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Observation of intersexuality in land hermit crabs (Anomura: Coenobitidae)

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We report here the first observation of intersexuality in a land hermit crab (Coenobitidae). Three species of land hermit crabs from Okinawa were investigated. In the population of the terrestrial hermit crab *Coenobita rugosus* two per cent of males were found to be intersex. Besides the true male gonopores on the coxae of the 5th pair of pereopods, they had additional openings on the coxae of the 3rd pair of pereopods. Both specimens examined had a normally developed male reproductive system. In the populations of two other species studied, *C. brevimanus* and *C. purpureus*, however, no intersex individuals have been found. Examples of intersexuality in decapod crustaceans are discussed and putative explanations of this phenomenon in land hermit crabs are proposed.

INTRODUCTION

Hermit crabs (Anomura) occur in various habitats of the world. A major step in the evolution of this group was adaptation to a fully-terrestrial life (Greenaway, 2003; Williams & McDermott, 2004). During this successful transition from marine to terrestrial ecosystems hermit crabs have developed many physiological and behavioural adaptations, including very complicated and still poorly-known reproductive behaviour (Helfman, 1977; Nakasone, 2001).

One of the most interesting features of reproductive morphology of hermit crabs is the phenomenon of intersexuality. Hermit crabs, like the other decapod crustaceans, are characterized by the sexual dimorphism. Males and females are clearly distinguishable by the position of their sexual openings (gonopores). The gonopores open typically on the coxae of the 3rd pair of pereopods (walking legs) in females, and on the coxae of the 5th pair of pereopods in males. However, some individuals have been found to possess gonopores in both 3rd and 5th pairs of pereopods (Turra, 2004, 2005). Such individuals are called intersex. Intersexuality has been previously described in other decapod crustaceans, but to-date there have been few reports in hermit crabs (McLaughlin & Lemaitre, 1993; Turra & Leite, 2000). Previously intersexuality has been observed only in aquatic species of Anomura, mostly members of families Paguridae and Diogenidae, but not in land hermit crabs (Coenobitidae). The present study aimed to screen the presence of specimens possessing the intersexual feature in the populations of the three species of Coenobitidae from the Iriomote Island (Okinawa, Japan). We report here the first observation of intersex individuals of terrestrial hermit crab *Coenobita rugosus* (Milne-Edwards, 1837). This species occurs from the mainland coast of East Africa through the Indo-Pacific to Tahiti and Tuamotu Islands. Like all members of this group, this species exhibits high levels of terrestrialization and can be found up to 300 m from the seashore (Burggren & McMahon, 1988).

MATERIALS AND METHODS

Males of examined species, including 98 specimens of *Coenobita brevimanus* (Dana, 1852), 95 specimens of *C. purpureus* (Stimpson, 1858) and 100 specimens of *C. rugosus* (Milne-Edwards, 1837) were obtained from the populations of these land hermit crabs randomly collected in Iriomote-jima Island, Okinawa, Japan (124°E 24°N), near the sandy seashore at night during the period from May to June 2004 (breeding season).

The animals were carefully examined using a Zeiss 2000-C binocular microscope. The thoracic sternites and coxae of pereopods with gonopores of specimens of interest were photographed using a digital camera and sketched. The shield (carapace) length of intersex specimens was measured to within 0.1 mm using digital calipers. The reproductive system of intersex individuals was examined. Terminology concerning external morphology is as in McLaughlin (2003); and terms concerning internal morphology of reproductive system—are as used in Hess & Bauer (2002) and Kronenberger et al. (2004).

