

SHORT COMMUNICATION

Austral Summer Bloom of Loricated Choanoflagellates in the Central Ross Sea PolynyaLaura Escalera^a , Olga Mangoni^{b,c}, Francesco Bolinesi^{a,b} & Maria Saggiomo^a^a Research Infrastructures for Marine Biological Resources, Stazione Zoologica Anton Dohrn, Naples 80121, Italy^b Department of Biology, Università degli Studi di Napoli Federico II, Complesso Universitario di Monte Sant'Angelo, Naples 80126, Italy^c CoNISMa, Piazzale Flaminio 9, Rome 00196, Italy**Keywords**

Acanthoecidae; Antarctica; colonies; food web.

Correspondence

L. Escalera, Research Infrastructures for Marine Biological Resources, Stazione Zoologica Anton Dohrn, Villa Comunale Naples 80121, Italy

Telephone number: (+39)0815833240; FAX number: (+39)0815833360; e-mail: laura.escalera@szn.it

Received: 4 September 2018; revised 8 February 2019; accepted February 10, 2019.
Early View publication 9 March, 2019

doi:10.1111/jeu.12720

THE aim of this study was to provide insights into the diversity of choanoflagellates and abundance in the Ross Sea, particularly during an unusual bloom recorded in austral summer 2017. Acanthoecid choanoflagellates are small bacteriophagous cells surrounded by a delicate siliceous armour (lorica). They are common within Antarctic and Arctic plankton communities (Buck and Garrison 1983; Chen 1994; Fritsen et al. 2001; Marchant et al. 1987) but we have found no data available about the choanoflagellate concentrations in the western area of the Ross Sea.

MATERIALS AND METHODS

Plankton samples were collected in January 2017 as part of an oceanographic cruise in the western Ross Sea within the framework of the Plankton biodiversity and functioning of the Ross Sea Ecosystems in a changing southern oceans (P-ROSE) project. The 500 ml water samples were collected using Niskin bottles. Four depths from surface to 60 m at the 28 stations (Fig. S1), with two of the stations (15 and 57) sampled twice, were collected based on the vertical fluorescence profiles of chlorophyll *a*. Samples for

ABSTRACT

A bloom of loricated choanoflagellates was recorded for the first time in the Ross Sea polynya during the austral summer 2017. Both individual cells and uncommon large-size colonies (200 µm length) represent the 42–55% of the total plankton community (i.e. specimens from 5 to 150 µm length). Choanoflagellates serve as a link between low and mid trophic levels since they prey on bacteria and in turn are ingested by zooplankton. This twofold role and the unusual abundance recorded in the Antarctic ecosystem may have relevant but still unknown effects on food web structure and dynamics in that area.

taxonomic analysis, of specimens from 5 to 150 µm length, were preserved in 4% CaCO₃ buffered formalin solution. Depending on cell concentrations, 3–10 ml of sample was settled in Utermöhl-type chambers (Lund et al. 1958; Utermöhl 1931) for 24–48 h respectively. Cell counts were performed at 400X magnification using a Zeiss Ax10 Observerz.1 inverted light microscope equipped with an AxioCam MRc5 (Carl Zeiss Microscopy GmbH, Jena, Germany) image capture system. Up to four transects with an area of 0.78 mm² each of the settling chamber, were examined to reach at least 200 cells per sample which is considered statistically valid.

RESULTS AND DISCUSSION

During January 2017, five species of loricated choanoflagellates (Fig. 1) were identified: *Bicosta spinifera* (Fig. 1A), *Calliacantha* sp. (Fig. 1B), *Parvicorbicula socialis* (Fig. 1C, D), *Diaphanoeca multiannulata* (Fig. 1E, F) and *Crinolina aperta* (Fig. 1G–I). In agreement with the description by Throndsen (1970) for *B. spinifera*, two longitudinal costae that run slightly spiral at the cell body level were visible on

