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The ghost crab *Ocypode mortoni* George, 1982 (Crustacea: Decapoda: Ocypodidae): redescription, distribution at its type locality, and the phylogeny of East Asian *Ocypode* species

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Abstract

The ghost crab *Ocypode mortoni* George, 1982 was first described from the sandy shores of Sai Wan, Hong Kong, and although also identified from South China and Japan, has not been subsequently recorded in Hong Kong. The taxonomic status of *O. mortoni* is not clear as its original morphological description was not detailed, and there remains no information on its vertical and horizontal ecological distribution in Hong Kong. In the present study, *O. mortoni* was found to be locally rare in Hong Kong, only present at three of seven surveyed sites. It was always sympatric with the common *O. ceratophthalmus* (Pallas, 1772) at the same tidal level, and there appeared to be no clear habitat/niche distinction. Relative abundance of *O. mortoni* vs. *O. ceratophthalmus* was at most 31%: 69% based on quantitative transect survey data from Sai Wan. Molecular phylogenetic analyses on 24 haplotypes (from 28 individuals) of 636-basepair (bp) mitochondrial cytochrome oxidase I (COI) gene of *Ocypode* spp. in East Asia show *O. mortoni* is a valid species most closely related to *O. stimpsoni* Ortmann, 1897, with an average difference of 37.64 bp, and nucleotide divergence of 6.25%. The two species are very similar morphologically, but adults of *O. mortoni* develop ocular stylets.

Key words: ghost crab, *Ocypode mortoni*, redescription, East Asia, Hong Kong

Introduction

Ocypode species, more commonly known as “ghost crabs”, are found along sandy shores within the tropics and subtropics (Dahl 1953; Barass 1963). They are typically predators (Hughes 1966; Smith 1975), scavengers (Wolcott 1978), and/or deposit feeders (Tweedie 1950; Robertson & William 1981) and play an important role in the ecology of sandy beaches.

At present, there are at least 26 recognized *Ocypode* species (reviewed by Ng *et al.* 2008). In East Asia (including China, Taiwan, Japan and Korea) at least 5 species of *Ocypode* have been reported, viz. *O. ceratophthalmus* (Pallas, 1772), *O. cordimanus* Latreille, 1818, *O. mortoni* George, 1982, *O. sinensis* Dai, Song & Yang, 1985, and *O. stimpsoni* Ortmann, 1897. *Ocypode ceratophthalmus*, *O. cordimanus* and *O. sinensis* are widespread species in the Indo-Pacific region; while *O. stimpsoni* has a narrower distribution, being recorded from Japan, Korea, China and Taiwan (see Tweedie 1937; Barnard 1950; Crosnier 1965; George & Knott 1965; T. Sakai 1976; Dai & Yang 1991; Huang *et al.* 1998). *Ocypode mortoni* was first described from Hong Kong (George 1982) and has been reported from Guangdong, Guangxi and Hainan in China (Dai *et al.* 1985 [as *O. macrocera* H. Milne Edwards, 1837]; Dai & Yang 1991) and the Pacific coast of Shikoku, Japan (K. Sakai 2000).

Ocypode mortoni can be diagnosed as “with a short ocular stylet in the adult condition, stridulating ridge on the major manus composed of up to 54 closely spaced striae” (George 1982). George (1982) further mentioned that *O. mortoni* morphologically resembles *O. nobilii* De Man, 1902 from Borneo, and *O. fabricii* H. Milne Edwards, 1837 from northern Australia, yet differs in the number of striae on the stridulating ridge. *O. stimpsoni* has also been believed to be morphologically closely related, except for the presence of the ocular stylets in *O. mortoni* (Dai *et al.*

1985; Dai & Yang 1991). Concerning the distribution pattern, George (1982) believed that *O. mortoni* was associated with freshwater discharge along sandy beaches, in contrast to the solely “marine” *O. ceratophthalmus* (but see below), that it showed deposit feeding behavior, and that it has a distinctive burrow opening surrounded by radiating “feeding lines” (see Plate 4 in George 1982, Fig. 8e of present paper).

Ocypode mortoni has not been reported from Hong Kong since its discovery, and there have been no further studies concerning its ecology or factors that might govern its distribution. Consequently, the present study aimed to: (1) provide a detailed morphological description of *O. mortoni*; (2) obtain a better ecological understanding of the species, including the appearance of burrows, vertical and horizontal distribution on the shore, and the extent of niche partitioning with the apparently sympatric *O. ceratophthalmus*; and (3) investigate molecular phylogenetic relationships with congeneric species. To achieve these aims we re-visited the type locality and conducted quantitative transect sampling to examine its abundance and distribution patterns. We further undertook molecular analyses on the segment of mitochondrial cytochrome oxidase subunit I (COI) for *O. mortoni* and other species from East Asia, to further confirm its taxonomic identify and try to better establish its phylogenetic position.

Material and methods

Horizontal distribution of *O. mortoni* in Hong Kong. Seven sandy shores were visited in Hong Kong in August 2008, including Big Wave Bay on Hong Kong Island, Starfish Bay near Ma On Shan, Pak Lap and Sai Wan in Sai Kung, and Bui O (as Pui O in George 1982), Cheung Sha and Fan Lau on Lantau Island (see Fig. 1) At each site, a 30-meter stretch of shoreline was chosen at the tidal level where the highest abundance of *Ocypode* burrows. All *Ocypode* burrow along the 30 meter-stretch of shoreline were dug and the crabs were identified. Representative specimens were collected and the rest were released upon identification.

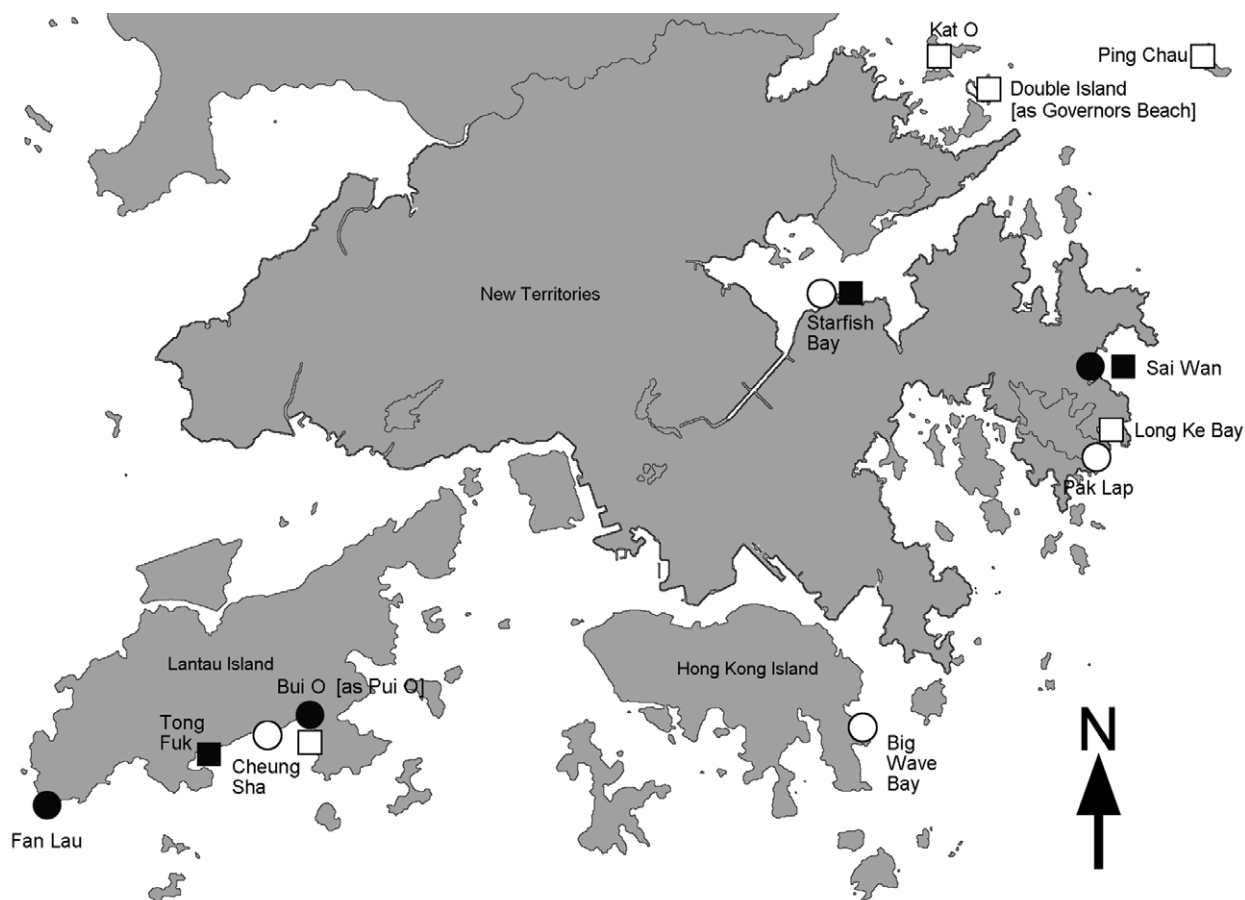


FIGURE 1. Map of Hong Kong showing sandy shore study sites. Squares indicate the sites visited by George (1982) and circles indicate the sites visited in the present study. Closed circle / square indicates the presence of *O. mortoni*. Empty circle / square indicated absence of *O. mortoni*.

