud bottom (ca. 8 m depth) of Kin Bay, eastern Okinawa Island (Fig. 1B), on February 9, 2020, (3) five individuals along with the host snapping shrimp *A. rapax* from a burrow in the upper subtidal sandy mud bottom (less than 1 m depth during the lowest tide) with seagrass beds, in Hamahiga Island, located

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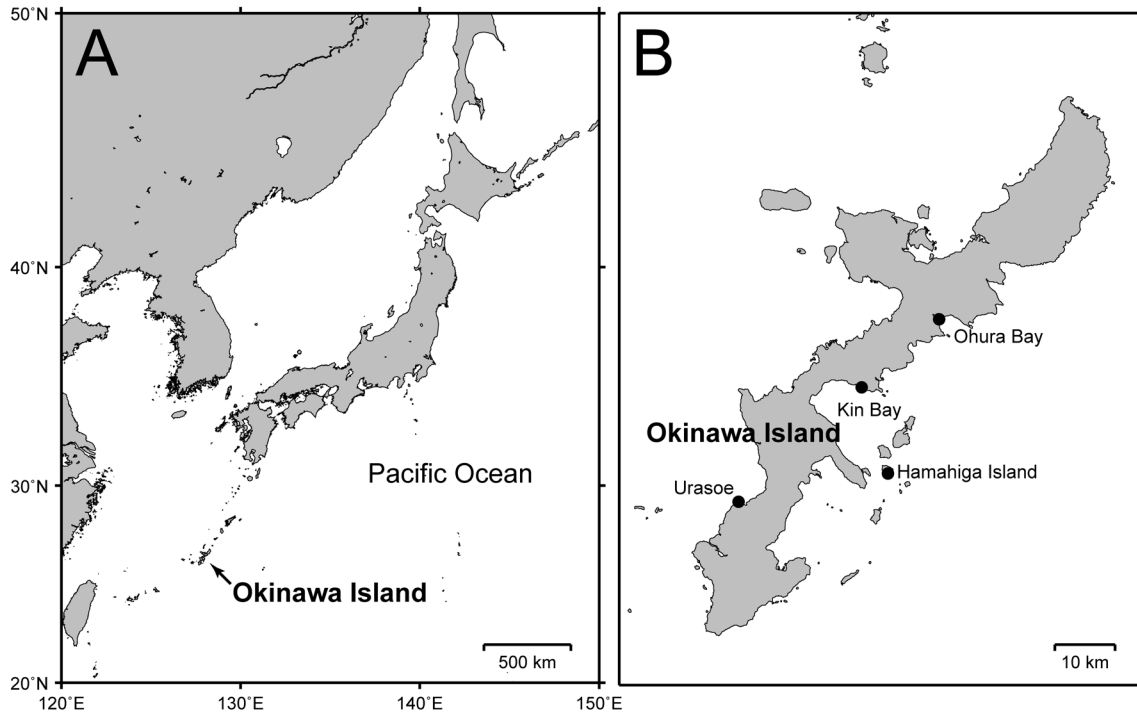


Fig. 1. Sampling sites of *Circulus cinguliferus*. (A) Geographic location of Okinawa Island. (B) Okinawa Island.

to the east of Okinawa Island (Fig. 1B), on March 10, 2020, and (4) three individuals from a burrow, likely of *Alpheus* sp., in the subtidal soft mud bottom (ca. 6–7 m depth) of Ohura Bay, eastern Okinawa Island (Fig. 1B), on June 13, 2020. The host was collected with the gastropods only at Hamahiga Island, and was released after identification. The exact number of crustacean burrows (mostly those of *N. acanthus* and *A. rapax*) surveyed by the yabby pump was not recorded; however, roughly 20% of the burrows contained *C. cinguliferus* individuals.

