

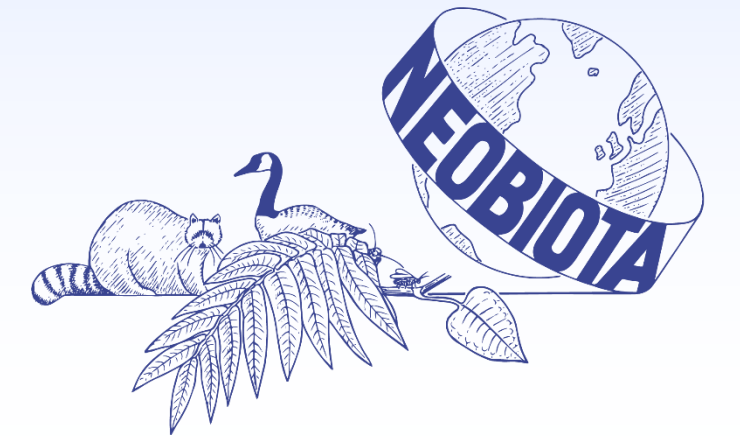
# Spreading of non-indigenous Indo-Pacific copepod *Pseudodiaptomus marinus* Sato, 1913 in eastern Adriatic coastal and transitional waters

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## Introduction

Indo-Pacific calanoid copepod *Pseudodiaptomus marinus* Sato, 1913 is spreading through ballast water release in port areas worldwide. From its native habitat in Japanese waters of the Northwestern Pacific, this passive way of transport has enabled its colonization of the western Atlantic coast and several European seas (Brylinski et al., 2012; Sabia et al., 2015) and *P. marinus* is now spreading rapidly through the Mediterranean coastal waters (Uttieri et al., 2020). The first records of *P. marinus* in the Adriatic Sea were confined to its northern part (Bay of Trieste, port of Koper, Venice lagoon) (de Olazabal and Tirelli, 2011; Lučić et al., 2015; Vidjak et al., 2019). However, in the last 5-year period we recorded its presence further to the south, at three new locations along the eastern coast closely related to port activities. Here we present the abundance data and morphological details of *P. marinus* specimens from coastal and transitional waters of the Croatian Adriatic.

## Material and methods

*P. marinus* individuals were isolated from zooplankton samples collected with vertically hauled plankton nets (53 and 125 µm mesh sizes) during night (after 20:00), early morning or late afternoon, when *P. marinus* migrates into the water column, since it is predominantly demersal during the day. We confirmed the species' identity through morphological identification using light and FE-SEM microscopy (MIRA3, TESCAN) and through mtCOI DNA sequencing (Folmer et al., 1994). Taxonomic drawings of habitus and body parts were made with a drawing tube mounted on BH2 OLYMPUS microscope, under 100x and 200x magnifications. Local zooplankton community was analyzed under inverted microscope and the abundances expressed as no. ind. m<sup>-3</sup>.

## Results and discussion

### Morphological characteristics:

The species' identity was unequivocally confirmed through DNA barcoding. The main morphological characteristics of *P. marinus* specimens of both sexes from the Šibenik port (Figure 1) were in general agreement with earlier descriptions (Grindley and Grice, 1969; Brylinski et al., 2012), regarding body segmentation and total body size range (female 1.37 mm; male 1.12 mm). Minor differences were observed in the minor armature elements of male fifth legs (Figures 2 and 3), interpreted in literature as geographical variations in individuals from different parts of the Japanese region, or ecophenotypes (Grindley and Grice, 1969; Brylinski et al., 2012).