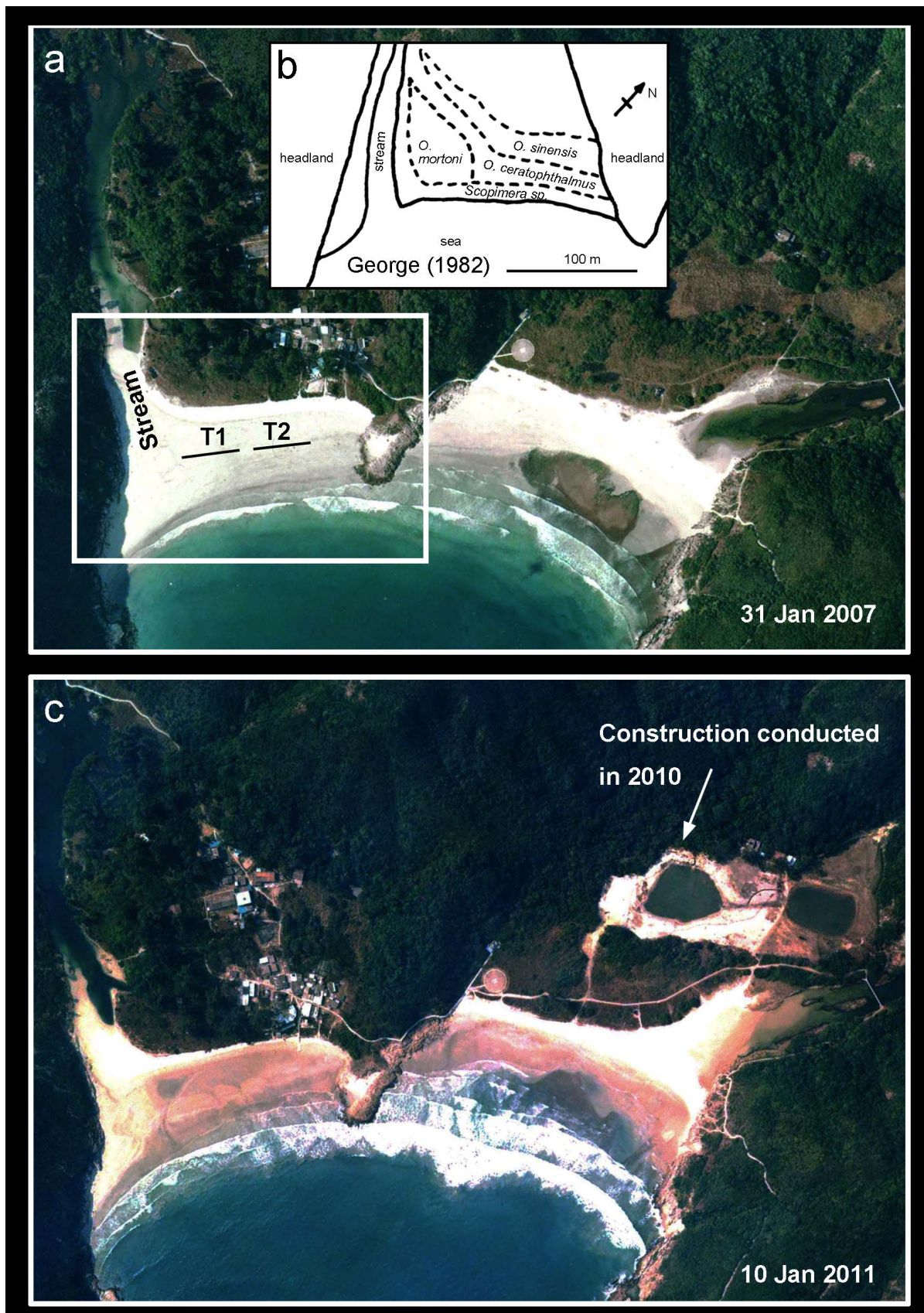


FIGURE 2. Sai Wan, Hong Kong, the type locality of *O. mortoni*. a. Aerial photos showing the topography of the site, the streams in the southwest part and the location of the two transects T1 and T2. b. map of Sai Wan, redrawn from George (1982) indicating the distribution of *Ocypode* species. c. same location, aerial photo taken in Jan 2011, showing the construction sites in the northern flank of the site. Aerial photo, courtesy of Map Office, Lands Department, the Hong Kong SAR Government.

Vertical distribution pattern and relative abundance of *O. mortoni* and *O. ceratophthalmus*. Preliminary studies revealed that *O. mortoni* was present at Sai Wan, Bui O and Fan Lau. Quantitative studies were further conducted on these three sites. In Sai Wan, two 30-meter parallel transects were established on the same tidal level at 2.0–2.3 metres above Chart Datum (C.D.). These levels contained the highest abundances of *Ocyropode* burrow openings. One transect (T1, see Fig. 2a) was located close to the stream, where George (1982) observed high abundances of *O. mortoni* (Fig. 2b). Another transect (T2, see Fig. 2a) was located further from the stream. No transects were placed above 2.5 m as levels higher up the shores were vegetated and only *O. sinensis* was present. Along each transect, 20 quadrats (50 × 50 cm²) were placed randomly. All *Ocyropode* burrows found in the quadrats were excavated and the crabs were preserved in 95% ethanol upon capture. In Bui O and Fan Lau, where there were fewer and more scattered burrows, random sampling would have resulted in very few burrows being included in the quadrats and therefore at these sites all burrows along the 30-meter transect located at 2.0–2.3 metres above C.D. were excavated and the crabs collected. The sampling in Bui O and Fan Lau can allow investigation of the abundance of *O. mortoni* and its relative abundance with sympatric species along the transect. Specimens have been deposited in National Museum of Natural Science, Taiwan (NMNS), the Zoological Reference Collection, Raffles Museum of Biodiversity Research, Singapore (ZRC), and the Coastal Ecology Laboratory of Academia Sinica, Taiwan (CEL).

Morphological observations. Morphology such as carapace, cheliped surfaces and gastric mill ossicles of *O. mortoni* specimens and additional material were observed and photographed under a stereomicroscope installed with a digital camera. Male gonopods (first pleopods; abbreviated as G1) were critical-point dried, gold coated and examined using a SEM (FEI Quanta 200, following SEM techniques in Chan *et al.* 2007a, b).

Molecular analyses. Total genomic DNA was extracted from soft tissue of specimens, using the commercial QIAamp Tissue Kit (QIAGEN, Hilden, Germany). Amplification of mitochondrial COI sequences through polymerase chain reaction (PCR) was performed by the universal primers LCO1490 and HCO2198 (Folmer *et al.* 1994) was used: 5'-GGTCAACAAATCATAAAGATATTGG-3' and 5'-TAAACTTCAGGGTGACCAAAAAATCA-3' under the following conditions: 2 minutes at 95°C for initial denaturing, then 35 cycles of 1 minute at 95°C, 1 minute at 50°C, 1 minute at 72°C with a final extension for 5 min. at 72°C. Sequences were generated using the same sets of primers and determined on an Applied Biosystems (ABI) 3100 automated sequencer using the ABI Big-dye Ready-Reaction mix kit, following the standard cycle sequencing protocol. The extracted sequences were aligned manually, with the aid of Clustal W (Thompson *et al.* 1994). A pairwise distance matrix is constructed using Kimura (1980) 2-parameter distance (K2P distance) in MEGA 5 (Tamura *et al.* 2011) to estimate inter- and intraspecific genetic diversity. Sequences of different haplotypes were deposited in the DNA Data Bank of Japan (DDBJ) database (accession nos. given in Table 1).

