INTERNATIONAL JOURNAL OF AGRICULTURE & BIOLOGY
ISSN Print: 1560–8530; ISSN Online: 1814–9596
19F–052/2019/22–4–763–768
DOI: 10.17957/IJAB/15.1127
http://www.fspublishers.org

Full Length Article

Production of Aldehydes in Diatoms and Dinoflagellates and the Detrimental Effect on Copepod Grazers

Jie Li[1*†], Yuanyuan Wang[1,2†], Yanjuan Liang[1], Jiantao Huang[1], Yiman Liu[1] and Jinfang Lu[1]
[1]*Institute of environmental and municipal engineering, Qingdao University of Technology, 266033 Qingdao, China*
[2]*Aquatic Ecology, Department Biology II, Ludwig Maximilian University of Munich, 82152 Munich, Germany*
[*]For correspondence: jli@qut.edu.cn
[†]These authors contributed equally to this work

Abstract

Marine algae, including diatoms, are the primary food source of oceanic ecosystems, sustaining the marine food chain to top consumers and playing a key role in marine food webs. Their beneficial role is questioned due to the finding of harmful compounds in their secondary metabolites, such as polyunsaturated aldehydes, which impair reproductive and developmental processes of their grazers. To investigate the response of dominant copepod grazers in the Jiaozhou Bay to different diets and how aldehydes vary in batch algal cultures over time, we carried out experiments in which two diatoms *Skeletonema marinoi* and *Chaetoceros didymus* were fed to the copepod *Calanus sinicus*. Controls were run with two dinoflagellates *Prorocentrum micans* and *Scrippsiella trochoidea*. In our two set of experiments, we found a C_7saturated aldehyde released by those four algal species, which is less reported. Our results showed that diatoms have greater potential for aldehydes production than dinoflagellates and the peak of aldehydes production occurred mostly in the first and second week, which clearly includes lag and exponential phase. However, the detrimental effect on copepods was species dependent. Some dinoflagellates are also potential aldehydes producers. Even though some dinoflagellates released fewer aldehydes than diatoms, the inhibitory effect on copepods was greater.

Keywords: Algae; Marine; *Chaetoceros didymus* Copepods

Introduction

Diatoms, one of the most common classes of autotrophic phytoplankton, contribute 40–45% of the total primary production in oceanic environments, playing a key role in marine food webs (Yool and Tyrrell, 2003). Traditionally, diatoms are regarded as a good food source for primary consumers of herbivorous plankton, such as the small crustacean copepods. However, recently, the traditional view of marine food web energy flow from diatoms to fish by means of copepods is challenged with the discovery that some diatom species produce cytotoxic secondary metabolites, including polyunsaturated fatty acids (PUFAs) (Grima *et al*., 1995; Li *et al*., 2008; Pezzolesi *et al*., 2017), polyunsaturated aldehydes (PUAs) (Ianora *et al*., 2012; Pezzolesi *et al*., 2017), and other compounds collectively termed as oxylipids. PUAs are mainly synthesized from enzymatic degradation of PUFAs upon cell wounding. It is confirmed that this wound-activated release takes place not only in the laboratory (Miralto *et al*., 1999; Pohnert, 2000) but also in the field (Ianora *et al*., 2004; Ribalet *et al*., 2014). This process is a form of allelopathy, utilized by diatoms and other marine phytoplankton as a defense mechanism against predation and inhibits competitors by producing certain chemicals (Strom, 2008; Ianora *et al*., 2011). It has been reported that 36% of the 51 investigated diatom species released aldehydes showing a species-specific and strain-dependent production (Wichard *et al*., 2005) and that the aldehydes are toxic to a wide variety of organisms, from bacteria to invertebrates, including phytoplankton (Pohnert, 2010). This defensive reaction can induce abortions, poor larval development and high offspring mortality in many invertebrates. Numerous studies (Miralto *et al*., 1999; Wichard *et al*., 2007; Ianora *et al*., 2012; Lavrentyev *et al*., 2015) indicate that aldehydeshave an adverse effect on reproduction and development of copepods which Ban *et al*. (1997) described as "diatom–copepod paradox". However, the effect can not only be species-strain-specific, but also environmental-specific. Aldehydes composition and production can also depend on nutrient limitation and growth phase stage (Pohnert, 2002; Ribalet *et al*., 2007). Additionally, the response of different copepod species to specific algal diets can vary (Ianora *et al*., 2003; Amin *et al*., 2011).

