

Octopicola huanghaiensis n. sp. (Copepoda: Cyclopoida: Octopicolidae), a new parasitic copepod of the octopuses Amphioctopus fangsiao (d'Orbigny) and Octopus minor (Sasaki) (Octopoda: Octopodidae) in the Yellow Sea

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Abstract A new species of parasitic copepod, Octopicola huanghaiensis n. sp., collected from the octopuses Amphioctopus fangsiao (d'Orbigny) and Octopus minor (Sasaki) (Octopoda: Octopodidae) in the Yellow Sea (off Qingdao, Shandong Province, China), is described. The new species is most similar to O. superba Humes, 1957, but can be distinguished from the latter by: (i) the third antennal segment having a different ornamentation; (ii) the fourth antennal segment of females much shorter than that in O. superba (49 vs 94 µm); (iii) males much smaller than females (mean body length 1.3 vs 2.0 mm, respectively) (vs similar male and female body size in O. superba, 1.9 mm and 1.8 mm respectively); and (iv) the presence of a spike at the posterior tip of each labrum flap. Octopicola huanghaiensis n. sp. is the first species of Octopicola Humes, 1957 reported from

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X. Du · C. Dong · S.-C. Sun (⊠) Institute of Evolution & Marine Biodiversity, Ocean University of China, Qingdao 266003, Shandong, People's Republic of China e-mail: sunsc@ouc.edu.cn *A. fangsiao* and *O. minor* and is the only species of the family Octopicolidae Humes & Boxshall, 1996 known in North Pacific waters.

Introduction

The genus Octopicola Humes, 1957 was included in the family Lichomolgidae Kossmann, 1877 in some earlier publications (Humes & Stock, 1972, 1973). Because it differs from the other members of the Lichomolgidae by retaining the primitive 6-segmented female urosome, Humes & Boxshall (1996) established a new family, Octopicolidae Humes & Boxshall, 1996, to accommodate it. Currently, the genus contains four species, all parasites of octopuses (Humes, 1957; Stock et al., 1963; Humes, 1963; Humes, 1974). Among these species, Octopicola superba Humes, 1957 (see Humes, 1957; Bocquet & Stock, 1960; Cavaleiro et al., 2013) and Octopicola antillensis Stock, Humes & Gooding, 1963 (see Stock et al., 1963; Humes & Stock, 1973) are distributed in the Atlantic Ocean, Octopicola stocki Humes, 1963 was only reported from the Indian Ocean (Humes, 1963), and Octopicola regalis Humes, 1974 was recorded off New Caledonia Islands and Marshall Islands (Humes, 1974). To date, no species has been reported from North Pacific waters.

During our study, copepod specimens were collected from the octopuses *Amphioctopus fangsiao* (d'Orbigny) and *Octopus minor* (Sasaki) (Octopoda: Octopodidae) in the Yellow Sea. Morphological studies revealed that they represent a hitherto unknown species, which is described here as a new species of *Octopicola*.

Materials and methods

Octopuses were caught by the commercial trawlers from the Yellow Sea (off Qingdao, Shandong Province, China; $\sim 36^{\circ}$ N, $\sim 120^{\circ}$ E). We obtained the octopuses from fishermen at local fishing ports. Host species, number, sex and size, as well as the sampling dates are summarized in Table 1. After being transported to laboratory, each octopus was moved into a jar containing $\sim 5\%$ ethanol in filtered seawater. After several hours, potential copepod parasites had abandoned the octopus and fallen to the bottom of the jar. Copepod specimens were picked out under a dissecting microscope (Zeiss Stemi 508) and preserved in 75% ethanol or 10% formalin seawater. The habitus was drawn and body length was measured from the front of the head to the end of caudal rami (not including setae on caudal rami). Then specimens were dissected on a clean slide under the dissecting microscope. Separated body parts and appendages were mounted with lactophenol clearing solution as a permanent mounting medium and subsequently sealed with clear nail varnish. Then they were examined under a compound microscope under the maximum magnification of $1,000 \times$. All drawings were made with a drawing tube fitted on a Nikon E600 microscope. Measurements are in micrometres unless otherwise indicated and are presented as the range followed by the mean (\pm standard deviation where applicable) in parentheses.

Family Octopicolidae Humes & Boxshall, 1996 Genus *Octopicola* Humes, 1957

Octopicola huanghaiensis n. sp.