The best-fitting model for sequence evolution of the COI dataset was determined to be TIM2+G by Jmodeltest (v. 0.1.1, Posada 2008; Guindon & Gascuel 2003), selected under the Bayesian information criterion (BIC), and the settings provided were applied in MrBayes (v. 3.2.1, Ronquist *et al.* 2012) for conducting Bayesian Inference (BI) analysis, with 4 independent runs of 10⁷ generations each. Trees were sampled every 1000 generations. The convergence of chains was determined by the effective sample size (ESS) (>200 as recommended) in Tracer (v. 1.5, Rambaut & Drummond 2009) and the first 1100 trees were discarded as the burnin (determined by the average standard deviation of split frequency values below the recommended 0.01; Ronquist *et al.* 2005). Maximum likelihood (ML) analysis was conducted in RAxML (v. 7.2.6, Stamatakis *et al.* 2008). The model GTR + G (i.e. GTRGAMMA) was used with 100 runs, and found the best ML tree by comparing the likelihood scores. The robustness of the ML tree was evaluated by 1000 bootstrap pseudoreplicates under the model GTRGAMMA. A consensus maximum parsimony (MP) tree was constructed using PAUP* (v. 4.0b10, Swofford 2003) with 2000 bootstrap replications of a simple heuristic search, tree bisection-reconnection (TBR) branch-swapping, and 100 random-addition sequence replications.

Results

Systematics

Family OCYPODIDAE Rafinesque, 1815

Genus *Ocypode* Weber, 1795

Ocypode mortoni George, 1982

(Figs 3–6)

Ocypode mortoni George, 1982: 187, figs. 1C, 2C, pls. 3. — Dai & Yang 1991: 457, fig. 203B. — K. Sakai 2000: 1159, figs. 2a–b, 3a–e.

Ocypode macrocera (not *O. macrocera* H. Milne Edwards, 1837) — Dai *et al.* 1985: 370, figs. 1–7, pls. 1–2.

Material examined. 6♂♂2♀♀ (CEL-Ocy-HK-003), Sai Wan, Hong Kong, 20 Jun. 2009, coll. K. Wong *et al.*; 3♂♂1♀ (CEL-Ocy-HK-021), Bui O, Hong Kong, 21 Jun. 2009, coll. H.-N. Chen *et al.*; 1♀ (CEL-Ocy-HK-017), Fan Lau, Hong Kong, 23 Jun. 2009, coll. K. Wong; 1♂1♀ (ZRC-2012.0141), Sai Wan, Hong Kong, 20 Jun. 2009, coll. K. Wong *et al.*; 1♂ (NMNS-6879-001), Sai Wan, Hong Kong, 20 Jun. 2009, coll. K. Wong *et al.*

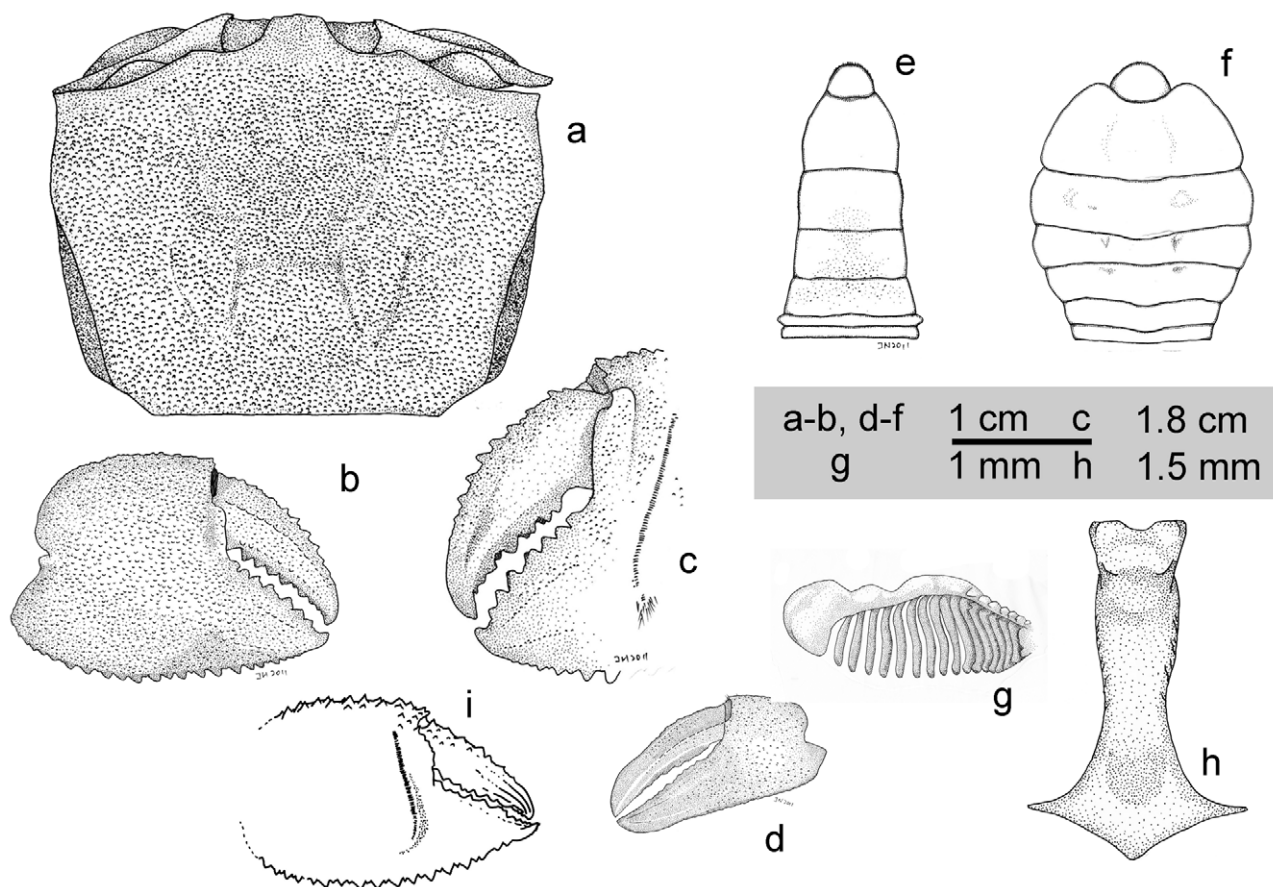


FIGURE 3. Morphology of *O. mortoni*: a, dorsal surface of carapace; b, outer surface of major cheliped palm; c, inner surface of major cheliped palm, showing stridulating ridge; d, outer surface of minor cheliped; e, male abdomen; f, female abdomen; g, zygocardiac ossicle; h, urocardiac ossicle; i, inner surface of major cheliped palm of the type specimen, redrawn from George (1982).

Diagnosis. Carapace subquadrangle, regions defined by grooves; cornea with ocular stylets on the distal end, never longer than cornea, even in adult males (Figs. 3a, 5a–b). Outer surface of major cheliped palm often lemon-yellow in color (Fig. 5b); stridulating ridge on inner surface of major cheliped consist of 44–65 evenly-spaced striation (Fig. 3c, i); base of immovable finger broad, compressed; finger tips of minor cheliped rounded (Fig. 3b–c).

Description. Carapace subquadrangle, broader than long, longitudinally convex, anterior 1/3 deflected, raised around branchial regions; surface evenly covered by beaded tubercles; regions markedly divided by grooves, those around cardiac region H-shaped, deeper; antero-lateral teeth absent; external orbital tooth forming right angle; anterior 1/3 of lateral margins roughly parallel, diverging posteriorly (Figs. 3a, 6a). Cornea on elongated stalks, bilobed, distal end extended with ocular stylets about half of cornea length in mature males (Figs. 3a, 6a). Small,

acute spine adjacent to antennule, suborbital ridge composed of 30–35 elongated granules; pterygostomian region sparsely covered by flattened granules (Fig. 6b). External maxillipeds flat, ischium longer than merus, both roughly quadrate (Fig. 6b), inner margins lined with long, light-colored stiff setae; exopod narrow, ill-developed, reaching 1/4 of merus length.