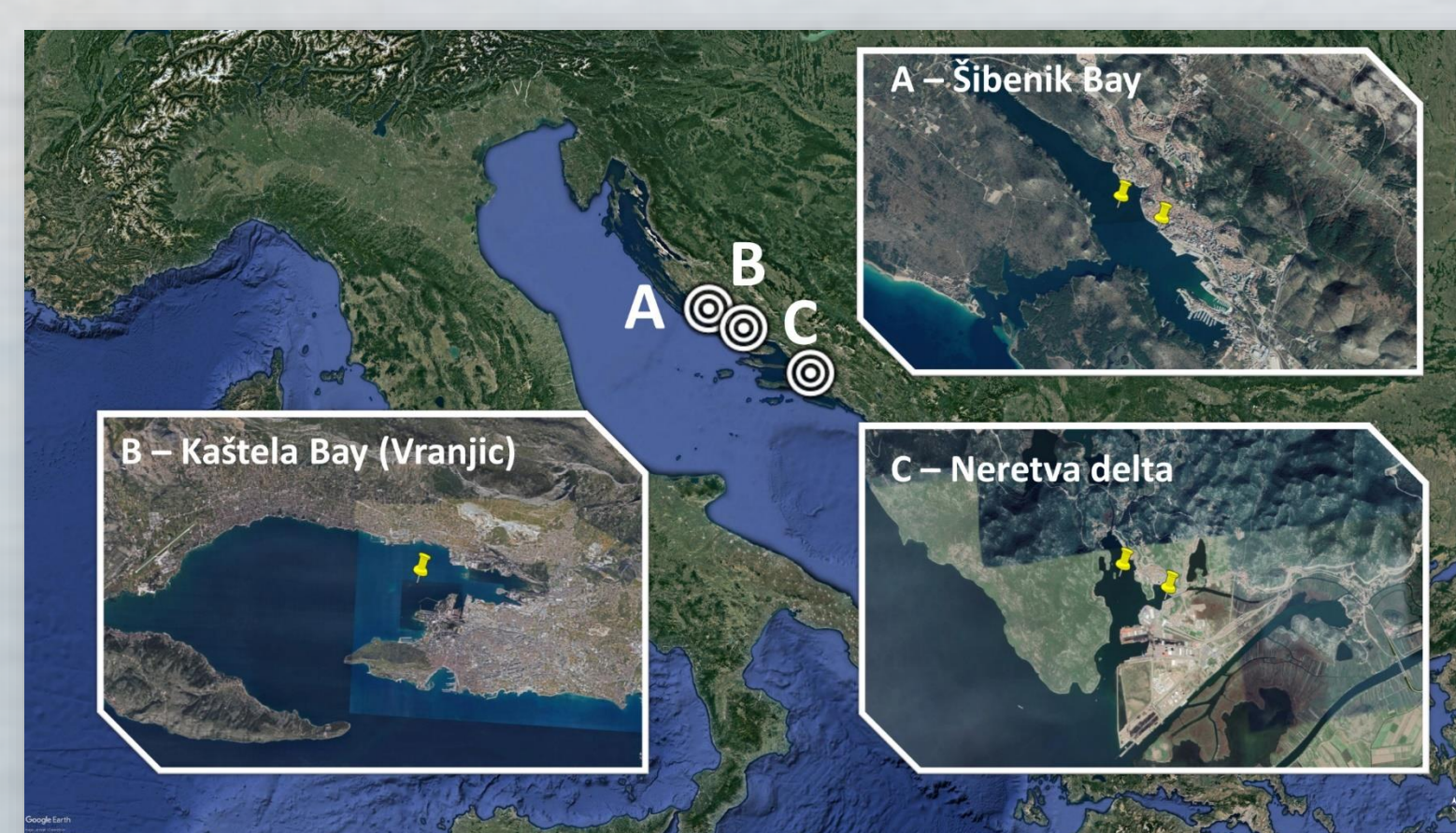


Figure 1. Study area with the sampling stations.

