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Notes on Early Development and Secondary Sexual Characteristics of the Mesopelagic Amphipod *Cyphocaris challengeri* (Gammaridea ; Lysianassidae)

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Abstract

Early development and expression of secondary sexual characteristics of the mesopelagic gammarid amphipod *Cyphocaris challngeri* from the Oyashio region are described and illustrated. The embryonic development pattern of *C. challengeri* is similar to that being reported for other amphipods. A marked morphological character of this species (e.g. "hump-head") becomes evident at Instar 4. Juveniles in females' marsupia are released into the surrounding water at Instar 5. External sexual characters (oostegites for females, genital papillae for males) develop from Instar 8. As mature characters, carrying eggs and juveniles in marsupia for females and presence of calceoli (possible pheromone receptor) on the antennal flagella for males are found for the specimens at Instars 10-13 and at Instars 11-12, respectively.

Key words : Mesopelagic, Amphipod, Early develoment, Secondary sexual characteristics, *Cyphocaris challengeri*

Amphipods are a common component of marine zooplankton communities throughout the world, though their contribution to the zooplankton biomass is usually low (cf. Raymont, 1983). There are two suborders (Hyperiidea and Gammaridea) of pelagic amphipods ; species in the former are exclusively pelagic, and the latter contains benthic and bentho-pelagic species, with a few pelagic forms (Vinogradov, 1970 ; Raymont, 1983). Pelagic hyperiids (such as genus *Themisto*) are numerous in the upper layers of higher latitude seas (Shih, 1982), and pelagic gammarids are largely deep-sea inhabitants (Vinogradov, 1970 ; Raymont, 1983).

The pelagic gammarid amphipod *Cyphocaris challengeri* is distributed worldwide (Birstein and Vinogradov, 1955 ; Bowman and McCain, 1967). It is the most abundant gammarid amphipod in the mesopelagic zone of the subarctic Pacific (Yoo, 1970 ; Birshtein and Vinogradov, 1972) and an important predator on the eggs of walleye pollock in the western Gulf of Alaska (Bailey, et al., 1993 ; Brodeur and Merati, 1993). Despite the possible importance of this species in pelagic food webs, little information is available on its development and maturation processes (cf. Yamada and Ikeda, 2000).

For gammarid amphipods, a marked external (= secondary) sexual character of females is the development of oostegites, four pairs of each arising from the posterior inner margin of the coxa of peraeopods 2-5 to form jointly a chamber (=marsupium). The function of the chamber is for the retention of eggs or newly hatched juveniles by females (Bousfield, 1973 ; Lincoln, 1979). The external sexual characters of male gammarids are genital papillae (penis papillae) and calceoli, and the latter is considered to act as a pheromone receptor (Dahl, et al., 1970). In addition to genital papillae, male characters are also expressed in the morphology of the gnathopod in some species (Charniaux-Cotton and Payen, 1985).

Previously, the presence of oostegites and genital papillae were used as diagnostic features of females and males, respectively, of *Cyphocaris challengeri* by Thurston (1976). Unfortunately, Thurston's (1976) brief description (without illustrations) for juveniles, mature males and mature females is of limited use for the evaluation of the biology of *C. challengeri*. In the present study, we describe and illustrate the development of oostegites and genital papillae in *C. challengeri* based on the instar number system established by Yamada and Ikeda (2000) for this species. Developmental features of eggs and early juvenile instars are also provided.

Cyphocaris challengeri specimens were collected from 200-400 m depth at a station off Cape Esan, southwest Hokkaido (42°N, 141°30′E) from May 1997 to April 1998, and preserved in 5% buffered formalin seawater. Eggs and larvae in the females' marsupia were removed for observations under a dissecting microscope fitted with a Nikon Digital Camera E-910.

The eggs are violet in color and oval in shape (Fig.