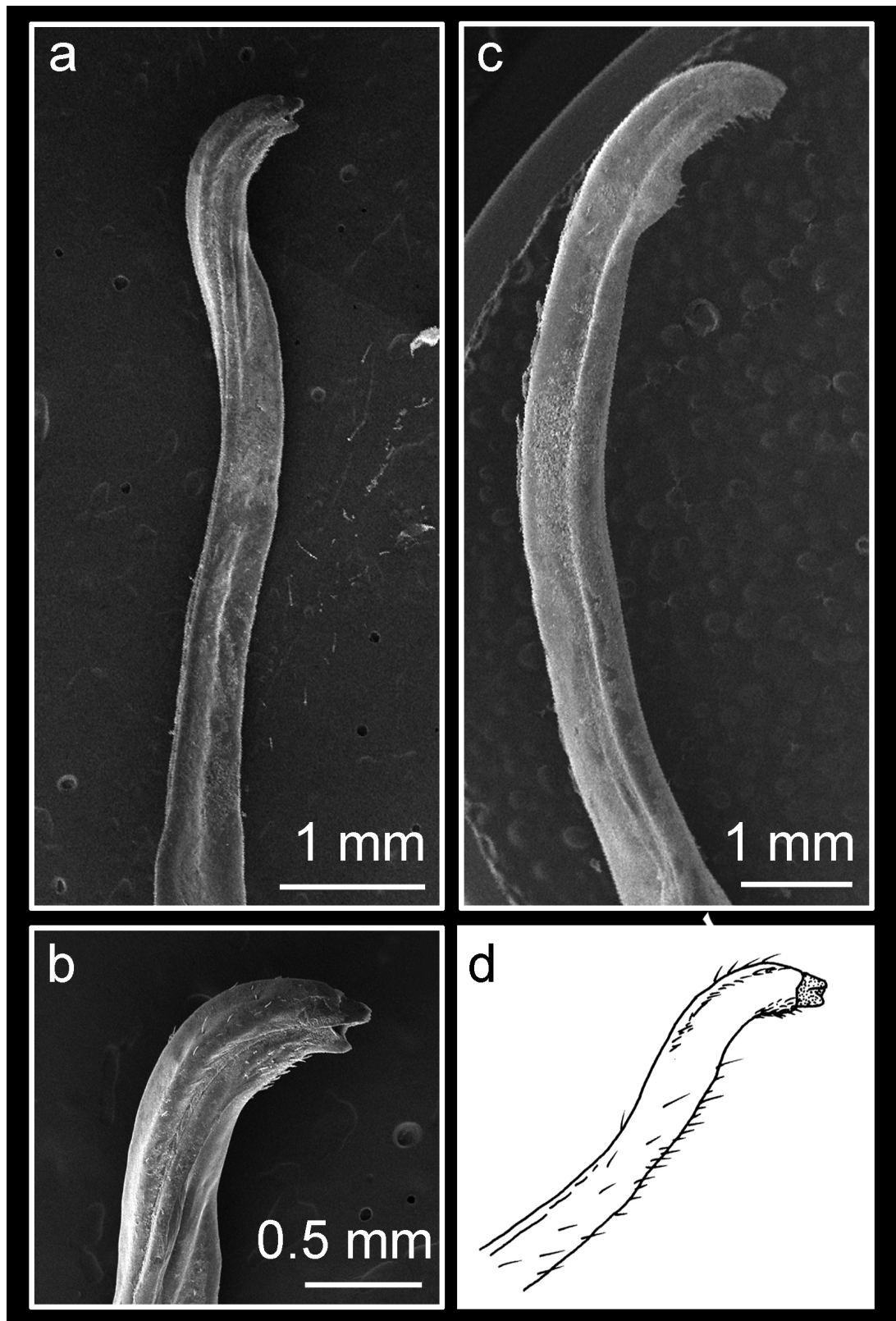


FIGURE 4. Morphology of male first pleopod (G1) of *O. mortoni*: a, ventral view of left G1; b, tip of left G1, ventral view; c, dorsal view of right G1; d, G1 of type specimen, redrawn from George (1982).

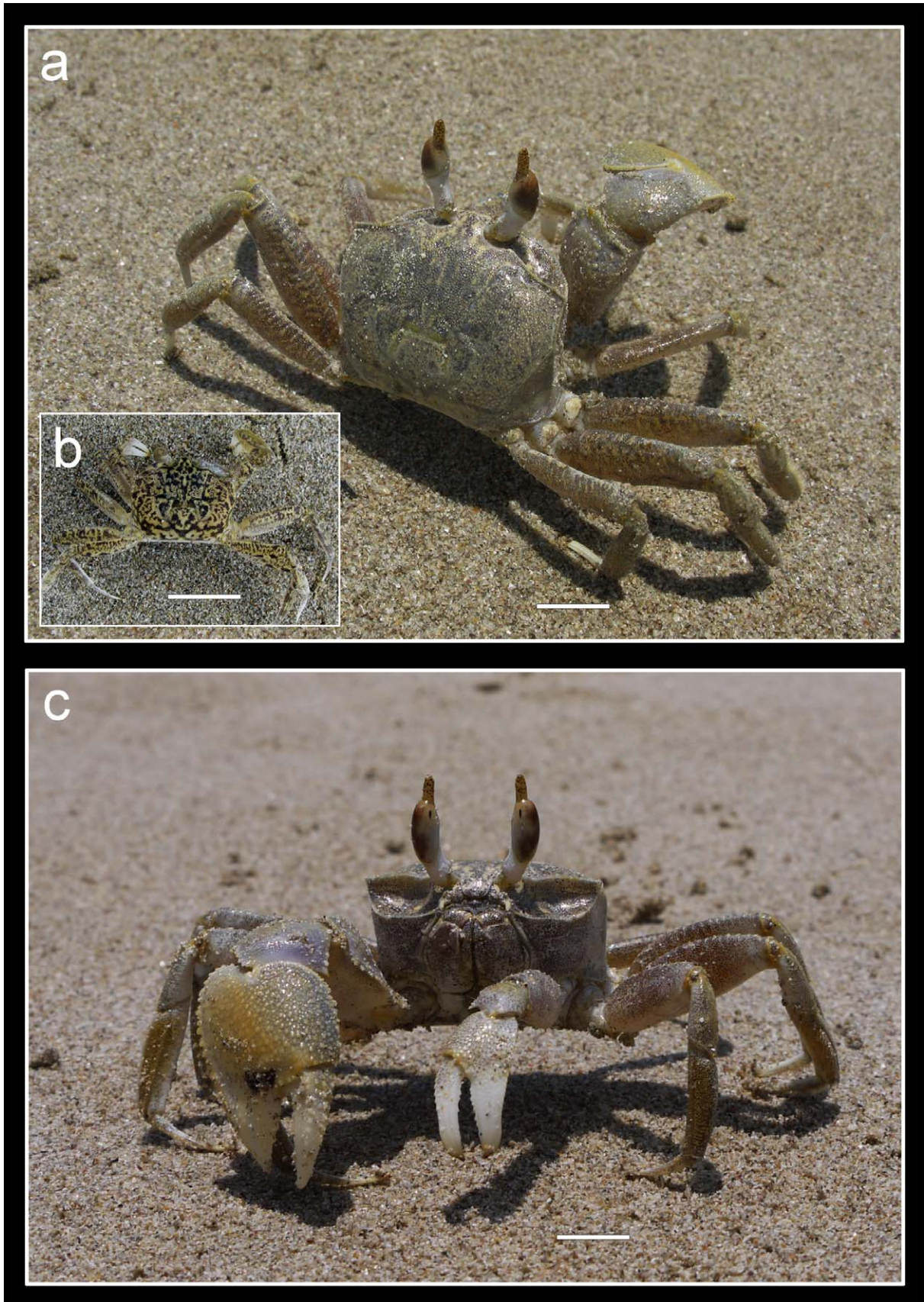


FIGURE 5. Life coloration of *O. mortoni*: a, dorsal view, adult; b, dorsal view, juvenile. c, frontal view, adult. Scale bars represent 1 cm.