Type-host: Amphioctopus fangsiao (d'Orbigny) (Octopoda: Octopodidae).

Other host: Octopus minor (Sasaki) (Octopoda: Octopodidae).

Type locality: Yellow Sea (off Qingdao, Shandong Province, China).

Type-specimens: Holotype, female (MBM286096) ex *A. fangsiao*; paratype, male (MBM286097) ex *A. fangsiao*. The type-specimens are deposited in the Marine Biological Museum (MBM), Chinese Academy of Sciences, Qingdao, China.

Voucher material: 11 females, 26 males and 4 copepodids ex *A. fangsiao* and 2 males ex *O. minor*. These specimens are deposited at the Institute of Evolution & Marine Biodiversity, Ocean University of China, Qingdao, China under the accession numbers INV20180430A01-INV20180430A43.

Infection levels: The prevalence and intensity of *O. huang-haiensis* n. sp. in two host species are shown in Table 1.

ZooBank registration: To comply with the regulations set out in article 8.5 of the amended 2012 version of the International Code of Zoological Nomenclature (ICZN, 2012), details of the new species have been submitted to ZooBank. The Life Science Identifier (LSID) for Octopicola huanghaiensis n. sp. is urn: lsid:zoobank.org:act:FD10ADE0-1561-496D-BA2E-82269D62F7C3.

Table 1 Sampling information and infection levels of Octopicola huanghaiensis n. sp. in two octopus species from the Yellow Sea(off Qingdao, Shandong Province, China)

| Hosts | Amphioctopus fangsiao | | Octopus minor | |
|--------------------------------------|------------------------------|---|------------------------------|--|
| Sampling date | 10 October 2016 ^a | 30 April 2018 | 10 October 2016 ^a | |
| No. of hosts examined | 10 (sex not recorded) | $4^{\circ}_{+} + 1_{\circ}^{\circ}_{-}$ | 9 (sex not recorded) | |
| Host mantle length ^b (cm) | 5.3 (4.5-6.0) | 7.1 (6.5–8.1) | 6.1 (5.4–6.5) | |
| Host total length ^b (cm) | 18.1 (16.5–21.0) | 21.7 (18.3–24.4) | 38.1 (33.3-48.5) | |
| Host weight ^b (g) | not recorded | 93.0 (71.0-122.9) | not recorded | |
| Prevalence (%) | 30.0 | 100.0 | 22.2 | |
| Intensity ^b | 1.3 (1–2) | 7.8 (2–13) | 1.0 (1, 1) | |

^aHosts of this batch were washed with filtered seawater before examining copepod parasites; ^bMean (Range)

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Etymology: The specific name '*huanghaiensis*' refers to the Chinese name (Huanghai) of the type-locality (Yellow Sea).

Description (Figs. 1–3)

Female [Based on 12 adult specimens; Figs. 1A, B, D, E; 2A–G; 3A–D.] Body (Fig. 1A) elongated and slender, 1.73–2.21 (2.03) mm long (not including setae on caudal rami), with maximum width (at level of first pedigerous segment) 0.39–0.57 (0.47) mm. Posterior and ventral edges of epimera of prosome segments each with hyaline, striated membrane. Urosome $c.1.08 \times$ longer than prosome. Dorsal surface of both prosome and urosome with scattered hairs.

Genital segment (Fig. 1A) $177-258 \times 247-319$ (228 × 293). Postgenital segments 4 (Fig. 1A), measuring (anterior to posterior): $112-144 \times 124-165$ (136 × 150); $107-145 \times 96-128$ (126 × 114); $94-113 \times 76-107$ (105 × 92); $84-113 \times 64-85$ (98 × 77).

Caudal ramus (Fig. 1B) 204–262 (236) long, 22–33 (27) wide (at middle); length $c.8.75 \times$ than width. Outer lateral seta 98–139 (121) long, haired outer side. Dorsal seta 31–54 (44) long. Outermost terminal seta 179–265 (223) long; innermost terminal seta 272–385 (327) long; both with lateral spinules. Median terminal seta 2, long, 360–493 (440) and 501–633 (555) in length respectively; both with lateral spinules.