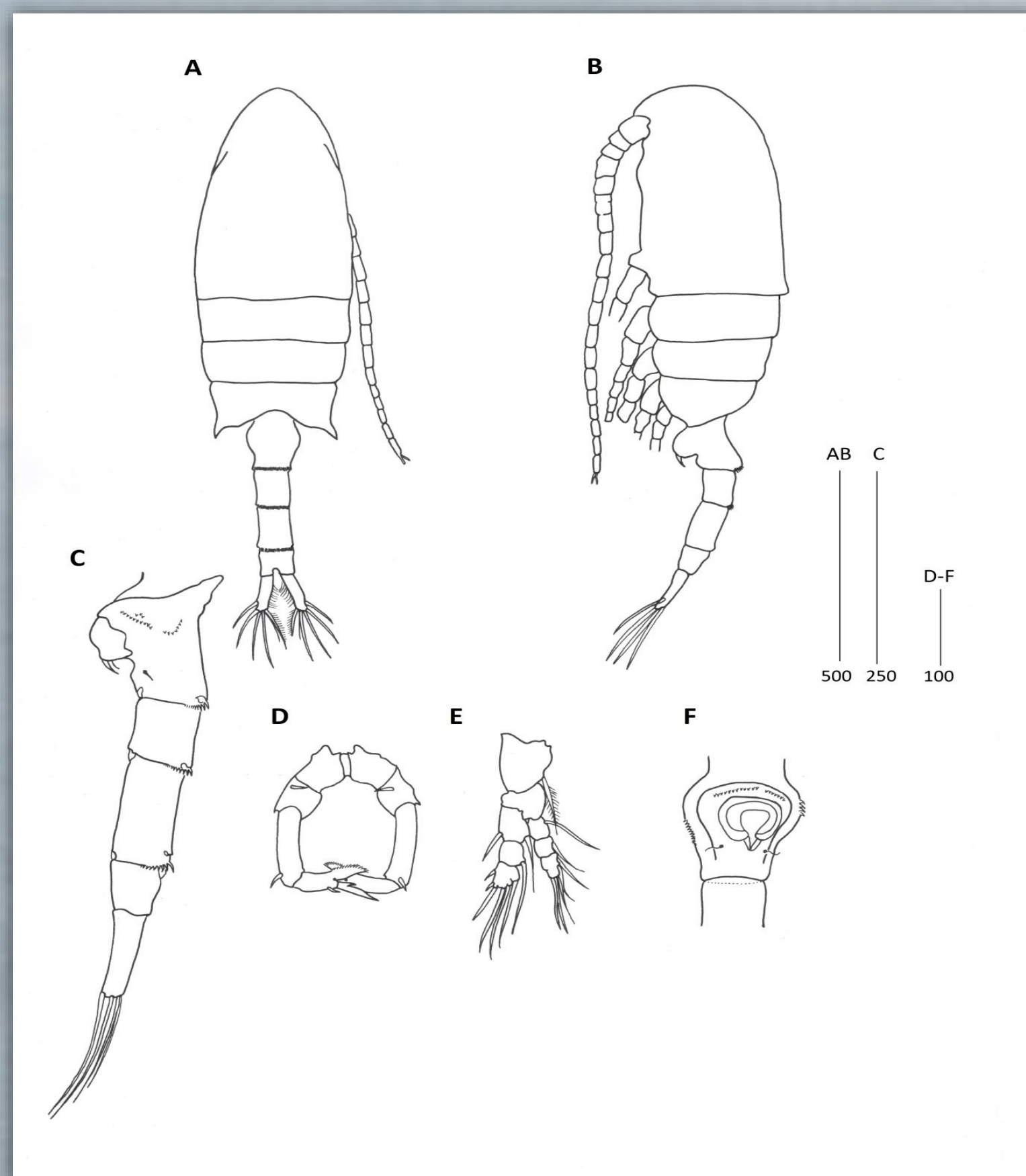


Figure 2.a *Pseudodiaptomus marinus*, adult female: A - habitus (dorsal); B - habitus (lateral) (P1-P4 not fully shown); C - female urosome in lateral view; D - female P5; E - female P1; F - female genital double somite in ventral view. All scales in micrometers.