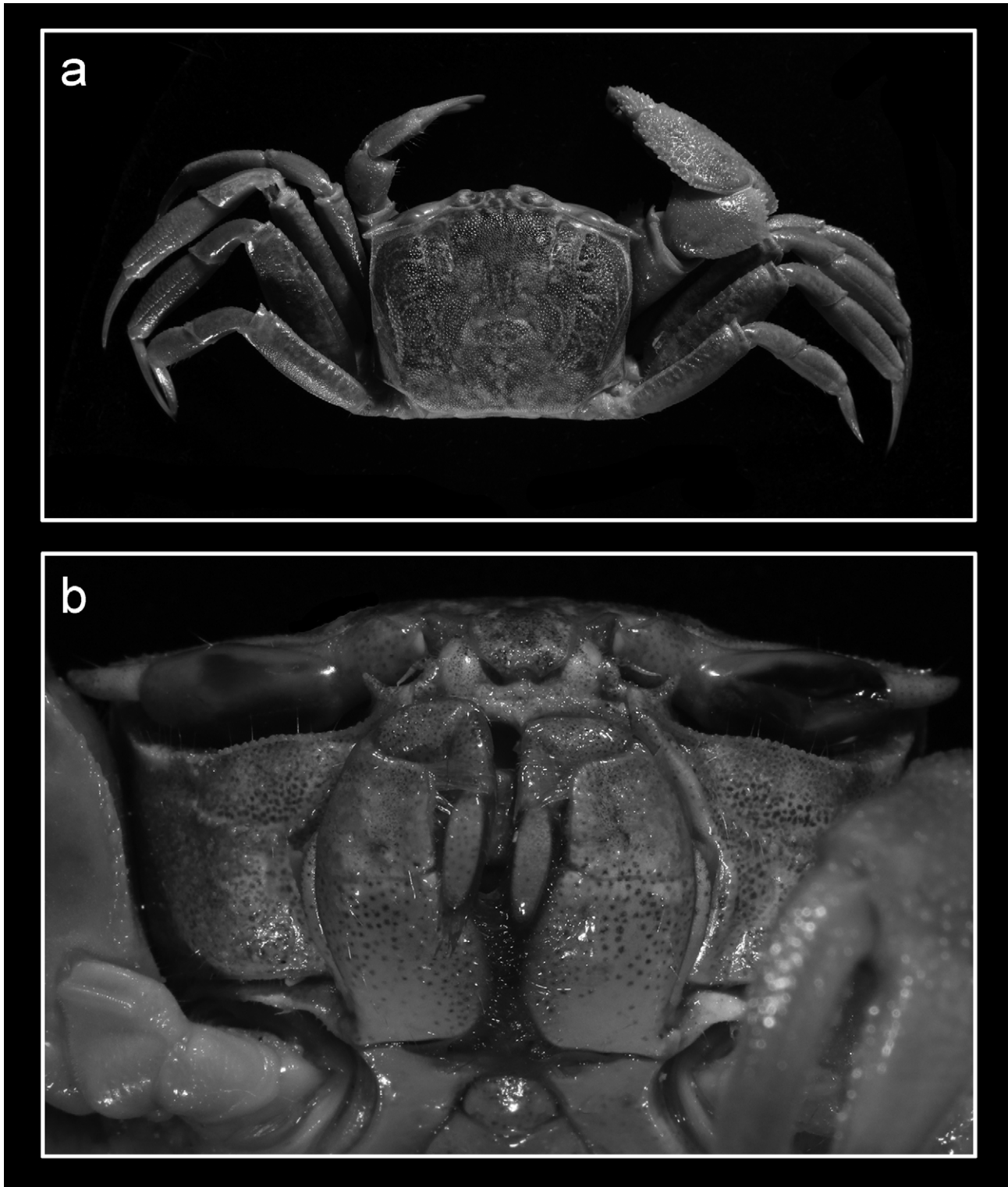


FIGURE 6. *Ocypode mortoni*: ♂ from Sai Wan, carapace 24.5mm x 21.2 mm (CEL-Ocy-HK-003): a, dorsal view; b, frontal view.

Chelipeds asymmetrical; major merus triangular in latitudinal cross-section; lower fringe of inner surface leaf-like, margin strongly serrated; carpus as long as broad, inner angle armed with strong tooth, outer-distal margin serrated; outer surface of palm covered with beaded tubercles of varies sizes, margins strongly serrated (Fig. 3b); inner surface with latitudinal stridulating ridge composed of 44–65 evenly-spaced striations (Fig. 3c, i); immovable finger triangular, high at base, which compressed and leaf-like, ridge of rounded tubercles lined subparallel to cutting edge, movable finger lined with ridge of large rounded tubercles; fingers shorter than palm, tips pointed

(Fig. 3b–c, i, 5b). Minor cheliped carpus slightly longer than broad, inner angle armed with strong tooth, fingers lined with ridge of rounded granules along respective median lines, tips rounded (Figs. 3d, 5b). Ambulatory legs slender, 2nd and 3rd subequal in total length, longest, 4th shortest; meri compressed, robust, distal end of anterior margin weakly armed with tooth (Fig. 6a). Propodi and dactyli of the first three pairs lined with rolls of soft plumose setae along median line on anterior surface; dactyli pointed, curved inwards for the first three pairs and outwards for the 4th (Fig. 6a).

Urocardiac ossicle constricted beneath the T-shaped bifurcation (Fig. 3h), and mildly expanded on the posterior half; zygocardiac ossicle bearing 16 comb-like teeth (Fig. 3g).

Ventral surface of carapace glabrous except for tuft of soft setae between bases of 2nd and 3rd ambulatory legs. Male abdomen elongated; telson rounded, proximal margin convex; 6th somite as long as broad, lateral margins convex; 5th somite broader than long, roughly rectangular (Fig. 3e). Male G1 slightly sinuous when viewed ventrally, not apparently tapering; distal part strongly curved outwards, tip divided into two chitinous beak-like lobes, upper lobe larger; neck of the distal curve fringed with short setae (Fig. 4a–b, d). Female abdomen broad, telson as semicircle, narrow, proximal margin convex, 6th somite broader than long, lateral margins convex; preceding somites much broader than long, roughly rectangular (Fig. 3f).

Coloration. Dorsal surface of carapace dark brownish with light-colored thin sinuate lines (Fig. 5a); pterygostomial region and external maxillipeds creamish with microscopic dark dots; orbits grayish, ocular stylets yellowish (Fig. 5b); ambulatory legs creamish with random faint blotches; outer surface of major cheliped distinctly yellow (Fig. 5b; might still be observable after alcohol preservation). Juveniles displaying a more conspicuous pattern with dark, sinuous markings on a pale yellowing background, on both dorsal surface of carapace and ambulatory leg meri (Fig. 5c). Cheliped palm also yellowish, with whitish fingers.

Distribution. Hong Kong (type locality), Guangdong, Guangxi, Hainan Island (Dai *et al.* 1985; Dai & Yang 1991), and the Pacific coast of Shikoku, Japan (K. Sakai 2000). See also “Remarks” below.

Remarks. As suggested in previous works, *O. mortoni* morphologically resembles *O. fabricii* and *O. nobilii* (George 1982), which all develop short ocular stylets above the cornea. *Ocypode mortoni* can be distinguished from the others based on the number of striations along the stridulating ridge on the inner surface of the major cheliped palm: up to 54 for *O. mortoni*, 120 for *O. nobilii* and 108–141 in *O. fabricii* (see George & Knott 1965; George 1982). In our specimens, there were 44–65 striations on the stridulating ridges which conforms to *O. mortoni* (see Fig. 3c, i). G1 morphology of our specimens, with two chitinous beak-like structures at the distal tip, also matches that of the original description (see Fig. 4).

Shen (1940) recorded *O. ceratophthalmus*, *O. cordimanus* and *O. stimpsoni* in his review of the brachyura of Hong Kong, based on his own collections, materials from other institutes and records in the earlier literature (Stimpson 1858, 1907; Balss 1922; Gee 1926). The presence of *O. ceratophthalmus* has long been validated (see Morton & Morton 1983), however the records of *O. cordimanus* has been shown to represent *O. sinensis* (Huang *et al.* 1998). The last species *O. stimpsoni*, appears to be a temperate species (Dai *et al.* 1985), and reported from Japan (T. Sakai 1939), Korea (Kim 1973), North China (Shen 1932) and Taiwan (Huang *et al.* 1992). Records from the coasts of South China (e.g. Shen 1936, 1940), thus, represents the southern limits for *O. stimpsoni* (see also Dai *et al.* 1986; Dai 1991; Dai & Yang 1991). However, studies on the genus from China (Dai *et al.* 1985) recognized material from Hong Kong as *O. mortoni* (then as *O. macrocera*) instead of *O. stimpsoni*.

Our molecular phylogenetic analyses (Fig. 9), show *O. stimpsoni* and *O. mortoni* to be closely related, which is not surprising as they are very similar morphologically except for the presence of ocular stylets in *O. mortoni* adults. It is very difficult to separate juveniles or younger individuals of the two species. As for the Hong Kong records of “*O. stimpsoni*” by Shen (1940), all four sites where he collected material (Shatin, Ngau Chi Wan, Repulse Bay and Stanley) have been altered or urbanized over the last 70 years, and no further records have been found since then (see also George 1982; Morton & Morton 1983). Extensive searches performed in this study also failed to acquire any material of this species. While it is tempting to regard the earlier records of *O. stimpsoni* as probable misidentifications, the past presence and subsequent local extinction is nevertheless still possible. As Hong Kong is within the range of the known southern distribution of *O. stimpsoni* we tentatively continue to include it as known from there, though with some hesitation.

For records in the East Asian region, aside from southern China (including Hong Kong), *Ocypode mortoni* has only been recorded on the Pacific coast of Shikoku, Japan (see K. Sakai 2000). The illustrations of the Japanese material agrees with our specimens, in terms of the number of striae along the stridulatory ridge (Shikoku

specimens: 60 in male and 52 in female; Hong Kong specimens: 44–65), and in the form of the zygo-cardiac ossicle (Fig. 3h). However, the distal tip of the G1 in material from Hong Kong, when viewed ventrally, ends with a pronounced chitinous bi-lobed, pointed, beak-like structure, with the upper lobe larger than the lower; and fine setae on the “neck” of the curvature beneath (see Fig. 4a–b, d). Both characters are somewhat wanting in Sakai’s (2000) illustrations (fig. 3d in K. Sakai 2000). The importance of these slight variations is uncertain, and we would opt to consider both the Hong Kong and Japanese records conspecific, at least for now. While *O. mortoni* could be expected to occur in Taiwan and adjacent areas, no specimens have been collected despite numerous efforts to find it, and none are present in museum collections. The biogeography of the species remains unclear, and needs further investigation based on more extensive targeted collecting.