The aim of the study is to clarify the response of dominant copepod grazers in the Jiaozhou Bay (JZB) to

To cite this paper: Li, J., Y. Wang, Y. Liang, J. Huang, Y. Liu and J. Lu, 2019. Production of aldehydes in diatoms and dinoflagellates and the detrimental effect on copepod grazers. *Intl. J. Agric. Biol*., 22: 763–768

different diets and how aldehydes vary in batch algal cultures over time. We hypothesized that copepods would prefer dinoflagellates over diatoms and aldehydes production would increase with time. JZB is a typical semi-enclosed bay located in the west of the Yellow sea with a water area of 340 km^2 and an average depth of 7 m (Gao *et al*., 2014), where harmful algal blooms (HABs) occur often (Yuan *et al*., 2017). So far, the correlation of phytoplankton blooms with several environmental parameters has been investigated in detail (Mundy *et al*., 2014; Carstensen *et al*., 2015), while we should also focus on how phytoplankton blooms can alter oceans biochemically not only to the water body but also to their grazers.

Materials and Methods

Copepod Sampling

Calanus sinicus are cosmopolitan, euryhaline and opportunistic copepod species. They were caught with a 500 μm net by vertical hauls on March 2009 from Jiaozhou Bay (36°06'N; 120°19'E), where they often account for the major part of the copepod assemblage. They were transported to the laboratory within one hour after sampling. Healthy mature females with intact appendages and active swimming behavior were sorted within 4 h and reared in 0.25 μm sieved ambient seawater.

Phytoplankton Cultures

Two diatoms *Skeletonema costatum* (SKE) and *Chaetoceros didymus* (CHA) and two dinoflagellates *Prorocentrum micans* (PRO) and *Scrippsiella trochoidea* (SCR) were selected as copepod food source. SKE and CHA are dominant diatom species in JZB, where they often form dense blooms. All the algae were collected from the ecological laboratory, Institute of Oceanology, Chinese Academy of Sciences, Qingdao. Monoclonal cultures were cultivated in f/2 medium at 20±0.5°C under a 12:12 light/dark cycle (Guillard, 1975).

Experimental Set-up

Two sets of experiments were carried out. The first was to detect how aldehydes vary in batch algal cultures within one month. The growth experiment were conducted in three independent replicates, inoculating four algal species in 5 L f/2 medium at 20±0.5°C under a 12:12 light/dark cycle. To investigate production of aldehydes during growth under standard conditions, several independent algal exudates were sampled every 7 days within one month, meanwhile, the growth curves (Fig. 1) were drawn according to the daily cell concentrations, which were monitored daily by Coulter Counter (Multisizer 3.5 μm aperture; measuring range: 2–30 μm). Every seven days, algal exudates were centrifuged and 3 mL concentrated algae was stored in 20 mL sealed headspace bottleat-20°C.

In a second set of experiments, we focused on the effects of different diets on the growth and recruitment of copepods. *Calanus sinicus*, after being captured, were acclimated in GF/F filtered seawater for 48 h. The egg production rate and hatching success of first day after sampling were recorded as the initial values to estimate the natural conditions. From the second day, females were acclimated 48 h under experimental conditions of various food types and concentrations. To study the effect of aldehydes on copepods, algal cultures were renewed with new f/2 medium to maintain cells in exponential phase (Fig. 1) and diluted to 0.5 μg C mL^{-1} using GF/F filtered seawater daily for feeding.