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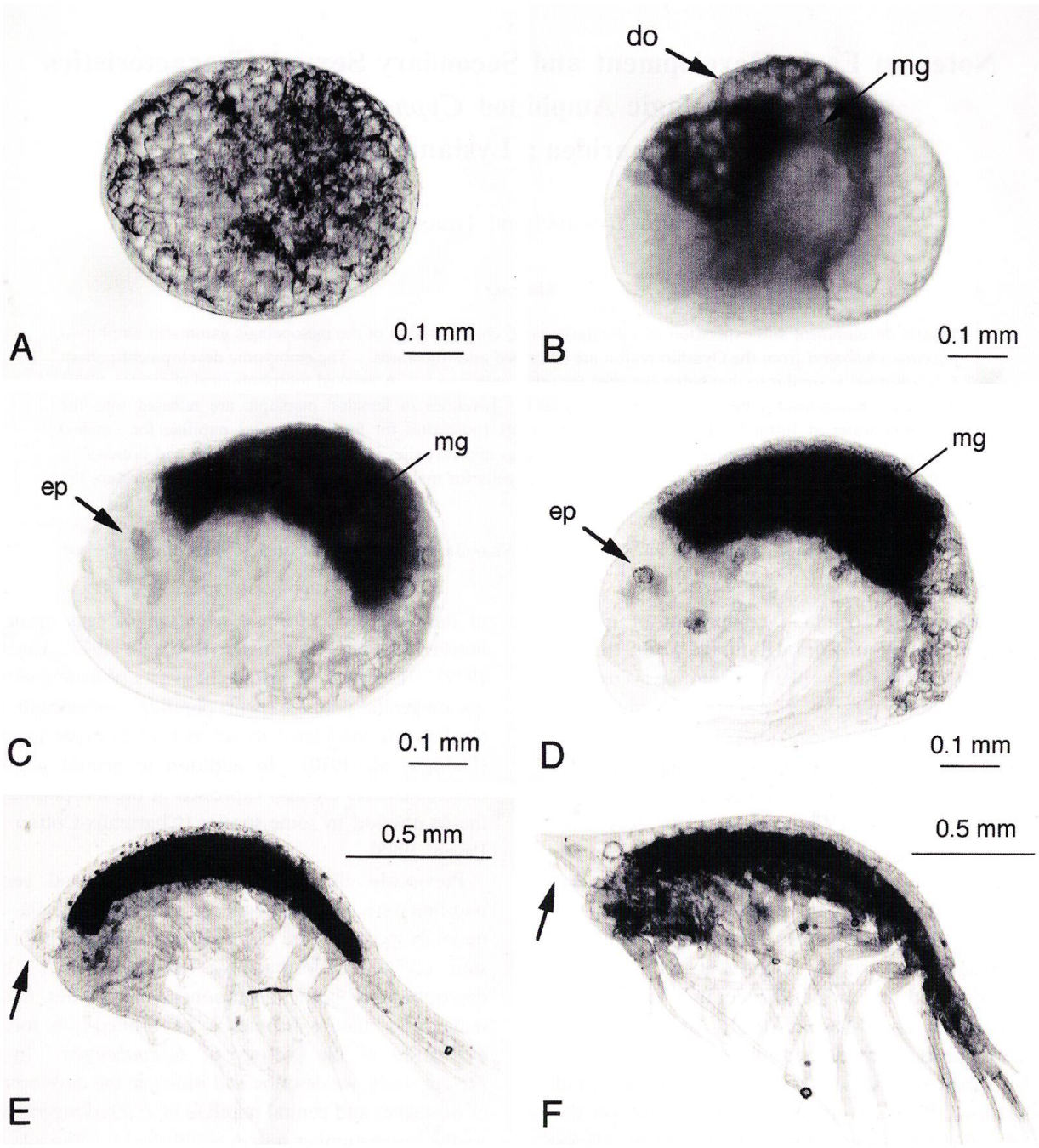

Fig. 1. *Cyphocaris challengeri.* (A) egg containing many oil globules. (B) Developing egg, in which dorsal organ (do) and rudiment of the mid-gut caeca (mg) formed. (C) Further developing egg in which eye pigment is visible and some appendage rudiments formed. (D) Newly hatched juvenile (Instar 1). (E) Instar 4 juvenile. The anterior portion of the pereonite 1 produces is blunt (arrowed). (F) Instar 5 juvenile (the first free-swimming instar). The anterior portion of the pereonite 1 extends more sharply (arrowed).