Rostrum (Fig. 1D) bending ventrally, sharply pointed, and armed ventrally with 4 pairs of setules; front edge with 4 smooth and 2 branched setules. Antennule (Fig. 2A) 7-segmented. Formula of armature: 4, 13, 5, 3, 4 + 1 aesthete, 2 + 1 aesthete, and 7 + 11 aesthete. All setae naked. Third segment possessing 1 intercalary piece. Antenna (Fig. 2B) 4-segmented. First segment short and with 1 distal seta. Second segment very long, with 1 marginal seta located about midway on inner edge, and numerous short spinules on its surface. Third segment very short, with 1 spine and 2 setae. Fourth segment 43-55 (49) long, 22-33 (27) wide (length taken along inner margin, width taken at middle of segment), bearing 4 short smooth claws and 3 setae. Posterior margin of labrum (Fig. 1E) deeply incised and possessing 2 elongated flaps each forming 1 spike at tip. Inner margin of each flap possessing 1 membrane extending to spike and forming inner and outer margins of the latter.

Mandible (Fig. 2C) with 1 strongly sclerotised, pointed tooth, and 1 broad, pointed lobe bearing 1 row of spinules along one margin. Maxillule (Fig. 2D) possessing 1 small seta and forming terminally 2 elements each with tiny denticles along one side. Maxilla (Fig. 2E) 2-segmented; first segment with numerous minute spinules; second segment terminating in 1 toothed lash, bearing 1 smooth seta and 1 spine-like seta with spinules along one side. Maxilliped (Fig. 2G) 3-segmented; first segment unornamented; second segment bearing 2 small naked setae and numerous minute spinules; third segment small, with 2 spines and 1 small seta, all naked.

Legs 1–4 (Fig. 3A-D) with 3-segmented rami except 1-segmented leg 4 endopod. Armature patterns of legs as follows (numbers of spines shown by Roman numerals, those of setae by Arabic numerals):

| | Coxa | Basis | Exopod | Endopod |
|-------|------|-------|--------------------|-------------------|
| Leg 1 | 0-0 | 1-0 | I-0; I-1; II, I, 5 | 0-1; 0-1; I, 5 |
| Leg 2 | 0-0 | 1-0 | I-0; I-1; I, II, 6 | 0-1; 0-2; I, I, 4 |
| Leg 3 | 0-0 | 1-0 | I-0; I-1; I, II, 6 | 0-1; 0-2; I, I, 3 |
| Leg 4 | 0-0 | 1-0 | I-0; I-1; II, 6 | 3 |
| | | | | |

Leg 4 exopod (Fig. 3D) 173–237 (211) long. Leg 4 endopod 127–155 (140) long, 33–53 (44) wide, with 3 setae measuring (from outer to inner) 104–145 (125), 154–233 (202) and 258–317 (293) in length, respectively. Leg 5 (Fig. 2F) with 1 unornamented small subrectangular lobe, 39–53 (48) long and 25–33 (29) wide, bearing 2 unequal, naked setae. Leg 6 (Fig. 1A) consisting of 1 small naked seta near oviducal opening and 1 large naked seta.

Male [Based on 29 adult specimens; Figs 1C; 2H, I.] Body shape resembling that of female (Fig. 1C). Body length 1.13–1.43 (1.32 \pm 0.08) mm; greatest width 0.27–0.36 (0.31 \pm 0.02) mm. Urosome *c*.1.06× longer than prosome.

Genital segment (Fig. 1C) $126-188 \times 169-219$ (170 ± 15 × 190 ± 13). Postgenital segments 4, measuring (anterior to posterior): 76-100 × 78-98 (89 ± 7 × 85 ± 5); 60-79 × 65-83 (69 ± 5 × 74 ± 4); 48-63 × 53-66 (54 ± 4 × 61 ± 3); 51-66 × 47-60 (58 ± 4 × 54 ± 3).



Fig. 1 Octopicola huanghaiensis n. sp. A, Female, dorsal view; B, Female, caudal ramus, dorsal view; C, Male, dorsal view; D, Female, rostrum, ventral view; E, Female, labrum. *Scale-bars*: A, C, 100 µm; B, D, E, 50 µm

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Fig. 2 Octopicola huanghaiensis n. sp. A, Female, antennule; B, Female, antenna; C, Female, mandible; D, Female, maxillule; E, Female, maxilla; F, Female, leg 5; G, Female, maxilliped; H, Male, maxilliped; I, Male, leg 6. Scale-bars: A–C, E–I, 50 µm; D, 20 µm

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Fig. 3 Octopicola huanghaiensis n. sp. A, Female, leg 1; B, Female, leg 2; C, Female, leg 3; D, Female, leg 4. Scale-bars: 50 µm

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Rostrum, antennule, antenna, labrum, mandible, maxillule and maxilla as in female.