RESULTS

No intersex individuals have been found among the *Coenobita brevimanus* and *C. purpureus* specimens. Two out of 100 male *C. rugosus* proved to be intersex individuals. Both are relatively large, mature males. It should be noted that growth dynamics and longevity of *Coenobita* remain unclear. It was shown that specimens with a total weight of 1 g and

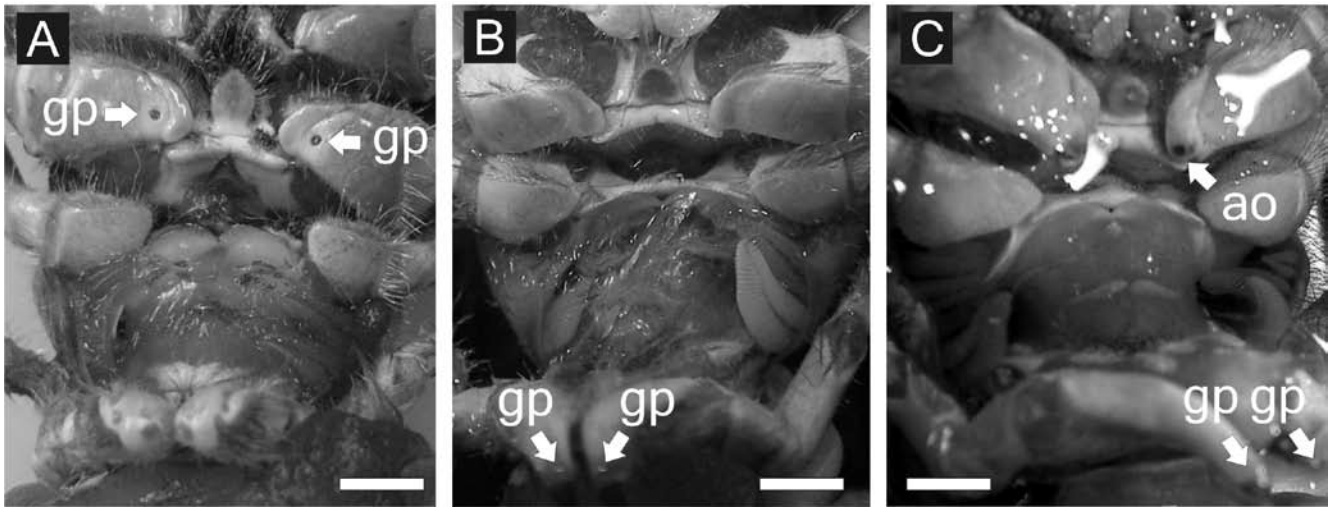

Figure 1. Ventral view of the coxae of pereopods of (A) normal female; (B) normal male; and (C) intersex individual of *Coenobita rugosus*. gp, gonopores; ao, additional opening. Intersex specimen possesses an additional gonopore-like opening on the 3rd pair of pereopods. Arrows indicate location of additional openings and gonopores. Scale bar: 3 mm.

a shield larger than 4.5 mm are already sexually mature, but the general age/weight/size equation is yet to be created, due to the strong influence shell and food availability, temperature and other environmental factors which have higher fluctuations (compared with the marine environment) on land (Nakasone, 2001). In our collections we focused on the mature crabs with the shield size higher that 8 mm. The whole pool of obtained *C. rugosus* could be roughly divided into three groups by the shield size: 10–20 mm (49 specimens), 22–29 mm (37 specimens) and larger than 30 mm (14 specimens). Both intersex individuals belonged to the second group. So far no statistically significant differences