The grazing experiment was conducted in five biologically independent replicates, placing six adult female copepods in four monoclonal (SKE, CHA, PRO, SCR) cultures (initial cell density 0.5 μg C mL^{-1}) respectively at exponential growth phase in 400 mL bottles. These bottles were plastic cylinders with a 220 μm sieve placed 5 cm above the bottom to separate eggs from copepods to prevent cannibalism of eggs. Each treatment with 300 ml algal suspension was incubated in a biochemistry cultivation cabinet under dim light at 6.5±0.5°C in accordance with the copepods sampling temperature.

Half a bottle of suspension was collected for egg counting and supplemented with equal amount of fresh algal culture. Over the period of 15 days, the number of surviving copepods was recorded and female survival (FS) as well as egg production rate (EPR) was calculated. After that, 30 eggs isolated from suspension were incubated in culture dishes with 20 ml filtered seawater under dim light at 6.5±0.5°C. This process was monitored with a microscope every 24 h until 72 h to ensure all vital eggs had sufficient time to hatch. Hatching success was defined as the number of hatched nauplii which developed into naupliar stage NII. Finally, the cultivated suspension for feeding copepods was harvested by centrifugation. After removing the supernatant, 3 mL concentrated algae was stored in 20 mL sealed headspace bottle at 4°C.

Aldehydes Quantification

The quantification of aldehydes concentration was carried out by HSSE (Head space sorptive extraction)-GC-MS technique. Firstly, 2 mL O-(2,3,4,5,6-pentafluorobenzyl) hydroxylamine hydrochloride solution (PFBHA, Sigma-Aldrich, U.S.) was incubated at 75°C for 3.5 min for headspace absorption onto the PDMS/DVB fiber. The algal sample was ultrasonicated for 30 sec and then incubated at 75°C for 10 min. Afterwards, the PFBHA-absorbed PDMS/DVB fiber was put into the headspace bottle with treated algal sample and incubated at 75°C for 20 min. After that, PUA concentration was determined by GC-MS and molecular identification was done by comparison of retention times and mass spectra with those of available commercial standards: trans-2-hexenal

(99%, Tokyo Chemical Industry, Japan), trans, trans-2,4-heptadienal (94%, Tokyo Chemical Industry, Japan), trans-2-decenal (95%, Tokyo Chemical Industry, Japan), trans, trans-2,4-decadienal (95%, Tokyo Chemical Industry, Japan), heptaldehyde (1000 mg/L, Accustandard, U.S.) and trans-2-heptenal (1000 mg/L, Accustandard, U.S.).

Results

Aldehydes Production in different Growth Phases

We set the first seven-day as stage Iand continue respectively. A time-dependent production of aldehydes was observed for all cultures (Fig. 2). The total aldehydes production of the four algae species all reached peak at stage I and stage II, which clearly includes lag and exponential phase (Fig. 1). Among the aldehydes, trans-2-hexenal (0 ~ 263.6910^{-3} μg/g DW) (DW represents dry weight of algae) and trans, trans-2, 4-heptadienal (0 ~ 469.19 10^{-3} μg/g DW) were always the most abundant, accounting for over 50%, during the whole period. Other aldehydes accounted for a lower proportion of total aldehydes, especially trans-2-decenal, which only appeared in trace amounts on SKE I and CHA III.

During the whole period, the average amount of aldehydes produced by diatoms was higher than the one produced by dinoflagellates (Fig. 3), which showed diatoms have greater potential for aldehydes production than dinoflagellates. Among them, CHA produced the maximum aldehydes and PRO the minimum. Additionally, there were six aldehydes released by diatoms, while only five were produced by dinoflagellates. Specifically, there was no trans-2-decenal in PRO and SCR and no trans-2-heptenal in PRO. The ability of diatoms to release aldehydes was significantly higher than the ability of dinoflagellates, especially in the late growth phase. Among all the aldehydes, we found a saturated aldehyde, heptaldehyde (Table 1), which is less reported.