1A). Long and short diameters of the eggs are 0.63 and 0.47 mm, respectively. Eggs are covered with a thin membrane and contain a large amount of oil globules of varying sizes. As with development of the eggs proceeds, the 'dorsal organ' develops at an early stage (Fig. 1B). This organ occurs during the development of many malacostracans and is lost prior to hatching (Sheader and Chia, 1970; Sheader, 1977). Formation of the mid-gut caeca (Fish, 1975) takes place. Then, body segments are formed and appendage rudiments (antennae, peraeopods, pleopods and uropods) are visible through the thin egg membrane (Fig. 1C). Red eye pigment appears at this stage. The development of the mid-gut caeca continues.

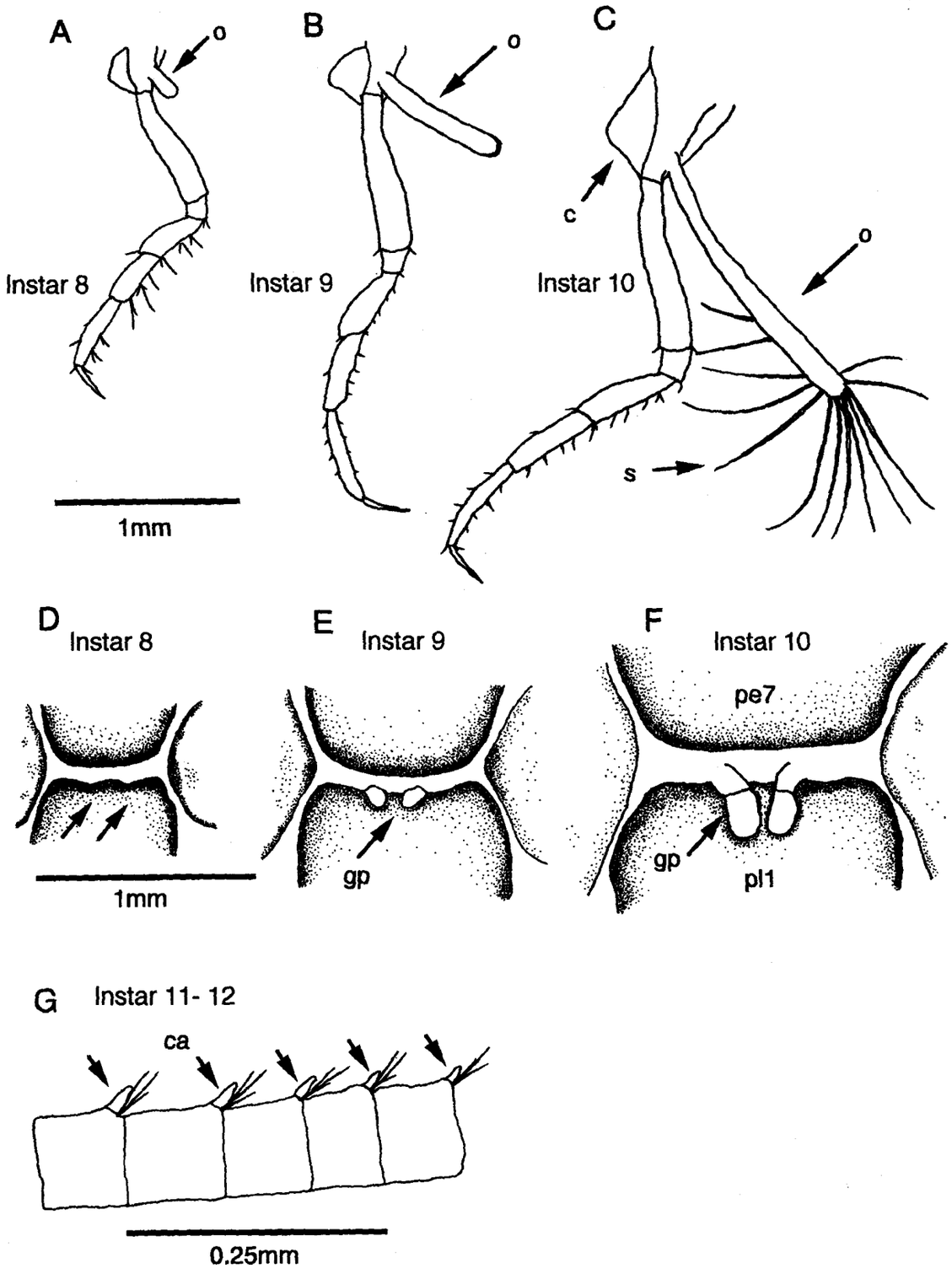

Fig. 2. *Cyphocaris challengeri.* (A)-(C) Serial development of oostegite (o) on the coxa (c) of right peraeopod 1 of an Instar 8, Instar 9 and 10 female. Oostegite of fully mature female bears long setae (s). (D)-(F) Serial development of genital papillae (gp) on the ventral surface between pereonite 7 (pe7) and pleonite 1 (pl1) on an Instar 8, Instar 9 and Instar 10 male. (G) Calceoli (ca) on the antennal flagellum of mature male.