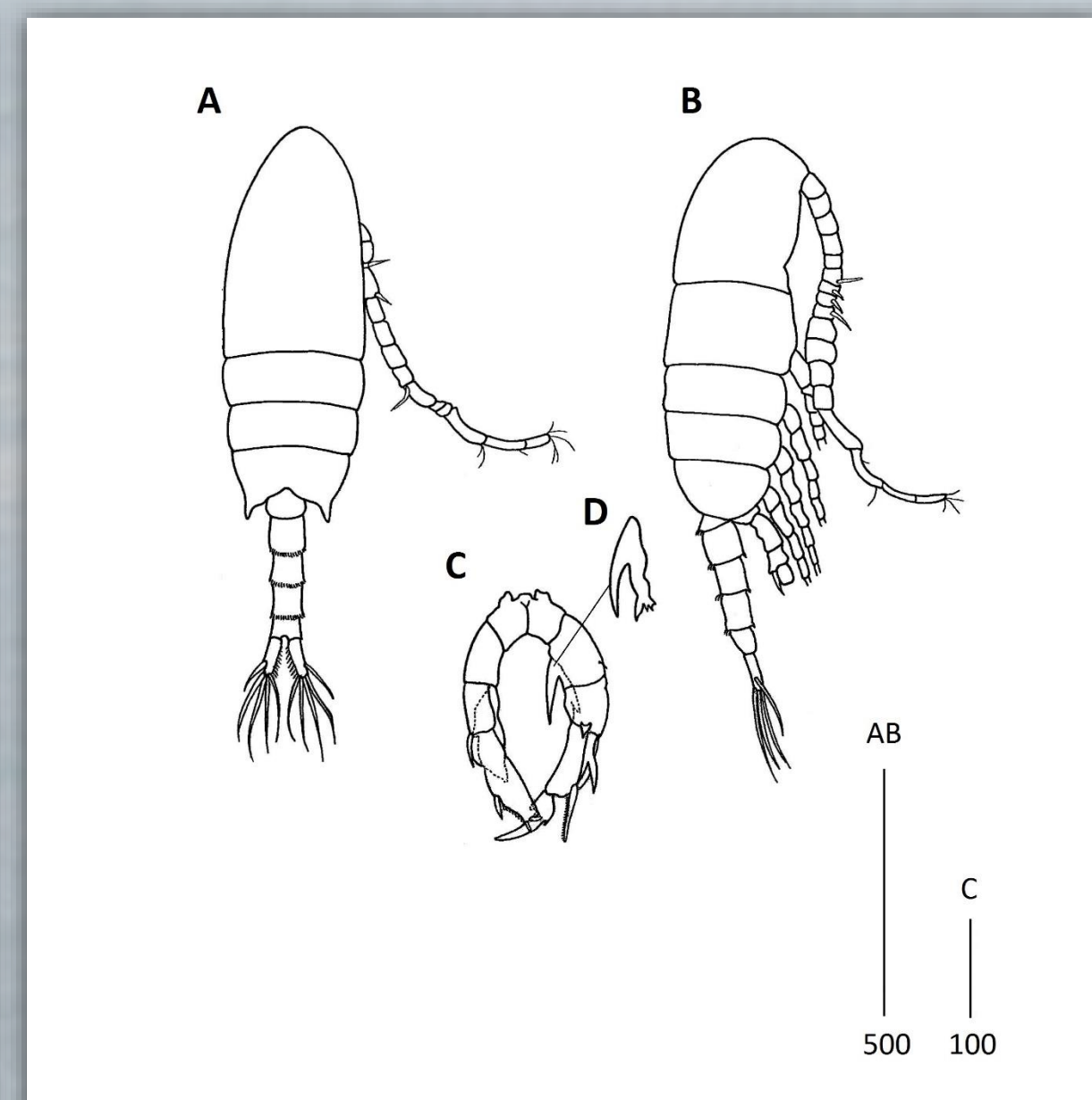


Figure 2.b *Pseudodiaptomus marinus*, adult male: A - habitus (dorsal); B - habitus (lateral) (P1-P4 not fully shown); C - male P5; D - detail of right P5 endopod. All scales in micrometers.