Female Survival, Egg Production and Hatching Success

In the second set of experiments, female survival (FS), egg production rate (EPR) and HS (hatching success) of *Calanus sinicus* varied with different diets (Figs. 4–6). FS of females fed on PRO (Fig. 4) was higher (average 95.11%) during 15 days, while FS of other cultures declined sharply from the sixth day and stayed below 90% (on average, SKE, CHA and SCR were 79.35%, 81.08% and 84.52%, respectively.). In general, viability of females fed on dinoflagellates was higher than of females fed on diatoms.

EPR and HS of females fed on PRO also remained at a high level (4.13±1.64 (mean ±s.d.) eggs female^{-1}day^{-1} and 70.93±14.73%, respectively), even though they were lower compared to the initial value (7.13±2.96 (mean ±s.d.) eggs female^{-1}day^{-1} and 81.11±2.94%, respectively). (Fig. 5). During the 15-day experiment, the average values of EPR

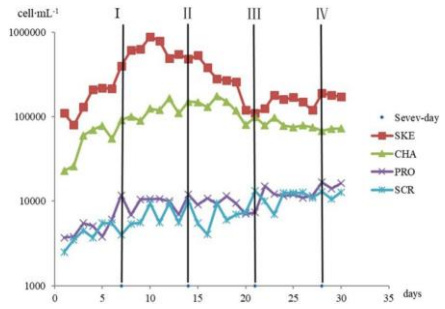

Fig. 1: Growth curves

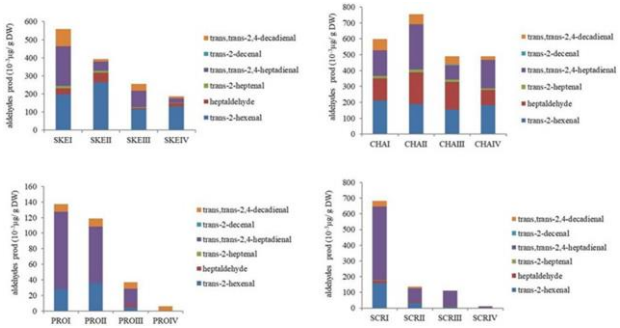

Fig. 2: Aldehydes production in every stage

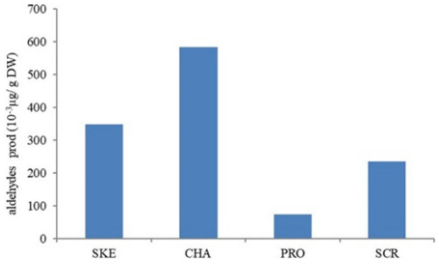

Fig. 3: Average aldehydes production during the whole period

for different diets ranged from 1.35±1.29 eggs female^{-1} day^{-1} to 4.42±1.64 eggs female^{-1} day^{-1}. Despite the low rate of the CHA treatment at the start, it climbed to 4.75±1.33 eggs female^{-1} day^{-1}at day thirteen and remained at 3.00±1.57 eggs female^{-1} day^{-1} after a small decrease. On the contrary, SKE and SCR treatments experienced a rapid increase at first, but then declined to below 2 eggs female^{-1} day^{-1}. HS of females fed on SKE, CHA and SCR were all below 60% (56.43, 55.91 and 53.40%, respectively). There was no big difference among these three species. All diets resulted in decreasing female survivor ship, viability and fecundity of eggs, while the PRO diet showed the lowest detrimental effect (Fig. 6).