The body of newly hatched juveniles (or Instar 1) is curled strongly (Fig. 1D), and the extended body length (from the base of the first antenna to the distal end of the telson) is about 1.50 mm. Instar 1 has a complete sets of appendage rudiments, i.e., 2 antennae, 7 peraeopods, 3 pleopods and 3 uropods. Each peraeopod is composed of 7 segments, which is the same as for adult specimens. The number of segments of pleopod rami of Instar 1, which can be used as a marker of instar number (cf. Yamada and Ikeda, 2000), is one. After the publication of Yamada and Ikeda (2000), we noticed that we overlooked Instar 1, and the number of segments we counted was in fact "the number of joints". Therefore, both instar number and the number of segments for Instars 1-17 given by Yamada and Ikeda (2000) should plus one for each, adding a new Instar 1 with one segment. Instar 1 molts to Instar 2, in which the body extends slightly with the further development progression of each appendage. The number of segments of pleopod rami increases to two. At Instar 4 the body extends almost fully (Fig. 1E), and the development of appendages is nearly complete. The anterior portion of the pereonite 1 projects bluntly (lateral view). Instar 4 is the last developmental stage in the female's marsupium, and they are released into the surrounding water and molt to Instar 5 as the first free-swimming instar. The anterior portion of the pereonite 1 projects sharply (Fig. 1F), a marked morphological character of *Cyphocaris challengeri* (Cyphocaris="hump-head" (Bowman and McCain, 1967)). No external sexual characters are evident yet for free-living Instars 5 through 7.

External sexual characters develop from Instar 8. As the female character, oostegites occur from Instar 8. Oostegites are small at Instar 8 females (Fig. 2A), and increase in size at Instar 9, but are still not fully developed yet (Fig. 2B). As a male character, genital papillae, arising from the ventral sternite in the segment 7, appear as very small paired-processes at Instar 8 (Fig. 2D), protrude slightly at Instar 9 (Fig. 2E), and extensively and flex each other at Instar 10 (Fig. 2F). Instar 8 and Instar 9 females and Instar 8 to Instar 10 males are designated as 'immature' adults (Yamada and Ikeda, 2000).

At Instar 10, oostegites project further and bear 13 interlocking setae at the posterior margin (Fig. 2C). The setae slip out when juveniles in the female's marsupium are released into the surrounding water. Ovigerous females are seen from Instars 10 through 13. The genital papillae of males develop no further after Instar 10. Instead, a small bud-shaped sense organ called the "calceoli" appears on each segment of the antennal flagellae at Instars 11 and 12 (Fig. 2G). Instar 10 to Instar 13 females and Instar 11 and Instar 12 males are designated as 'mature' adults (Yamada and Ikeda, 2000).

Compared with the planktonic hyperiid amphipods *Themisto gaudichaudii* (Kane, 1963) and *T. compressa* (formally *T. gaudichaudii*; Sheader, 1977), the results of our observations on *Cyphocaris challengeri* appear to be almost identical in egg development patterns, morphology of newly hatched juveniles in females' marsupia, the juvenile instar number leaving from the marsupia, and development patterns of oostegites in females (although oostegites of *Themisto* spp. lack interlocking setae). A large difference between *Themisto* spp. and *Cyphocaris challengeri* is the expression of male's secondary sexual characters. For *Themisto* spp., male characters appear as a long, multi-articulated second antenna and an 'excavated' organ developing opposite to one another on the inner margin of the rami of the first pair of urosomes; both of these characters are easily observable under a microscope. Neither marked male characters of *Themisto* spp. are developed in *C. challengeri*, making identification of males of this species rather difficult. While the present observations were based on preserved specimens, more detail study on live specimens such as the studies on *Themisto compressa* by Sheader (1977) is needed to deepen our understanding about mating behavior, embryonic development time, hatching success, etc., of *C. challengeri*.