All the specimens of *C. cinguliferus* were brought back to the laboratory, except for those collected in Kin Bay, which were released on site after identification. The soft body of eight specimens, including one from Urasoe, four from Hamahiga, and three from Ohura, were withdrawn from the shells for observation after being boiled for 30 s, and then fixed with 10% formalin solution or 99.5% ethanol. We measured the shell diameter and height of all the collected specimens using digital calipers, which were as follows: 5.4×3.0, 5.6×3.0, and 6.2×3.0 mm (Urasoe); 4.9×2.9, 5.1×3.4, 5.3×3.4, 5.5×3.3, and 5.9×3.4 mm (Hamahiga); and 4.3×2.4, 4.5×2.5, and 4.6×2.8 mm (Ohura). These specimens were then deposited in the collections of the Seto Marine Biological Laboratory, Kyoto University.

The living specimens collected from Urasoe, Hamahiga, and Ohura were observed in petri dishes filled with seawater. The head-foot bore a pair of long, blackish cephalic tentacles with immobile terminal bristles and with eyes in a small swellings at the bases (Fig. 2). A snout protruded between the cephalic tentacles (Fig. 2). As reported in other vitrinellids (e.g., Fretter 1956; Bieler & Mikkelsen 1988; Ponder 1994),

two pallial tentacles projected from the right side of the mantle edge through the posterior corner of the shell aperture (Fig. 2A): the upper tentacle was whitish with terminal bristles, whereas the lower tentacle was blackish without bristles (Fig. 2A). The foot was elongated and flattened (Fig. 2A). The anterior end of the foot projected laterally (Fig. 2A), whereas the posterior end was slightly indented at the middle and lacked a metapodial tentacle (Fig. 2A). A circular, multispiral operculum was present on the posteroventral side of the foot (Fig. 2B). The external anatomy of the visceral mass (Fig. 2C, D) was similar to that reported for other vitrinellids in previous studies (e.g., Bieler & Mikkelsen 1988).

Of the eight individuals whose soft body was examined, only the two smallest specimens collected from Ohura (maximum shell diameter: 4.3 and 4.5 mm) possessed a penis, which was large and located posteriorly to the cephalic tentacles and covered by the mantle (Fig. 3). It originated from the slightly right of the dorsal midline and was directed posteriorly (Fig. 3). The testes were orangish in color (Fig. 3A). Larger specimens from Ohura (4.6 mm), Urasoe (6.2 mm), and Hamahiga (4.9, 5.3, 5.9, and 5.5 mm) did not possess a penis, suggesting that females may be larger than males in *C. cinguliferus*. In contrast, males and females appear to be similar in size in *Circulus striatus* (Philippi, 1836) and *Circulus mortonii* Ponder, 1994 (Fretter 1956; Ponder 1994).

Our field collections suggest that *C. cinguliferus* inhabits crustacean burrows, although further ecological studies are required to confirm whether it is an obligate or facultative burrow commensal. Multiple individuals of *C. cinguliferus* were recorded in the same burrow, despite their relatively low occurrence across the burrows (roughly 20%). It is pos-