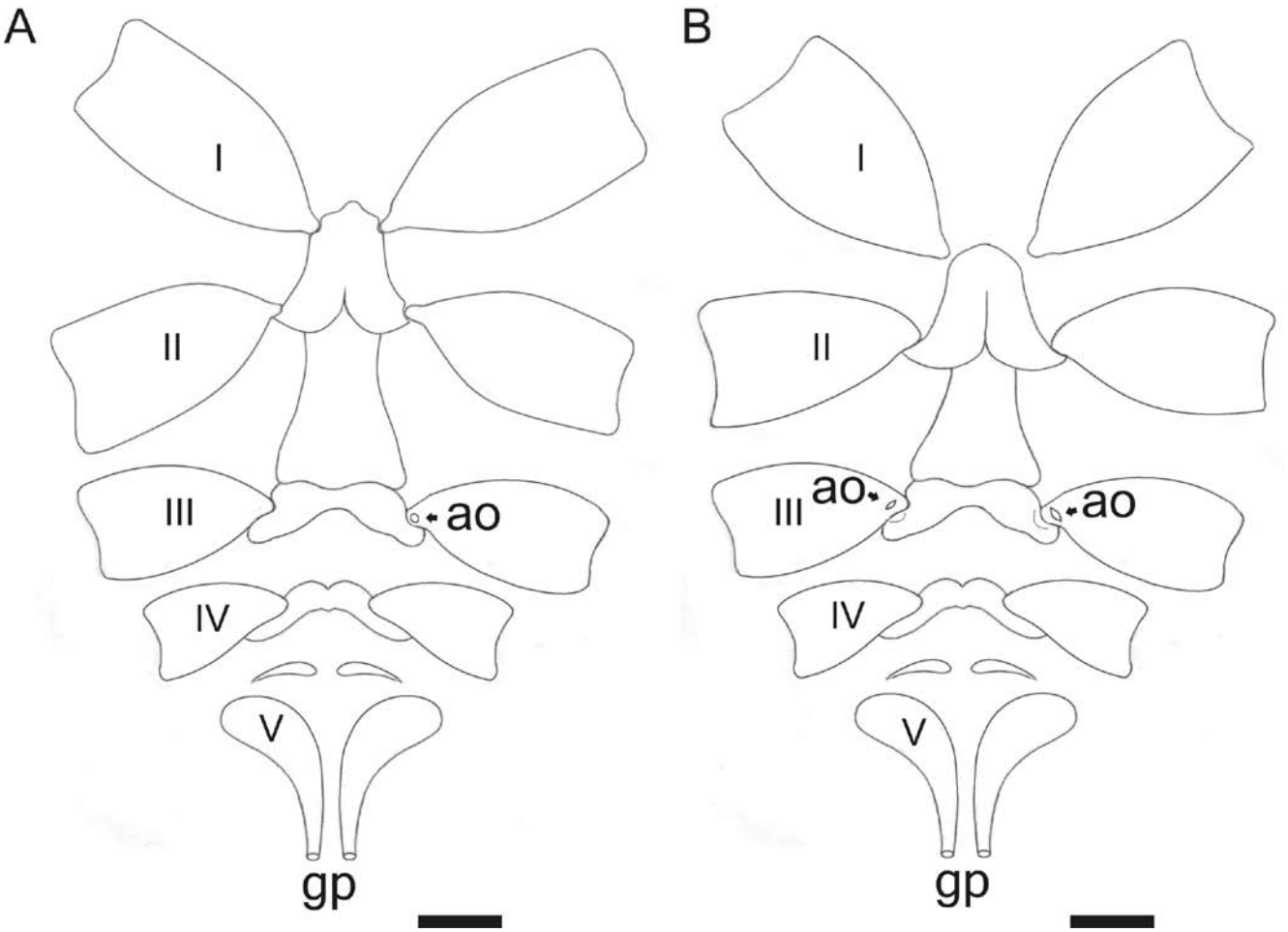

Figure 2. Location of the additional openings on the coxae of pereopods of two intersex individuals. (A) Shield length 27.8 mm; and (B) shield length 25.7 mm of *Coenobita rugosus*. Pereopods are numbered by Roman numerals (I–V). gp, gonopores; ao, additional openings. Scale bar: 1.5 mm.

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were observed between shield length in females and males of the same size group obtained by the random collections and the M:F sex ratio in the population of *C. rugosus* tended to be 1:1.2–1.3 and favoured females in all size groups (Nakasone 2001; our unpublished observations).