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References

Bailey, K.M., Brodeur, R.D., Merati, N. and Yoklavich, M.M. (1993). Predation on walleye pollock (*Theragra chalcogramma*) eggs and yolk-sac lavae by pelagic crustacean invertebrate in the western Gulf of Alaska. *Fish. Oceanogr.*, **2**, 30-39.

Birshtein, Y.A. and Vinogradov, M.E. (1972). On the fauna of pelagic gammarids in the Kurile-Kamchatka area of the Pacific Ocean. p. 419-438, Bogorav, V.G. (ed). *Fauna of the Kurile-Kamchatka Trech and its environment*, Israel Program for Scientific Translations, Jelsalem.

Birstein, J.A. and Vinogradov, M.E. (1955). Pelagic gammarids (Amphipoda-Gammaridea) of the Kurile-Kamchatka Trench. *Trudy. Inst. Okeano. Akad. Nauk. SSSR*, **12**, 210-287.

Bousfield, E.L. (1973). *Shallow-water gammaridean amphipoda of New England*. Cornell University Press, Ithaka & London.

Bowman, T.E. and McCain, J.C. (1967). Variation and distribution of the pelagic amphipod *Cyphocaris challen-*

geri in the Northeast Pacific (Gammaridea : Lysianassidae). *Proc. U.S. Nat. Mus.,* **122**, 1-14.

Brodeur, R.D. and Merati, N. (1993). Predation on walleye pollack (*Theragra chalcogramma*) eggs in the western Gulf of Alaska : the roles of vertebrate and invertebrate predators. *Mar. Biol.,* **117**, 483-493.

Charniaux-Cotton, H. and Payen, G. (1985). Sexual differentiation. p. 217-299, Bliss, D.E. and Mantel, L.H. (eds.). *The Biology of Crustacea 9. Integument, pigments, and hormonal processes*, Academic Press, Inc., New York.

Dahl, E., Emanuelsson, H. and Mecklenburg, C.V. (1970). Pheromone reception in the males of the amphipod *Gammarus duebeni* Lilljeborg. *Oikos,* **21**, 42-47.

Fish, J.D. (1975). Development, hatching and brood size in *Bathyporeia pilosa* and *B. pelagica. J. Mar. Biol. Ass. U.K.,* **55**, 357-368.

Kane, J.E. (1963). Stages in the early development of *Parathemisto gaudichaudii* (Guér.) (Crustacea Amphipoda : Hyperiidea), the development of secondary sexual characters and of the ovary. *Trans. Royal Soc. New Zealand,* **3**, 35-45.

Lincoln, R.J. (1979). *British marine amphipoda : Gammaridea.* British Museum (Natural History), London.

Raymont, J.E.G. (1983). *Plankton and productivity in the oceans 2nd edition.* Pergamon Press, Oxford.

Sheader, M. (1977). Breeding and marsupial development in laboratory-maintained *Parathemisto gaudichadi* (Amphipoda). *J. Mar. Biol. Ass. U.K.,* **57**, 943-954.

Sheader, M. and Chia, F.S. (1970). Development, fecundity and brooding behaviour of the amphipod, *Marinogammarus obtusatus. J. Mar. Biol. Ass. U.K.,* **50**, 1079-1099.

Shih, C.T. (1982). Hyperiidea. p. 285-292, Parker, S.P. (ed). *Synopsis and classification of living organisms, 2*, McGraw-Hill, New York.

Thurston, M.H. (1976). The vertical distribution and diurnal migration of the crustacea amphipoda collected during the Sond Cruise, 1965 I. The gammaridea. *J. Mar. Biol. Ass. U.K.,* **56**, 359-382.

Vinogradov, M.E. (1970). *Vertical distribution of the oceanic zooplankton.* Israel Program for Scientific Translations, Jelsalem.

Yamada, Y. and Ikeda, T. (2000). Development, maturation, brood size and generation length of the mesopelagic amphipod *Cyphocaris challengeri* (Gammaridea ; Lysianassidae) off southwest Hokkaido, Japan. *Mar. Biol.,* **137**, 933-942.

Yoo, K.I. (1970). Ecological studies on pelagic amphipods in the western North Pacific. Ph. D Thesis. Tokyo University.