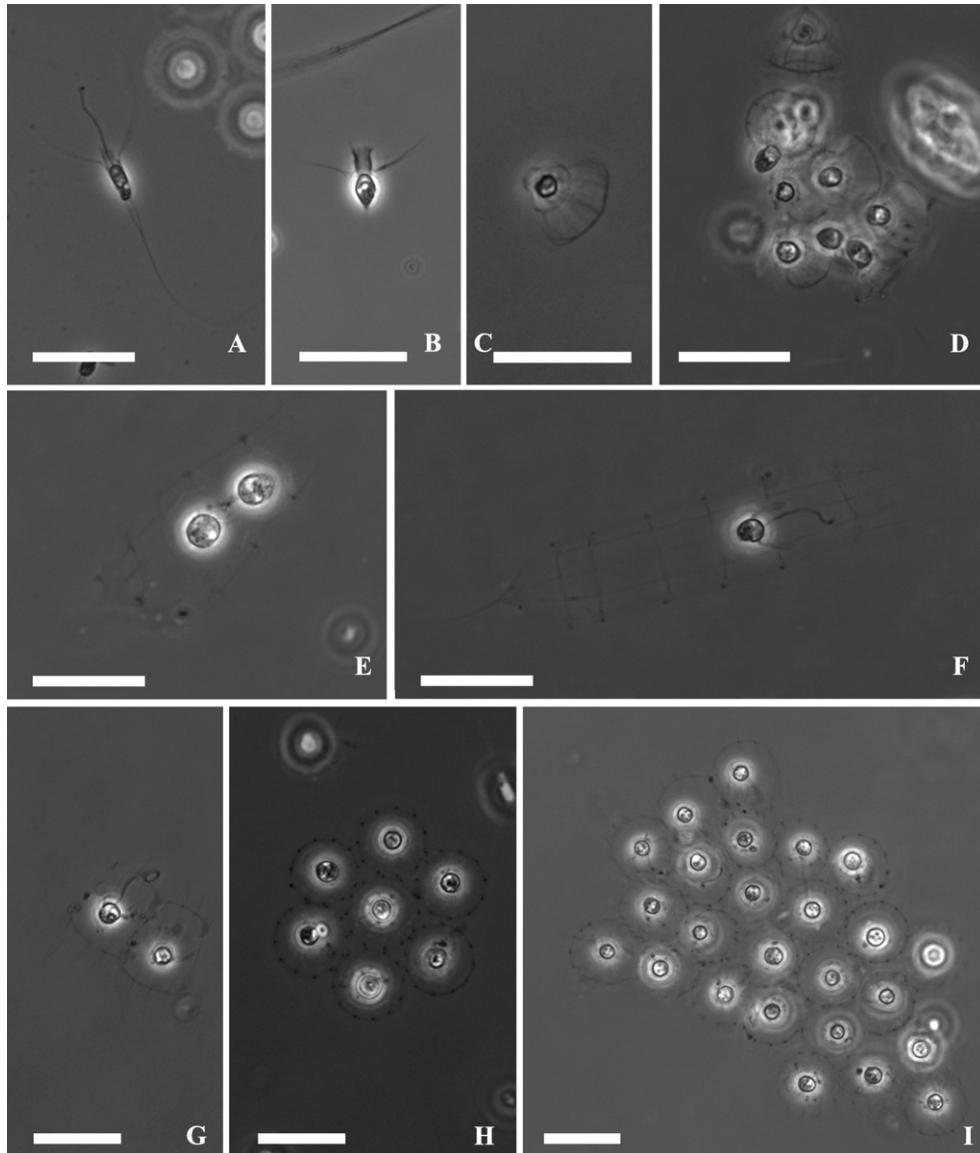


Figure 1 Acanthoecid choanoflagellates of the Ross Sea. **A.** *Bicosta spinifera*. **B.** *Calliacthanta* sp. **C** and **D.** *Parvicorbicula socialis* single cell (C) and in colonial form (D). **E** and **F.** *Diaphanoeca multiannulata* with a recently divided daughter cell inside the parental lorica (E). **G–I.** *Crinolina aperta* in pairs (G) or in colonies (H and I). Cells of *C. aperta* colonies remained attached by longitudinal costae (G and H). Scale bar = 20 μ m.