The first intersex individual, with shield length of 27.8 mm, had both male gonopores on the coxae of the 5th pereopod and only a single opening on the coxa of the left 3rd pereopod. It was approximately the same size as the gonopores of typical females, but was located closer to the proximal edge of the coxa (Figures 1 & 2).

The second individual, with shield length 25.7 mm, also had both male gonopores on the coxae of 5th pereopod and paired openings on both coxae of the 3rd pereopods. Two openings were different in their size and shape (Figure 2). They were also both located closer to the proximal edge of the coxa than typical female gonopores (Figure 1). In addition, this individual had two small cavities on the posterior portion of the 6th sternite of the thorax, located close to the openings on the coxae of the 3rd pereopods (Figure 2).

In both cases normal male gonopores on the coxae of the 5th pereopods were gaping. Previously, several authors (Turra & Leite, 2000; Turra, 2004, 2005) observed the mating behaviour of intersex individuals of aquatic hermit crabs and confirmed that they successfully copulated, as males, with females. In the present study, primarily dissection of the intersex individuals revealed the presence of the male reproductive system. At the same time, there is some documented evidence of the presence of small oviducts and other morphological features of the female reproductive system in the intersex individuals of crayfish believed to be functioning as males (Almeida & Buckup, 2000). Taking this into account, fine morphological and histological examination of the internal reproductive systemof two intersex individuals of *C. rugosus* are being conducted with special attention to the structure of testis and seminal ducts.

DISCUSSION

In the studied population of *Coenobita rugosus* 2% (2 out of 100 collected specimens) of males were found to be intersex. Our results are similar to those reported by Turra (2004), where the percentage of intersex individuals in the populations of three species of aquatic hermit crabs (*Clibanarius antillensis*, *C. sclopetarius* and *C. vittatus*) was 2–7%.

In the present study the intersex individuals were not observed in populations of *C. brevimanus* and *C. purpureus*. The putative absence of intersex individuals in these populations can hardly be explained by our present knowledge upon differences in ecology and reproductive strategies of studied species (Burggren & McMahon, 1988). It is more likely that such an apparent difference in the studied populations may have been due to the lower frequency of intersex individuals or possibly the seasonal variation of the reproductive period. At the same time, we can not declare a total absence of intersex individuals in *C. brevimanus* and *C. purpureus*, due to the relatively small sampling at the present time.

The function of intersex individuals is still poorly-known. It is still debatable as to whether intersexuality in decapods can be a part of a true hermaphroditic condition, or only an aberrant event. To date this phenomenon has been described in different groups of decapod crustaceans, such as Caridea, Thalassinidea, Astacidea and Brachyura. The caridean shrimp *Lysmata wurdemanni* shows protandric hermaphroditism and develops first as male-phase individuals and later changes to female-phase (Bauer & Holt, 1998). However, the female-phase individuals retain functional male gonopores and ejaculatory ducts, and the testicular portions of gonads (ovotestes) continue to produce sperm (Baldwin & Bauer, 2003). Both males and females of the freshwater astacidean crayfish *Parastacus brasiliensis* have intersex genital ducts connected to a gonad with male or female components depending on the sex of the individual. A third sexual form has recently been described—an intermediate between male and female, with a few morphotypes (Almeida & Buckup, 2000). Such individuals have two sets of gonopores, but the female gonopores are closed, and they function as males. The intersex individuals of the thalassinidean mud shrimp *Upogebia snelliusi* with both pairs of gonopores also function as males (Sakai et al., 2004; Hirano et al., 2006). There are only a few described examples of intersexuality patterns in Brachyura (Zou & Fingerman, 2000; C. McLay, personal communication). The best known is gynandromorphism—the presence of uniramous male pleopods on the first abdominal segment as well as the normal biramous pleopods on segments 2–5 in the dynomenid crab *Metadynomene tanensis*. Summarizing all these data, we see that representatives of different groups show both functional (Caridea, Astacidea – in part) and non-functional (Astacidea – in part, Thalassinidea) hermaphroditism. All these examples are protandric hermaphrodites. Protogyny has not been reported for decapod crustaceans yet, although some recent genetic investigations suggest that intersex males of crayfish *Cherax quadricarinatus* have developed from females (Parnes et al., 2003). In addition, the possibility of partial hormonal control of sex determination in crayfish *Procambarus clarkii* was confirmed by implantation of male androgenic glands into immature females (Taketomi et al., 1996; Taketomi & Nishikawa, 1996). Taking these examples into consideration, male individuals of *Coenobita* with accessory female openings may be just isolated instances of specimens with abnormalities in the functioning of their endocrine system. However, there is still no explanation of such distinction in frequency of the intersex individuals in populations of different groups of decapod crustaceans—from very low (2–7%) in the case of hermit crabs (Turra & Leite, 2000), to relatively high (more than 90%) in the case of the thalassinidean *Upogebia snelliusi* (Hirano et al., 2006). Moreover, in the case of *Upogebia snelliusi* it was proposed that the appearance of two pairs of gonopore openings on the 3rd and 5th periopods is a normal step of sexual maturation of the male, while the immature males possess 'classical type' of a single pair of openings on the 5th pereopods (Sakai et al., 2004; Hirano et al., 2006).