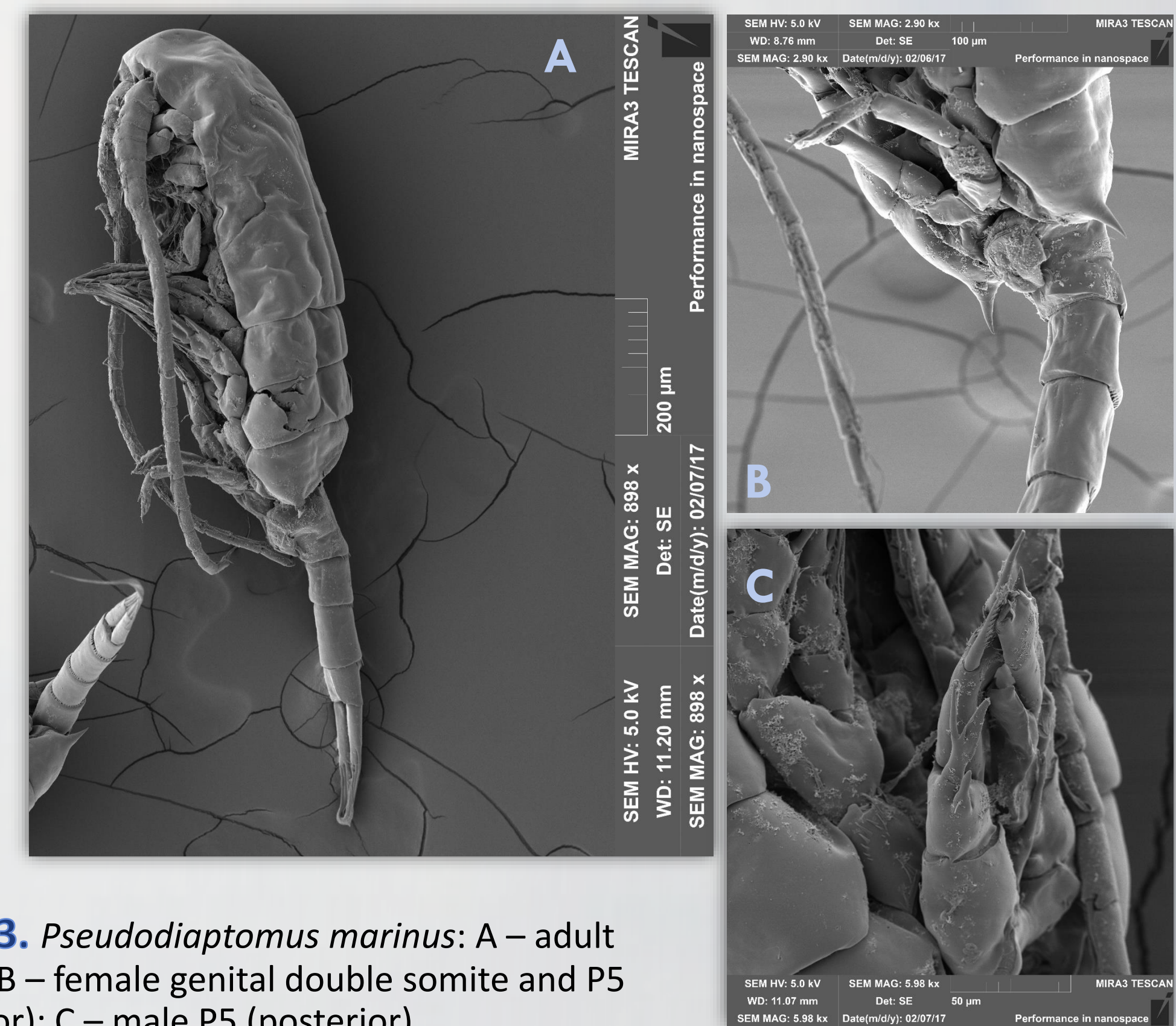


Figure 3. *Pseudodiaptomus marinus*: A - adult female; B - female genital double somite and P5 (posterior); C - male P5 (posterior).

### Distribution and abundance in Croatian waters:

**Port of Šibenik** (Figure 4): *P. marinus* has been present continuously from 2015, at anchoring site in the port (6-9 m), as well as in the central part of Šibenik Bay (38 m). **Neretva delta and port of Ploče** (Figure 4): *P. marinus* was recorded twice in the central part of the inner lagoon (11 m) and at anchoring site in the port (6 m). **Vranjic Bay** (Figure 4): few individuals of *P. marinus* were found during night sampling near the industrial North Port of Split (18 m). There was no evidence of changes in local planktonic copepod communities in any of the investigated areas, as their taxonomical compositions and relative abundances were consistent with the usual assemblages reported for eastern Adriatic shallow coastal embayments (Vidjak et al., 2012). The relative contribution of *P. marinus* to total copepods was >2%, while the abundances ranged between 0.1 and 59 ind. m<sup>-3</sup>. The presence of individuals of both sexes, ovigerous females and juveniles was recorded at all investigated sites, indicative of unimpeded reproduction and population establishment.