the outer surface of the cell and continued as spines. The absence of transverse costa was observed in some organisms (Fig. 1A). Specimens belonging to the genus *Calliacthanta* had a conical shaped lorica and the lorica chamber delimited, at the anterior part of the cell body, by a transverse costa (Leadbeater 1978). Those features were observed in some of our cells (Fig. 1B). *Parvicorbicula socialis* was distinct by the formation of colonies (Fig. 1D), a conical basket lorica (Fig. 1C) in which the cell body was located, two circular transverse costae with marked differences in diameter and approximately 10 longitudinal costae, all of which were present. *Diaphanoeca multiannulata* (Buck 1981) was described to possess 4–5 transverse circular costae and barrel-shaped siliceous lorica (Fig. 1E).

Few years later, Thomsen et al. (1990) highlighted that lorica dimensions of *D. multiannulata* were highly variable together with length and number of transverse and longitudinal costae and strips. The most frequently observed specimens had morphological characteristics reported in the original description (Buck 1981). Very few longer (~80 μ m) specimens with a straight outline and seven transverse costae (Fig. 1F), similar to fig. 43 in Thomsen et al. (1990) were also observed within samples. Finally, the open lorica, the number of longitudinal costae (11–12), the length of their posterior projections and the slight differences in diameter of the two transverse costae that confers them an almost cylindrical shape, led us to identify those specimens as *C. aperta* (Fig. 1G–I) and not as

C. isefjordensis, a quite similar species of the same genus. All the species mentioned above, except for *P. socialis*, possessed characteristic posterior “tails” or projections that are thought to be indicators of the presence of ice (Manton et al. 1975). Although our sampling area is an ice-free zone (polynya), ice floes can retain choanoflagellates (Fritsen et al. 2001) and may transport them to other areas.

Loricata choanoflagellates were the most abundant group within the plankton community with peak concentrations above 10^6 cells/liter reached in a transect that included stations from 43 to 48 (75°18.8'S–76°36'S, 168°53.2'E–177°59.8'E). Maxima densities (above 3.5×10^6 cells/liter) were observed at the end of January at stations 44 (45 m) and 48 (42 m). These abundances contributed 55% and 42% respectively of the total plankton abundance. Within the choanoflagellate group, *P. socialis* contributed 44% and 35.6%, whereas *C. aperta* reached the 45.8% and 54.5% respectively. *Diaphanoeca multiannulata* and *Calliantha* spp. appeared at <10% and *B. spinifera* was undetectable within those samples. Previous studies reported maximal choanoflagellates concentrations of 2×10^6 cells/liter at the ice edge zone (Buck and Garrison 1983) and 2.7×10^5 cell/liter at 31 m (Buck and Garrison 1988) in the Weddell Sea. Abundances of 10^5 and 2.1×10^6 cell/liter have been reported in the freeboard sea ice of the eastern zone and in the sea ice area of the Ross Sea respectively (Fritsen et al. 2001; Garrison et al. 2005). Dennett et al. (2001) observed high average biomass (65%) of heterotrophic nanoplanktoners during autumn in the southeastern part of the Ross Sea, where choanoflagellates dominated but the numerical contribution to the total biomass was not indicated. Thus, the cell concentrations of loricata choanoflagellates observed in this study are the highest reported in the Antarctic Ocean so far. Moreover, because these observations, were made along a transect and across the water column in the central Ross Sea polynya, they add new information about the biogeographical distribution of the loricata choanoflagellates in a poorly studied area (see fig. 9.12. in Leadbeater 2015).