According to several authors (Turra & Leite, 2000; Turra, 2004), the closure of female gonopores in intersex individuals of hermit crabs after moulting might suggest that they could have previously been females. But, to our knowledge, intersex individuals with both pairs of gonopores, functioning as females, have not yet been found and described among

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Anomura. In the case of observing such females, it would be possible to foretell stages of transition from normal females through intersex females and intersex males to normal males (or, in the case of protandry, reverse direction from normal males to normal females). To our knowledge, such data have yet to be presented, although during the collection of specimens for the present study, we also observed a female of coconut crab *Birgus latro* with abnormal 5th pair of pereopods with an unusually soft thin-chitinized area, where male gonopores are usually located (unpublished observations).

As a result, the phenomenon of intersexuality in Anomura seems to occur at all grades of terrestrialization from typically aquatic species, like Diogenidae and Paguridae, to fully-terrestrial species, like Coenobitidae. All coenobitid hermit crabs depend on water only during the breeding season. Since the natural habitat of land hermit crabs is usually located relatively far from the seashore, the reproductive behaviour of both males and females has to be strictly synchronized with the timing of high tide (Nakasone, 2001). Considering that, male-to-female sex reversal patterns should be an ecologically 'costly' process, since the occurrence of intersex individuals should be followed by changing the sexual behaviour which is much more complicated in terrestrial species than in marine ones. Nevertheless the present study confirms the presence of intersex specimens even among the fully-terrestrial species of decapod crustaceans. Further detailed studies, including wide population monitoring and fine internal morphological investigation of intersex individuals are necessary to definitively understand sex reverse phenomena in this crustacean group.

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REFERENCES

Almeida, A.O. & Buckup, L., 2000. Occurrence of protandric hermaphroditism in a population of the neotropical freshwater crayfish *Parastacus brasiliensis* (Parastacidae). *Journal of Crustacean Biology*, **20**, 224–230.

Baldwin, A.P. & Bauer R.T., 2003. Growth, survivorship, life-span, and sex change in the hermaphroditic shrimp *Lysmata wurdemanni* (Decapoda: Caridea: Hippolytidae). *Marine Biology*, **143**, 157–166.

Bauer, R.T. & Holt, G.J., 1998. Simultaneous hermaphroditism in the marine shrimp *Lysmata wurdemanni* (Caridea: Hippolytidae): an undescribed sexual system in the decapod Crustacea. *Marine Biology*, **132**, 223–235.

Burggren W.W. & McMahon, B.R., 1988. *Biology of the land crabs*. Cambridge: Cambridge University Press.

Greenaway, P., 2003. Terrestrial adaptations in the Anomura (Crustacea: Decapoda). *Memoirs of Museum Victoria*, **60**, 13–26.