Distribution pattern of *Ocypode* spp. in Hong Kong. From the seven sandy shores surveyed, *Ocypode ceratophthalmus* was collected in all sites while *O. mortoni* was only limited at Sai Wan, Tong Fuk and Bui O (Fig. 1). *Ocypode mortoni* was found at the same tidal levels as *O. ceratophthalmus* (2.0–2.3 m above C.D.) but not on higher shores, where the zone is solely dominated by *O. sinensis*.

Vertical distribution pattern at Sai Wan and relative abundance of *O. mortoni* and *O. ceratophthalmus*. At Sai Wan, along the transect close to the stream (T1), average density of *O. ceratophthalmus* reached 2.2 individuals m⁻², while for *O. mortoni* only occurred at 0.4 individuals m⁻². On the transect further beyond the stream (T2), *O. ceratophthalmus* maintained an average density of 1.8 individuals m⁻², and *O. mortoni* at 0.8 individuals m⁻² (Fig. 7). In Fan Lau and Bui O, the relative abundance of *O. ceratophthalmus* to *O. mortoni* was 96:4%, and 83:17% respectively (Fig. 7).

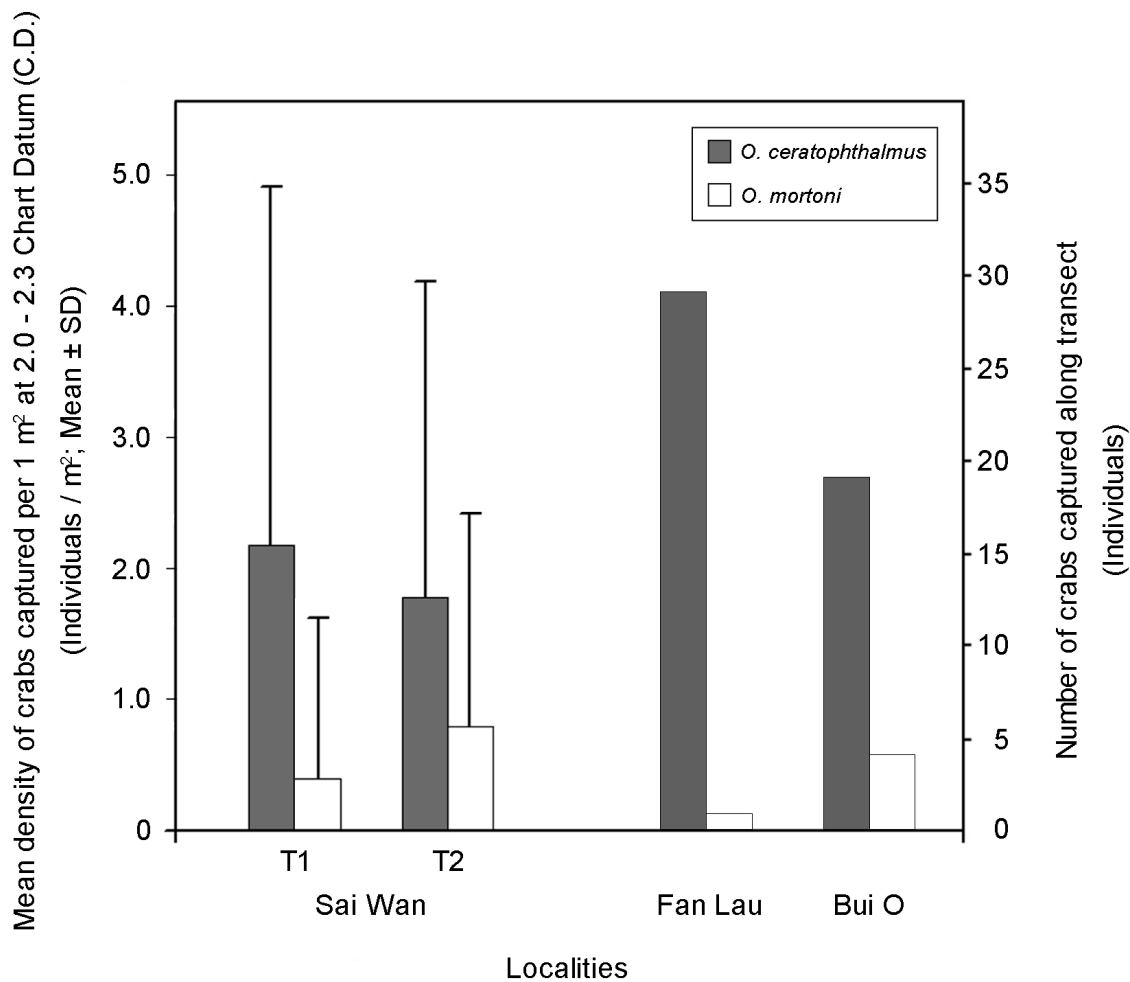


FIGURE 7. Density of *O. mortoni* and *O. ceratophthalmus* along T1 and T2 in Sai Wan (refer to Y-axis on left hand side); and the total number of crabs collected in the transects at Fan Lau and Bui O along the 30 meter-transects (refer to the Y axis on the right hand side).

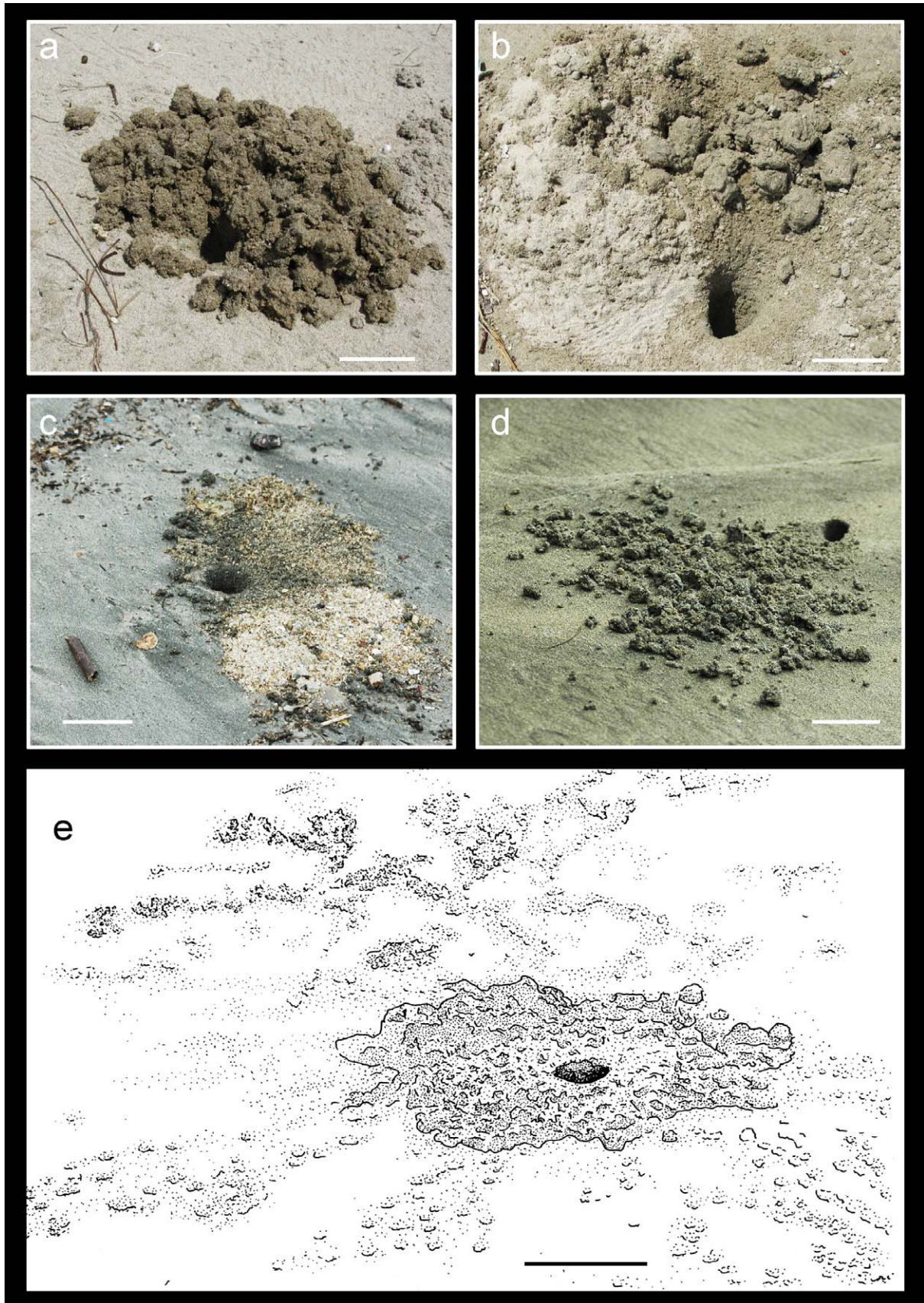


FIGURE 8. Burrow morphology of *Ocyptode* spp.: Burrow opening: a–b from Sai Wan (*O. ceratophthalmus* / *O. mortoni* were both found in these two types of burrows, see Discussion); c–d from Bui O (*O. ceratophthalmus* / *O. mortoni* were both found in these two types of burrows). Note the variation of arrangements of sediment piles around the opening; e, burrow opening of *O. mortoni* redrawn from George (1982). Scale bars represent 5 cm.