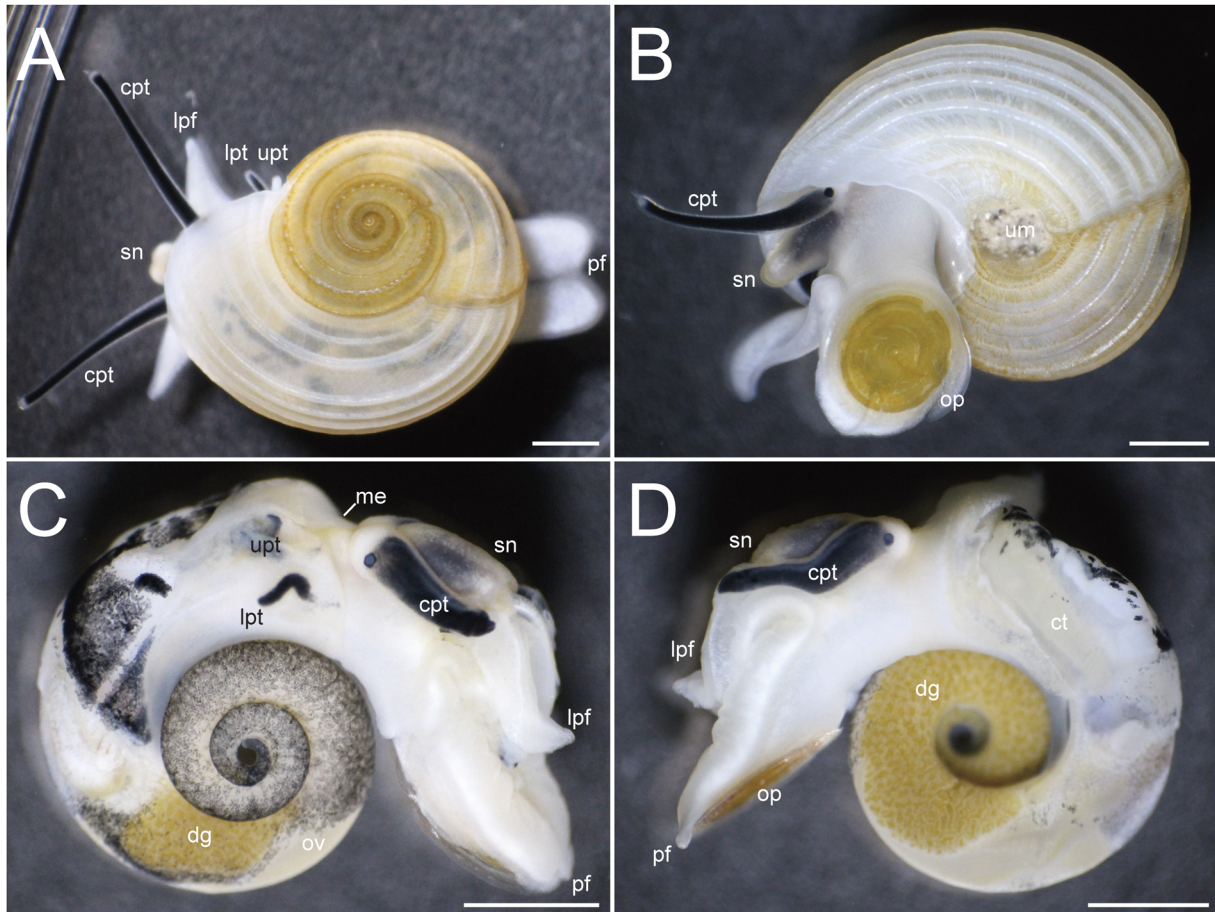


Fig. 2. Female *Circulus cinguliferus* (maximum shell diameter: 5.5 mm) collected from a burrow of *Alpheus rapax* at Hamahiga Island, Okinawa, Japan. (A) A crawling individual in dorsal view. (B) A living individual in ventral view. (C) Right side of the soft body withdrawn from the shell and not fixed. (D) Left side of the soft body. Abbreviations: cpt, cephalic tentacle; ct, ctenidia; dg, digestive gland; lpf, lateral projection of the anterior end of foot; lpt, lower pallial tentacle; me, mantle edge; op, operculum; ov, ovary; pf, posterior end of foot; sn, snout; um, umbilicus; upt, upper pallial tentacle. Scale bar=1 mm.