Aldehydes Production with Copepods Grazing

Aldehydes composition and production were distinctly different between diatom diets (SKE and CHA) and

Table 1: Properties and structures of aldehydes

Name	Molecular Formula	Molecular Structure	CAS Number	Molecular Weight	Boiling Point (°C)
trans-2-hexenal	$C_6H_{10}O$		6728-26-3	98.15	81.6
heptaldehyde	$C_7H_{14}O$		111-71-7	114.19	153
trans-2-heptenal	$C_7H_{12}O$		18829-55-5	112.17	90
trans,trans-2,4-heptadienal	$C_7H_{10}O$		4313-03-5	110.15	84.7
trans-2-decenal	$C_{10}H_{18}O$		3913-81-3	154.23	78~80
trans,trans-2,4-decadienal	$C_{10}H_{16}O$		25152-84-5	152.23	115.2

dinoflagellate diets (PRO and SCR) (Fig. 7). Firstly, the total amount of aldehydes released by diatoms was much higher than by dinoflagellates. Secondly, there were also six aldehydes released by diatoms, while only five were released by dinoflagellates, without trans-2-decenal in PRO and SCR. Four algal species, however, had in common that trans-2-hexenal and trans, trans-2,4-heptadienal were the most abundantly produced aldehydes, accounting for over 60%.

Discussion

Two diatom isolates (SKE and CHA) and two dinoflagellate isolates (PRO and SCR) were analyzed for aldehydes production over time or coexisting with copepods. The result of the first set of experiments showed that the intact diatoms and dinoflagellate cells could release aldehydes dependent on the growth stage. The peak of aldehydes production was mostly in stage I and II, which clearly includes lag and exponential phase. Vidoudez and Pohnert (2008) reported that intact cells of SKE released a pronounced burst of PUAs which only happened during a defined time span during stationary phase. Our results, however, showed that aldehydes release happened not only in stationary phase, but also in lag and exponential phase. What's more, the type and average amount of aldehydes released by diatoms was bigger than the one released by dinoflagellates, which demonstrated a great potential of aldehydes-formation of diatoms. Similarly, in the second set of experiments, diatoms also showed a better ability to produce aldehydes, when their cells were damaged by copepods grazing.

Traditionally, diatoms represent a quality food source for copepods, due to the well-balanced amino acid composition which can be stored in copepods eggs and utilized for nauplii development (Nahon *et al*., 2010). Copepods are unable to synthesize essential amino acids, that are important in regulating cell membranes and precursors for tissue hormones, de novo. However, some diatoms lack or are deficient in some essential nutrients, such as C, N, P, polyunsaturated fatty acids (PUFAs), sterols and minerals (Brett and Müller-Navarra, 1997; Vehmaa *et al*., 2011). Therefore, a mineral and biochemical inadequacy of a pure diatom diet cannot satisfy the nutrient demand of

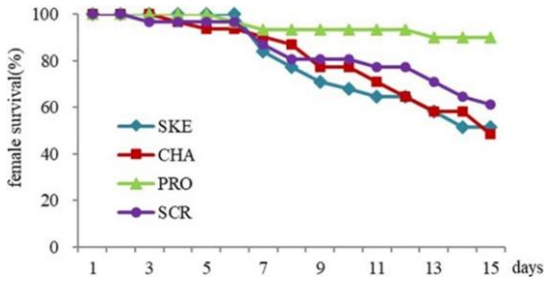

Fig. 4: Female survival (%) of *Calanus sinicus* fed on 4 diets (SKE: *Skeletonema costatum*; CHA: *Chaetoceros didymus*; PRO: *Prorocentrum micans*; SCR: *Scrippsiella trochoidea*)

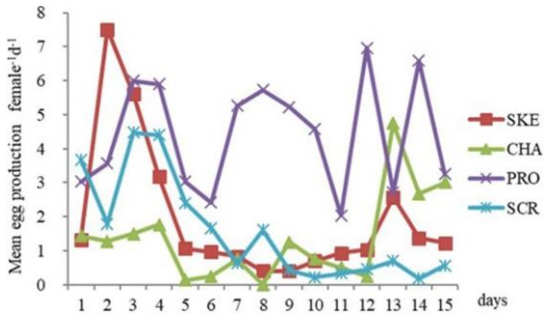

Fig. 5: Daily egg production of female *Calanus sinicus* fed on 4 diets (SKE: *Skeletonema costatum*; CHA: *Chaetoceros didymus*; PRO: *Prorocentrum micans*; SCR: *Scrippsiella trochoidea*)

copepods. In the field, the growth peak of copepods and diatoms is always unsynchronized and copepods generally peak well after diatom blooms occur, suggesting that diatoms are not an ideal diet (Ianora *et al*., 2004).