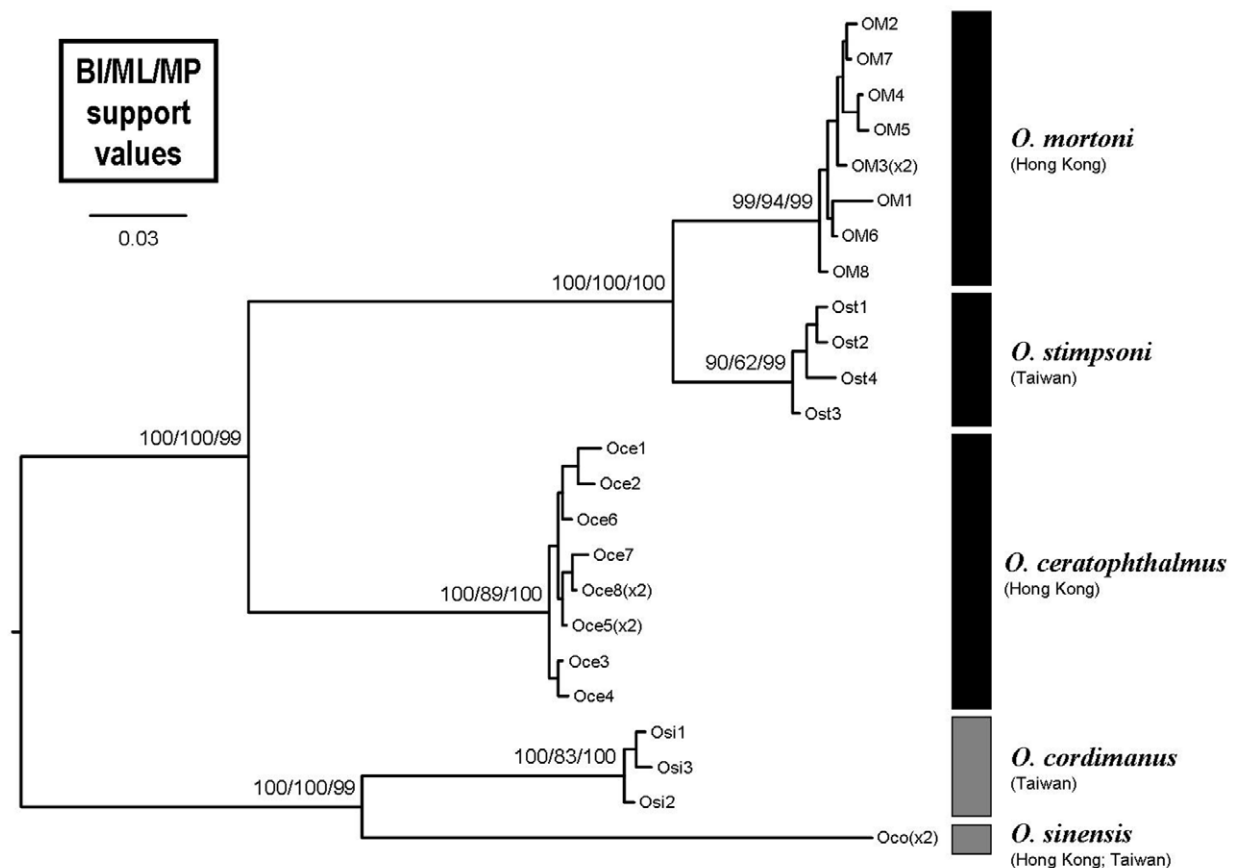


FIGURE 9. Bayesian Inference (BI) tree showing phylogenetic relationships of East Asian *Ocypode* spp., based on COI gene. Additional support values of BI, maximum likelihood (ML) maximum parsimony (MP) analyses printed at the nodes. For details of haplotypes see Table 1, with species and localities behind. Black bars represent “ridge-bearing” species, and gray bars represent “ridge-less” species (see Discussion).

Molecular analysis. From 28 individuals of *Ocypode* spp., 24 haplotypes (see Table 1) of COI sequences, each of 636 basepairs were obtained. The alignment are AT-rich (in average A: 27.6%, T: 34.3%, G: 17.0%, C: 21.2%), within which 156 sites are variable, and 130 of these are parsimoniously informative.

Pairwise nucleotide divergences (K2P distance) and the mean number of differences of the COI sequences between five East Asian *Ocypode* species are displayed in Table 2. Apart from *O. cordimanus* which involved a single haplotype, the remaining 4 species show a certain degree of intraspecific variation. The percentage of bp differences and K2P can be as much as 1.57% (10/636) and 1.60% (for *O. mortoni*), respectively.

A phylogenetic (BI) tree of COI of East Asian *Ocypode* species is shown in Figure 9, with the additional support values of ML and MP analyses beside the nodes. Results show all the studied *Ocypode* species formed well-supported, distinct clades at the species level. *Ocypode mortoni* and *O. stimpsoni* are genetically the closest related (average K2P distance: 6.25%, see Table 2). Of the 24 haplotype analysed, the East Asian *Ocypode* can be divided into two main clades with robust clade support: one contains the species with a stridulating ridge on the inner surface of major cheliped, i.e. *O. mortoni*, *O. stimpsoni* and *O. ceratophthalmus*; while the other clade contains species that lack ridges on the inner side of the chelae: *O. cordimanus* and *O. sinensis*. Within the first clade, *O. ceratophthalmus* is sister to the *O. mortoni* and *O. stimpsoni* subclade.

Discussion

In the present study, *Ocypode mortoni* George, 1982, was redescribed in detail, and new data on its distribution and abundance in Hong Kong was obtained. George (1982) surveyed eight sandy shores around Hong Kong, among which *O. mortoni* was collected from only three, Sai Wan, Tong Fuk and Starfish Bay (Fig. 1), and Sai Wan was designated the type locality. The present study also recorded *O. mortoni* from Sai Wan, but additionally from Bui O and Fan Lau on Lantau Island (Fig. 1). George’s (1982) original sites in Starfish Bay and Tong Fuk were searched

thoroughly over multiple visits, but *O. mortoni* could not be found. Very probably changes to these beaches over the last 30 years have made them no longer favourable for this species.

George (1982) provided a map showing the distribution of *Ocypode* species in Sai Wan (see Fig. 2b), and reported that *O. mortoni* was restricted in the vicinity of a stream in the southwestern part of Sai Wan. He considered that *O. mortoni* may prefer the presence of freshwater influence, while *O. ceratophthalmus* prefer a fully seawater environment. In the present study, we found that the abundance of *O. mortoni* dug along the transect closer to the stream (T1: 2 individuals; 0.4 individuals / m²) remains lower than that further away (T2: 4 individuals; 0.8 individuals / m²). There was no observed decreasing trend in abundance from southwest to northeast (with decreasing freshwater input). Additional night searches in the stream area also failed to collect many individuals of *O. mortoni*, and thus our results do not support George's (1982) preliminary ecological hypothesis.

In the present study, *O. mortoni* is sympatric with *O. ceratophthalmus*, but *O. mortoni* remains much lower in relative abundances (at most 31%: 69% along T2 of Sai Wan, see above and Fig. 2a). Among the three sandy shores where *O. mortoni* was collected, the abundance of *O. mortoni* was always less than half that of *O. ceratophthalmus*. This confirms the observation of George (1982), that *O. mortoni* is relatively rare. *Ocypode sinensis* is distributed higher up along the supratidal zones on shores of Hong Kong, often among the coastal vegetation, and is clearly ecologically separated from *O. mortoni* and *O. ceratophthalmus* (personal observation).

George (1982) reported that *O. mortoni* has distinctive burrow openings, with "feeding lines" of sediment pellets radiating from the opening (see also Fig. 8e). In the present study, burrows with the distinctive appearance described by George (1982) were not found. Indeed, apart from typical *O. ceratophthalmus* burrow openings, where excavated sediments are scattered in a single direction (see Fig. 8b–d; Chan *et al.* 2006), burrow openings generally showed a range of appearances, including a peculiar entrance with enormous lumps of loosened substrate piled around the opening (Fig. 8a). Thus there seems to be no basis to George's assertion that *O. mortoni* burrows have a distinctive form.