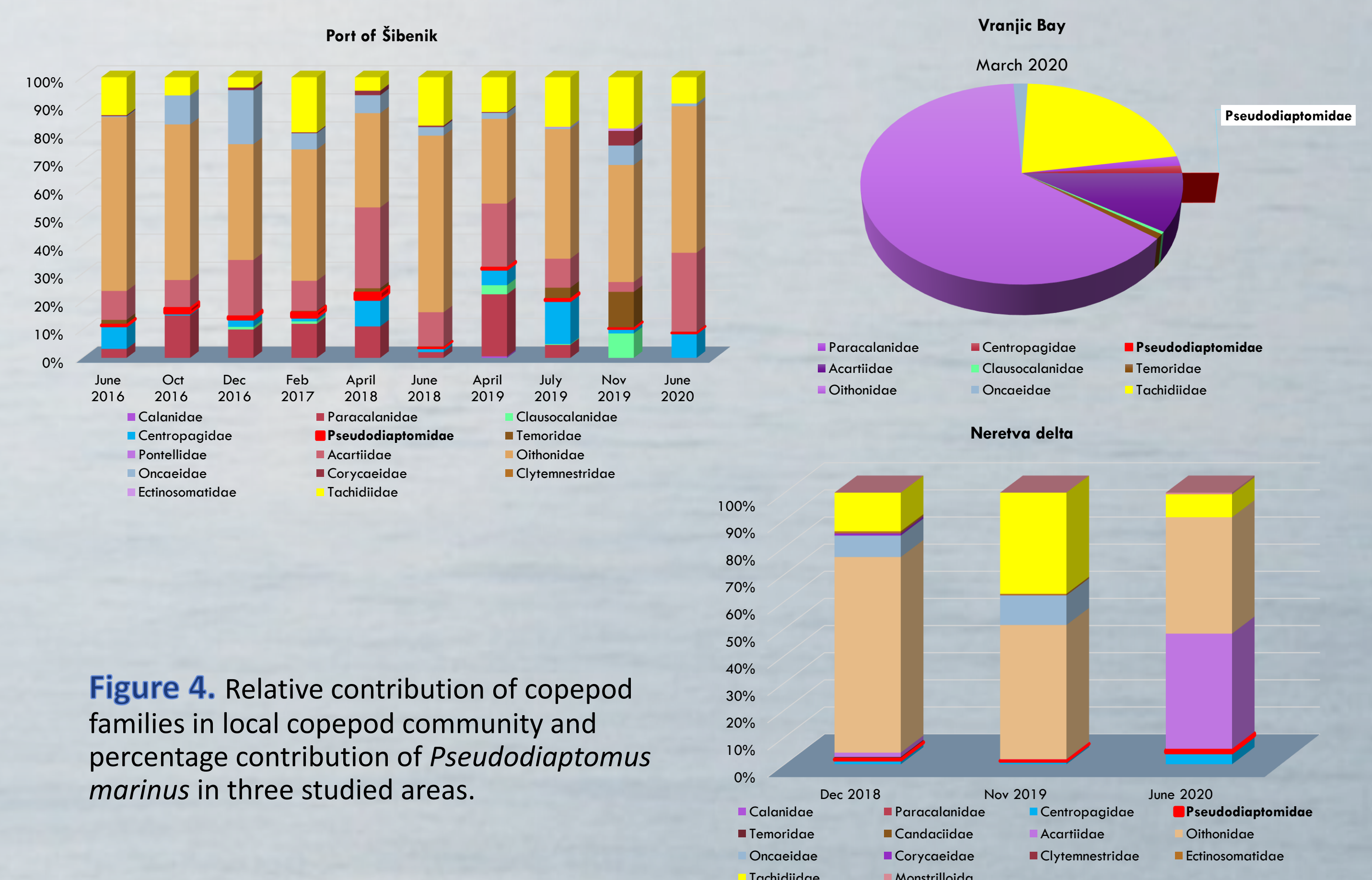


Figure 4. Relative contribution of copepod families in local copepod community and percentage contribution of *Pseudodiaptomus marinus* in three studied areas.

## Conclusion

So far, no adverse effects on local zooplankton were found on many sites of *P. marinus* worldwide colonization, including the Adriatic locations reported here. Low abundances that we recorded could be related to the limitations of sampling methodology, as the demersal phase of the species' life cycle remained unassessed. Nevertheless, high tolerance to physical environment and specific biological and behavioral traits of *P. marinus* (egg-carrying strategy in females, high plasticity in feeding mode and habitat occupation) could potentially increase its invasive behavior under right conditions. We are therefore advocating careful monitoring of the affected localities, improvement of sampling methodology and extension of targeted surveillance to other major Croatian ports.

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