The choanoflagellate bloom observed in this study was characterized by the presence of *P. socialis* (Fig. 1E, F) and *C. aperta* colonies (Fig. 1G–I), that reached ~200 μ m length and 100 μ m width in the latter species. The formation of colonies in some choanoflagellates has been suggested to be a consequence of extremely calm weather conditions during which the daughter cell completes the formation of the lorica close to the parental cell and the new lorica remains attached to it by the longitudinal costae of the foremost costal rings (Thomsen 1976). Colonies formed by *P. socialis* have been described as a mat formed by clusters of four cells (Braarud 1935; Manton et al. 1976) or spherical with clusters of six cells (Pavillard 1917). The former authors suggested that globular colonies were unnaturally formed as a consequence of the weak contact between cells after sample fixation and handling. However, Thomsen (1976) reported that spherical colonies were the result of the physical contact between cells with different diameters of the transverse costal

rings. Conversely, we observed *C. aperta* specimens with a quite straight outline of the siliceous lorica that formed almost flat colonies with up to 25 cells in which one cell was contacted to six surrounding cells that resulted in a zigzag pattern (Fig. 1G–I). Thus, the cell disposition in *C. aperta* (Fig. 1H, I) coincided with those for *P. socialis* drawn by Pavillard (1917). The arrangement of cells within the colony (one cell surrounded by six) and the different outlines of the lorica, conical, in *P. socialis* (Fig. 1C, D), and cylindrical, in *C. aperta* (Fig. 1G–I), are in agreement with Pavillard (1917) and Thomsen (1976). Previous studies described *C. aperta* colonies (Buck and Garrison 1983; Marchant et al. 1987) but the cell pattern in Fig. 1H, I is shown in this work for the first time.

Choanoflagellates play an important ecological role in the transfer of carbon and energy from surface to deep water communities and from lower to higher trophic levels (Vincent 2004). Studies on natural samples and cultures indicated that their diet includes bacteria and particulate organic matter (Buck and Garrison 1988). At the same time, choanoflagellates are potential preys of zooplankton, such as the Antarctic krill (Tanoue and Hara 1986). Thus, the concentrations reached and the formation of such large colonies suggests that choanoflagellates may play a pivotal role by connecting primary producers and higher trophic levels in the Antarctic pelagic food web. Our observations support the recent trend of changes in the plankton succession that are potentially restructuring trophic relationships in the Antarctic food web (Mangoni et al. 2017; Rivaro et al. 2017).

ACKNOWLEDGMENTS

This study was carried out in the framework of the activities of the Italian National Program for Antarctic Research (P-ROSE Project, PNRA, 2016/A3.06) which provided logistic support. We thank the officers, crew and technical personnel on board the R/V *Italica*, G. Zazo for his help during the cruise and F. Iamunno for technical support. Our special thanks to V. Saggiomo for his valuable criticism of this work.

LITERATURE CITED

- Braarud, T. 1935. The “Ost” expedition to the Denmark Strait 1929. II The Phytoplankton and its conditions of growth. *Hvalråd. Skr.*, 10:1–173.
- Buck, K. R. 1981. A study of choanoflagellates (Acanthoecidae) from the Weddell Sea, including a description of *Diaphanoeca multiannulata* n. sp. *J. Protozool.*, 28:47–54.
- Buck, K. R. & Garrison, D. L. 1983. Protists from the ice-edge region of the Weddell Sea. *Deep-Sea Res. Pt I*, 30:1261–1277.
- Buck, K. R. & Garrison, D. L. 1988. Distribution and abundance of choanoflagellates (Acanthoecidae) across the ice-edge zone in the Weddell Sea, Antarctica. *Mar. Biol.*, 98:263–269.
- Chen, B. 1994. Distribution and abundance of choanoflagellates in Great-Wall Bay, King George Island, Antarctica in austral summer. *Proc. NIPR Symp. Polar Biol.*, 7:32–42.
- Dennett, M. R., Mathot, S., Caron, D. A., Smith, W. O. & Lonsdale, D. J. 2001. Abundance and distribution of phototrophic