As a note on dietary habits of the two superficially sympatric species at Sai Wan, preliminary gastric content analysis remained statistically inconclusive due to small sample sizes. However, qualitatively, crustacean fragments and fish tissue have been found in samples from *O. ceratophthalmus* but not in *O. mortoni*. Fine traces of unidentified organic materials were often seen from samples of *O. mortoni* (personal observation). This temptingly verify past reports of predatory / scavenging nature of the *O. ceratophthalmus* (see Barass, 1963; Hughs, 1966; Smith, 1975). Three out of seven retrieved samples from *O. mortoni* consisted of large proportion of inorganic sediments. Hypothesis raised in George (1982), "behaves much as the sand bubblebers (*Scopimera* sp.)" based on burrow opening appearance might be true, yet this potential aspect in niche segregation cannot be statistically justified at this stage. Further investigation on dietary habits of *Ocypode* sp. in Hong Kong, both intra- and interspecies, can be conducted in the future, and hopefully possible conditions and mechanisms of niche partitioning of the two *Ocypode* spp. can be better understood.

Our molecular analyses show that all *Ocypode* species in the present study can be well differentiated at species level. Each species-level taxon had at least 6.25% average divergence in COI sequences (in *O. mortoni* vs. *O. stimpsoni*), which can be considered sufficient for specific recognition. Other studies for identifying species level differences among intertidal brachyurans have concluded lower percentage differences to be valid, e.g. 2.79% (*Uca splendida* vs. *U. crassipes*; Shih *et al.* 2012); 3.62% (*Mictyris guinotae* vs. *M. brevidactylus*; Davie *et al.* 2010); 4.43% (*Scopimera ryukyuensis* vs. *S. globosa*; Wong *et al.* 2010); 4.74% (*Helice tridens* vs. *H. latimera* clade; Shih & Suzuki 2008); and 5.32% (*Uca jocelynae* vs. *U. neocultrimana*; Shih *et al.* 2010). *Ocypode* spp. presented in this study can be divided into 2 main clades, one containing species with no stridulating ridge on the inner surface of major cheliped (the "ridge-less clade", represented by gray bars in Fig. 9), and the other with a conspicuous stridulating structure (the "ridge-bearing clade", represented by black bars in Fig. 9). Dai *et al.* (1985) concluded a close relationship between *O. mortoni* and *O. stimpsoni*, based on the morphological similarities of the stridulating ridge, the chelipeds and the G1, and this is supported by our phylogenetic tree (Fig. 9). Among the "ridge-bearing clade", *O. ceratophthalmus* is distinguished from an *O. mortoni* and *O. stimpsoni* subclade. This is supported morphologically by the form of the stridulating ridge (*O. ceratophthalmus* consisting tubercles and striations; *O. mortoni* and *O. stimpsoni* with only striations), and the form of major cheliped (the immovable finger of both *O. mortoni* and *O. stimpsoni* has a much compressed, broad triangular base). A true understanding of the broader phylogenetic relationships within the genus *Ocypode* will require a larger study with many more species included from across its entire range.

TABLE 1. Haplotypes of COI gene of *Ocypode* spp. from East Asia used in this study.

Species	Haplotype of COI	Sample size	Sequence no.	Specimen Catalogue no.	Locality	DDBJ access. no.
<i>O. mortoni</i>	OM1	1	BOO_1	CEL-Ocy-HK-021	Bui O, Hong Kong	AB751384
	OM2	1	BOO_2	CEL-Ocy-HK-021	Bui O, Hong Kong	AB751385
	OM3	2	BOO_3	CEL-Ocy-HK-021	Bui O, Hong Kong	AB751386
	OM4	1	SWO_16	CEL-Ocy-HK-021	Bui O, Hong Kong	AB751387
	OM5	1	BOO_4	CEL-Ocy-HK-021	Bui O, Hong Kong	AB751388
	OM6	1	SWO_11	CEL-Ocy-HK-003	Sai Wan, Hong Kong	AB751389
	OM7	1	SWO_12	CEL-Ocy-HK-003	Sai Wan, Hong Kong	AB751390
	OM8	1	SWO_13	CEL-Ocy-HK-003	Sai Wan, Hong Kong	AB751391
<i>O. stimpsoni</i>	Ost1	1	XSO_1	CEL-Ocy-TW-012	Siangshan, Hsinchu, Taiwan	AB751395
	Ost2	1	XSO_2	CEL-Ocy-TW-012	Siangshan, Hsinchu, Taiwan	AB751396
	Ost3	1	XSO_3	CEL-Ocy-TW-012	Siangshan, Hsinchu, Taiwan	AB751397
	Ost4	1	XSO_4	CEL-Ocy-TW-012	Siangshan, Hsinchu, Taiwan	AB751398
<i>O. ceratophthalmus</i>	OCe1	1	SWO_2	CEL-Ocy-HK-002	Sai Wan, Hong Kong	AB751375
	OCe2	1	SWO_3	CEL-Ocy-HK-003	Sai Wan, Hong Kong	AB751376
	OCe3	1	SWO_4	CEL-Ocy-HK-004	Sai Wan, Hong Kong	AB751377
	OCe4	1	SWO_5	CEL-Ocy-HK-005	Sai Wan, Hong Kong	AB751378
	OCe5	2	SWO_6	CEL-Ocy-HK-006	Sai Wan, Hong Kong	AB751379
	OCe6	1	SWO_7	CEL-Ocy-HK-007	Sai Wan, Hong Kong	AB751380
	OCe7	1	SWO_8	CEL-Ocy-HK-008	Sai Wan, Hong Kong	AB751381
	OCe8	2	SWO_9	CEL-Ocy-HK-009	Sai Wan, Hong Kong	AB751382
			SWO_10	CEL-Ocy-HK-010	Sai Wan, Hong Kong	
			HKO_1	CEL-Ocy-HK-030	Silver Mine Bay, Hong Kong	
<i>O. cordimamus</i>	Oco	2	ATO_17	CEL-Ocy-TW-015	Taitung, Taiwan	AB751383
			GIO_1	CEL-Ocy-TW-023	Lyudao (Green Island), Taitung Taiwan	
<i>O. sinensis</i>	Osi1	1	KTO_4	CEL-Ocy-TW-024	Kenting, Pingtung, Taiwan	AB751392
	Osi2	1	TFO_4	CEL-Ocy-HK-015	Tong Fuk, Hong Kong	AB751393
	Osi3	1	TFO_6	CEL-Ocy-HK-015	Tong Fuk, Hong Kong	AB751394

TABLE 2. Matrix of intra- and interspecific percentage divergences, with K2P distances based on the COI sequences from 28 specimens of East Asian *Ocyptode* spp. Under interspecific values, pairwise K2P distances are printed in the lower-left portion of the table and mean number of differences on upper-right portion of the table. Values of range are given in parentheses.

Species	Intraspecific		Interspecific				
	Nucleotide divergence (%)	Mean nucleotide difference	<i>O. mortoni</i>	<i>O. stimpsoni</i>	<i>O. ceratophthalmus</i>	<i>O. cordimanus</i>	<i>O. sinensis</i>
<i>O. mortoni</i>	0.56 (0–1.60)	3.56 (0–10)	—	37.64 (33–41)	72.62 (69–77)	103.44 (102–105)	90.85 (87–93)
<i>O. stimpsoni</i>	0.74 (0.32–0.95)	4.67 (2–6)	6.25 (5.44–6.84)	—	70.30 (68–73)	105 (104–107)	94.33 (93–96)
<i>O. ceratophthalmus</i>	0.52 (0–1.11)	3.33 (0–7)	12.50 (11.81–13.34)	12.04 (11.64–12.53)	—	100.50 (98–103)	84.87 (82–88)
<i>O. cordimanus</i>	0 —	0 —	18.55 (18.24–18.88)	18.92 (18.70–19.34)	18.07 (17.56–18.57)	—	77 (75–79)
<i>O. sinensis</i>	0.523 (0.47–0.63)	3.33 (3–4)	15.94 (15.17–16.37)	16.65 (16.37–16.99)	14.83 (14.26–15.46)	13.50 (13.10–13.91)	—

The type locality of *Ocypode mortoni*, is an undisturbed sandy shore site at the southern end of Sai Wan, Hong Kong, where it is apparently rare. The type locality appears to have remained relatively pristine for the last 30 years (Fig. 2a–b), however in mid-2010 construction works commenced on the vegetated land above the beach of the northern flank, altering stream flows, and severely changing the landscape (see Cheung 2010; Fig. 2c paper). Impacts on intertidal organisms, including *Ocypode* species remain unknown. Such events are increasingly common in coastal areas of the region, and highlight the long-term conflicts of land ownership, development, and ecological conservation in Hong Kong, where recent efforts in preserving biodiversity have not been particularly satisfactory (see Kilburn & Cheng 2011).

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