Helfman, G.S., 1977. Copulatory behavior of the coconut or robber crab *Birgus latro* (L.) (Decapoda, Anomura, Paguridea, Coenobitidae). *Crustaceana*, **33**, 198–202.

Hess, G.S. & Bauer R.T., 2002. Spermatophore transfer in the hermit crab *Clibanarius vittatus* (Crustacea, Anomura, Diogenidae). *Journal of Morphology*, **253**, 166–175.

Hirano, Y., Ikeda, H., Nanri, T., Takada, Y., Sakai, K. & Saigusa, M., 2006. Morphology and ecology of *Upogebia snelliusi* Ngoc-Ho, 1989 (Crustacea; Decapoda; Thalassinidea) inhabiting sandstone at intertidal zone in Iriomote Island. *Aquabiology*, **28**, 95–103. [In Japanese.]

Kronenberger, K., Brandis, D., Turkay, M. & Storch, V., 2004. Functional morphology of the reproductive system of *Galathea intermedia* (Decapoda: Anomura). *Journal of Morphology*, **262**, 500–516.

McLaughlin, P.A., 2003. Illustrated keys to families and genera of the superfamily Paguroidea (Crustacea: Decapoda: Anomura), with diagnoses of genera of Paguridae. *Memoirs of Museum Victoria*, **60**, 111–144.

McLaughlin, P.A. & Lemaitre, R., 1993. A review of the hermit crab genus *Paguritta* (Decapoda: Anomura: Paguridae) with descriptions of three new species. *Raffles Bulletin of Zoology*, **41**, 1–29.

Nakasone, Y., 2001. Reproductive biology of three land crabs (Decapoda: Anomura: Coenobitidae) in Okinawa, Japan. *Pacific Science*, **55**, 157–169.

Parnes, S., Khalaila, I., Hulata, G. & Sagi, A., 2003. Sex determination in crayfish: are intersex *Cherax quadricarinatus* (Decapoda, Parastacidae) genetically females? *Genetic Research*, **82**, 107–116.

Sakai, K., Hirano, Y. & Saigusa, M., 2004. A new record of *Upogebia snelliusi* Ngoc-Ho, 1989 (Thalassinoidea, Upogebiidae) from Iriomote Island, Ryukyus, Japan. *Crustaceana*, **77**, 741–749.

Taketomi, Y. & Nishikawa, S., 1996. Implantation of androgenic glands into immature female crayfish, *Procambarus clarkii*, with masculinization of sexual characteristics. *Journal of Crustacean Biology*, **16**, 232–239.

Taketomi, Y., Nishikawa, S. & Koga, S., 1996. Testis and androgenic gland during development of external sexual characteristics of the crayfish *Procambarus clarkii*. *Journal of Crustacean Biology*, **16**, 24–34.

Turra, A., 2004. Intersexuality in hermit crabs: reproductive role and fate of gonopores in intersex individuals. *Journal of the Marine Biological Association of the United Kingdom*, **84**, 757–759.

Turra, A., 2005. Reproductive behavior of intertidal hermit crabs (Decapoda, Anomura) in southeastern Brazil. *Revista Brasileira de Zoologia*, **22**, 313–319.

Turra, A. & Leite, F.P.P., 2000. Population biology and growth of three sympatric species of intertidal hermit crabs in south-eastern Brazil. *Journal of the Marine Biological Association of the United Kingdom*, **80**, 1061–1069.

Williams, J.D. & McDermott, J.J., 2004. Hermit crab biocoenoses: a worldwide review of the diversity and natural history of hermit crab associates. *Journal of Experimental Marine Biology and Ecology*, **305**, 1–128.

Zou, E. & Fingerman, M., 2000. External features of an intersex fiddler crab, *Uca pugilator* (Bosc, 1802) (Decapoda, Brachyura). *Crustaceana*, **73**, 417–423.

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