- and heterotrophic nano- and microplankton in the southern Ross Sea. *Deep-Sea Res. Pt II*, 48:4019–4037.
- Fritsen, C. H., Coale, S. L., Neenan, D. R., Gibson, A. H. & Garrison, D. L. 2001. Biomass, production and microhabitat characteristics near the freeboard of ice floes in the Ross Sea, Antarctica, during the austral summer. *Ann. Glaciol.*, 33:280–286.
- Garrison, D. L., Gibson, A., Coale, S. L., Gowing, M. M., Okolodkov, Y., Fritsen, C. H. & Jeffries, M. O. 2005. Sea-ice microbial communities in the Ross Sea: autumn and summer biota. *Mar. Ecol. Prog. Ser.*, 300:39–52.
- Leadbeater, B. S. C. 1978. Renaming of *Salpingoeca sensu Grøntved*. *J. Mar. Biol. Assoc. U.K.*, 58:511–515.
- Leadbeater, B. S. C. 2015. Choanoflagellate ecology. In: Leadbeater, B. S. C. (ed.), *The Choanoflagellates: Evolution, Biology and Ecology*. Cambridge University Press, Cambridge. 9:202–240.
- Lund, J. W. G., Kipling, C. & Le Cren, E. D. 1958. The inverted microscope method of estimating algal numbers and the statistical basis of estimations by counting. *Hydrobiologia*, 11:143–170.
- Mangoni, O., Saggiomo, V., Bolinesi, F., Margiotta, F., Budillon, G., Cotroneo, Y., Mistic, C., Rivaro, P. & Saggiomo, M. 2017. Phytoplankton blooms during austral summer in the Ross Sea, Antarctica: driving factors and trophic implications. *PLoS ONE*, 12:e0176033. <https://doi.org/10.1371/journal.pone.0176033>.
- Manton, I., Sutherland, J. & Leadbeater, B. S. C. 1976. Further observations on the fine structure of marine collared flagellates (Choanoflagellata) from arctic Canada and west Greenland: species of *Parvicorbicula* and *Pleurasiga*. *Can. J. Bot.*, 54:1932–1955.
- Manton, I., Sutherland, J. & Thomsen, H. A. 1975. Four new species of choanoflagellates from Arctic Canada. *Proc. R. Soc. Lond. B Biol. Sci.*, 189:15–27.
- Marchant, H., Van den Hoff, J. & Burton, H. 1987. Loricatae choanoflagellates from Ellis Fjord, Antarctica including the description of *Acanthocorbis tintinnabulum* sp. nov. *Proc. NIPR Symp. Polar Biol.*, 1:10–22.
- Pavillard, M. J. 1917. Protistes nouveaux ou peu connus du plancton mediterraneen. *C. R. Hebd. Seanc. Acad. Sci.*, 164:92–928.
- Rivaro, P., Ianni, C., Langone, L., Ori, C., Alicino, G., Cotroneo, Y., Saggiomo, M. & Mangoni, O. 2017. Physical and biological forcing of mesoscale variability in the carbonate system of the Ross Sea (Antarctica) during summer 2014. *J. Mar. Syst.*, 166:144–158.
- Tanoue, E. & Hara, S. 1986. Ecological implications of fecal pellets produced by the Antarctic krill *Euphausia superba* in the Antarctic Ocean. *Mar. Biol.*, 91:359–369.
- Thomsen, H. A. 1976. Studies on marine choanoflagellates. II. Fine structural observations on some silicified choanoflagellates from the Isefjord (Denmark), including the description of two new species. *Norw. J. Bot.*, 23:33–51.
- Thomsen, H. A., Buck, K. R., Coale, S. L., Garrison, D. L. & Gowing, M. M. 1990. Loricatae choanoflagellates (Acanthoecidae, Choanoflagellida) from the Weddell Sea, Antarctica. *Zool. Scr.*, 19:367–387.
- Thronsen, J. 1970. *Salpingoeca spinifera* sp. nov., a new planktonic species of the Craspedophyceae recorded in the Arctic. *Br. Phycol. J.*, 5:87–89.
- Utermöhl, v. H. 1931. Neue Wege in der quantitativen Erfassung des Planktons (mit besondere Berücksichtigung des Ultraplanktons). *Verh. Int. Ver. Theor. Angew. Limnol.*, 5:567–595.
- Vincent, W. F. 2004. *Microbial ecosystems of Antarctica*. Cambridge University Press, Cambridge. p. 320.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Figure S1. Map of the sampling stations in the central Ross Sea polynya (Antarctica). Note that stations 15 and 57 were sampling twice.