Maxilliped (Fig. 2H) 4-segmented; first segment unornamented; second segment bearing 2 naked setae and 2 rows of spinules; third segment small and unornamented; fourth segment bearing 1 claw possessing 2 setae and 1 small process at base.

Segmentation and armature of legs 1–4 similar to those in females. Leg 4 exopod 128–165 (146 ± 8) long. Leg 4 endopod 81–104 (95 ± 5) long, 28–38 (33 ± 2) wide, bearing 3 setae measuring (from outer to inner) 76–97 (87 ± 6), 114–162 (144 ± 10) and 136–190 (174 ± 13) in length, respectively. Leg 5 similar to that of female. Leg 6 (Fig. 2I) bearing 2 setae.

Discussion

To date only handful studies have been documented for Octopicolidae taxa; these described four species all assigned to the genus *Octopicola*, and recorded four (plus one unidentified) species of hosts (Cavaleiro et al., 2013). The present study extends the species number of *Octopicola* to five, adds two octopus species to the host list and extends the distribution of *Octopicola* spp. to North Pacific region.

Among the four Octopicola spp., O. stocki has two spines and one seta on the third antennal segment (figure 8 in Humes, 1963), which can be easily distinguished from the present species (one spine and two setae; Fig. 2B). Another distinct difference between these two species is that in O. stocki the second segment of maxilla is armed with two similarly-sized smooth setae (figure 12 in Humes, 1963), while in the new species one (of the two) seta is markedly larger and with a row of spinules along one side (Fig. 2E). Additionally, the outermost seta on the leg 4 endopod of the male in O. stocki is distinctly spiniform, sinuous and armed with prominent lateral spinules (figure 29 in Humes, 1963) (vs not spiniform, not sinuous and unarmed with lateral spinules; Fig. 3D).

Octopicola regalis differs from the new species by its third antennal segment bearing a claw-like jointed spine, a small naked seta and a blunt spine with rows of long hairs along inner margin (figure 10 in Humes, 1974) (vs 1 smooth spine and 2 smooth setae in the new species; Fig. 2B), the inner surface of the second segment of male maxilliped bears three rows (figure 26 in Humes, 1974) (*vs* two rows; Fig. 2H) of spinules.

Octopicola antillensis was originally described as a subspecies, Octopicola superba antillensis Stock et al., 1963, but was recently elevated to species level (Cavaleiro et al., 2013). In this species, the third segment of the antenna bears a prominent projection densely covered with long spinules (figure 2a in Stock et al., 1963) (vs absent in the new species; Fig. 2B); the posterior seta on genital segment of both males and females extends only slightly beyond the posterior margin of its own somite (figure 1b, d in Stock et al., 1963) (vs extending beyond the posterior margin of next urosomal somite in present species; Fig. 1A, C); the inner surface of the second segment of male maxilliped possesses three rows plus clusters of spinules (figure 2d in Stock et al., 1963) (vs two rows of spinules in the new species; Fig. 2H).

Octopicola huanghaiensis n. sp. is most similar to O. superba in morphology. However, the third antennal segment of O. superba has a finely denticulated triangular process (figure 3 in Bocquet & Stock, 1960), while that of the new species lacks process (Fig. 2B). The fourth antennal segment in female O. superba is nearly two times longer than that in female O. huanghaiensis n. sp. (94 vs 49 µm). The two species also differ in that the male of O. superba is similar to the female in size (mean body length 1.9 mm for males and 1.8 mm for females), while in the present species the male is considerably smaller than the female (mean body length for males 1.3 mm vs 2.0 mm for females). Moreover, the posterior margin of the labrum of the new species has 2 elongated flaps each forming a spike at the tip (Fig. 1E), which has not been reported for the four congeners known to date.

The above comparisons, clearly indicate that the present specimens represent a new species. *Octopicola huanghaiensis* n. sp. is the first species of *Octopicola* reported from *A. fangsiao* and *O. minor* and is the only species of the family Octopicolidae known from the North Pacific waters. Among the \sim 300 species of octopuses (Octopoda) (see Allcock et al., 2014), only a few have been examined for the presence of octopicolids may be discovered by future intensive surveys, as commented by Cavaleiro et al. (2013).

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All applicable institutional, national and international guidelines for the care and use of animals were followed.

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