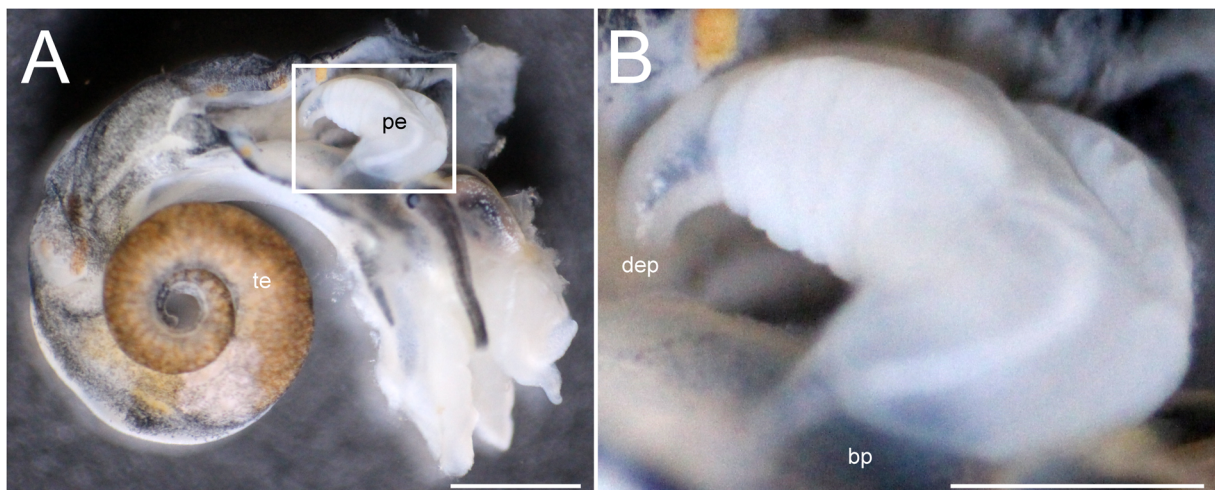


Fig. 3. Male *Circulus cinguliferus* (maximum shell diameter: 4.5 mm) collected from a burrow, likely of *Alpheus* sp., in Ohura Bay, eastern Okinawa Island, Japan. (A) Right side of the soft body withdrawn from the shell and not fixed. The mantle was partially dissected to show the penis, which is indicated by an open rectangle. (B) Close up of the penis. Abbreviations: bp, base of penis; dep, distal end of penis; pe, penis; te, testis. Scale bar=1 mm (A) and 0.5 mm (B).

sible that they were attracted by conspecifics for the purpose of mating. Barnes and Laurie (2018) investigated the spatial patterns of the entire macrobenthic assemblage in the intertidal seagrass beds in subtropical Moreton Bay, Queensland, Australia, by extensive core sampling, and noted that *C. cinguliferus* were extremely patchily distributed in terms of abundance. Although it is necessary to confirm that *C. cinguliferus* from Queensland are truly conspecific to those from Okinawa, it is possible that each patch of *C. cinguliferus* observed by Barnes and Laurie (2018) corresponded to a cluster of individuals inhabiting the same crustacean burrow.

The genus *Circulus* includes approximately 30 species (WoRMS 2020). Although the biology of most species remains unknown, at least *C. texanus* (Moore, 1965) is known to inhabit burrows of the stomatopod crustacean *Lysiosquilla scabricauda* (Lamarck, 1818) in Florida (Bieler & Mikkelsen 1988). In addition, *C. mortoni* is possibly associated with the burrowing holothurian *Protankyra bidentata* in Hong Kong (Woodward & Barrett, 1858) (Ponder, 1994). Taking our findings into consideration, it is probable that numerous species of *Circulus* occur as commensals of burrowing invertebrates. In addition to *Circulus*, some truncatelloideans (mostly tornids) are known to form commensalistic associations with other invertebrates; for example, *Cochliolepis parasitica* Stimpson, 1858 is an ectocommusal of acoetid annelids (Moore 1972); *Cyclostremiscus beauii* (P. Fischer, 1857) inhabits stomatopod burrows (Bieler & Mikkelsen 1988); *Sigaretornus planus* (A. Adams, 1850) inhabits echinuran burrows (Morton 1988; Ponder 1994); *Sigaretornus* sp. inhabits stomatopod burrows (Yamashita et al. 2011; Fukuda 2020); *Teinostoma* sp. inhabits ghost-shrimp burrows (Goto et al. 2014); and *Vitrinella oldroydi* Bartsch, 1907 is an ectocommusal of chitons (Eernisse et al. 2007). Molecular phylogenetic studies are required to understand their evolutionary relationships and the origin(s) of the commensalistic lifestyles in these groups.

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