

A New Species of the Genus *Pseudodiaptomus* (Copepoda, Calanoida) from Korean Estuarine Waters with Remarks on its Molecular Variation

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Introduction

Recently many studies show that genetic characters can be used to provide unambiguous taxonomic discrimination when morphological characters are not practical for routine identification to species level (Bucklin et al., 1996, 1998; Lindeque et al., 1999). The internal transcribed spacer (ITS) regions of the nuclear ribosomal DNA are also one of appropriate markers for species-level studies because it contains sufficient diversity to address intra- and interspecific phylogenetic relationships in invertebrates (Odorico and Miller, 1997; Schizas et al., 1999).

Based on the morphology of its habitus and genital area, we divide a species previously known as *Pseudodiaptomus inopinus* in Korean waters into two species, *P. inopinus* and *P. koreanus* n. sp.. In this study, using rDNA ITS we address the genetic differentiation between their species.

Methods and Materials

Materials examined: In Mangyeong and Seomjin River Estuaries zooplankton collections were monthly made from January to December 2000. All pseudodiaptomids were sorted from the samples. For morphological taxonomy pseudodiaptomids were dissected and mounted on polyvinyl lactophenol. The bodies and appendages were observed with a differential interference contrast microscope (Zeiss SV6) equipped with a drawing tube. The female genital structures of *Pseudodiaptomus* species were examined with a scanning electron microscope (Hitachi S-4700). The morphological terminology follows Huys and Boxshall (1991).

Sequencing of rDNA ITS regions: Plasmids containing the rDNA ITS fragments were isolated by using a QIAquick plasmid minikit (Qiagene

GmbH). Purified plasmids were manually sequenced by using a T7 sequencing kit (Pharmacia Biotech, Inc., Piscataway, N.J.) and the fmol DNA sequencing system (Promega Co.).

Results and Abstract

In Korean waters *Pseudodiaptomus koreanus* n. sp. has been identified as *P. inopinus* (Burckhardt, 1913) because of morphological similarities in their habitus and fifth legs of both sexes. However, the morphology of the female genital area distinguishes *P. koreanus* from *P. inopinus*: in the former processes of the genital flaps are wider and duller than those in the latter. The morphological differences of the female genital area within pseudodiaptomid species are species-specific. This is also supported by the ITS sequence differences ranging from 5.8% (between *P. koreanus* from Seojin River and *P. inopinus*) to 6.7% (between *P. koreanus* from Hyeongsan River and *P. inopinus*). On the other hand, in the male fifth legs of *P. koreanus* two types, finger-like form and shoe-like one, are found as in other estuarine copepods *P. inopinus* and *P. poplesia*. Although the morphological differences could be important enough to separate two forms into each of an independent species, it is considered as an intraspecific variability because of the similarities of other morphological characters and sympatric occurrence. ITS sequence analyses for solving the taxonomic problem indicate that the sequence difference between two types is <1.5%, suggesting that two types are genetically identical species.

References

- Bucklin, A., T.C. LaJeunesse, E. Curry, J. Wallinga and K. Garrison. 1996. Molecular diversity of the copepod, *Nannocalanus minor*: Genetic evidence of species and population structure in the North Atlantic Ocean. *J. Mar. Res.*, 54, 285-310.
- Bucklin, A., A.M. Bentley and S.P. Franzen. 1998. Distribution and relative abundance of *Pseudocalanus moultoni* and *P. newmani* (Copepoda: Calanoida) on Georges Bank using molecular identification of sibling species. *Mar. Biol.*, 132, 97-106.
- Lindeque, P.K., R.P. Harris, M.B. Jones and G.R. Smerdon. 1999. Simple molecular method to distinguish the identity of *Calanus* species (Copepoda: Calanoida) at any developmental stage. *Mar. Biol.*, 133, 91-96.
- Odorico, D.M. and D.J. Miller. 1997. Variation in the Ribosomal Internal Transcribed Spacers and 5.8S rDNA among five species of *Acropora* (Cnidaria; Scleractinia): Patterns of variation consistent with reticulate evolution. *Mol. Biol. Evol.*, 14, 465-473.
- Schizas, N.V., G.T. Street, B.C. Coull, G.T. Chandler and J.M. Quattro. 1999. Molecular population structure of the marine benthic copepod *Microarthridion littorale* along the southeastern and Gulf coasts of the USA. *Mar. Biol.*, 135, 399-405.