On the other hand, secondary metabolites released by diatoms are another important factor that is causing reproductive failure in copepods. Our results are consistent with studies (Ban *et al.*, 1997; Ianora *et al.*, 2004; Wichard *et al.*, 2007; Li *et al*., 2008; Pohnert, 2010; Brugnano *et al*., 2016) showing that aldehydes have a detrimental effect on the development and reproduction of copepods. Some studies (Niehoff, 2004; Poulet *et al*., 2007) found that higher EPR and HS were produced on dinoflagellate diets rather than on diatom diets, while our results indicate that the adverse effect was species dependent. The aldehydes

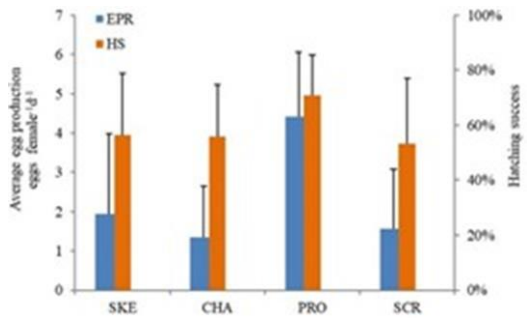

Fig. 6: Average egg production rate and hatching success of female *Calanus sinicus* fed on 4 diets (SKE: *Skeletonema costatum*; CHA: *Chaetocerosdidymus*; PRO: *Prorocentrummicans*; SCR: *Scrippsiellatrochoidea*)

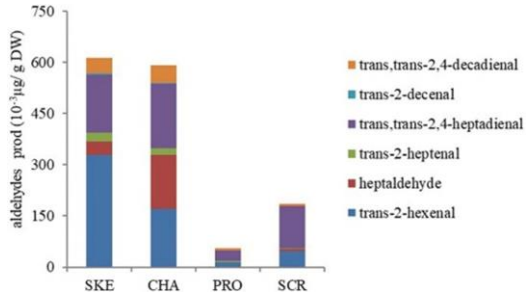

Fig. 7: aldehydes composition and production

production of SKE (diatom) was about three times of the amount of SCR (dinoflagellate), while the EPR of SKE (diatom) was higher than the one of SCR (dinoflagellate) and their HS was almost the same. Thus, some dinoflagellates are also potential aldehydes producer. We should not focus only on diatoms, but also on dinoflagellates in future studies. In addition, a mixed diet may serve to dilute toxins, a concept that is already explored in some studies (Ianora *et al*., 2004; Li *et al*., 2010).

Currently, most studies focus on unsaturated aldehydes, such as heptadienal, octadienal, octatrienal, decadienal and decatrienal (Pezzolesi *et al*., 2017). In our study, however, we found a new released aldehyde, heptaldehyde, which is a C_7saturated aldehyde. At present, little is known about the effect of saturated aldehydes, which could be explored in the future work. Adolph *et al.* (2003) and Romano *et al.* (2011) reported that the proliferation inhibition activity of aldehydes depends on the degree of unsaturation and length of the chain. Unsaturated aldehydes showed greater activity than saturated aldehydes of the same chain length. Moreover, Pichierri *et al.* (2016) confirmed that the anti-cell-growth activity is stronger in longer-chain aldehydes than in shorter-chain ones. So far, the mechanism for aldehydes-mediated effect is not completely clear yet. Miralto *et al.* (1999) demonstrated that the decadienal can induce apoptosis in mammalian tumor cells. Subsequently, decadienal-induced apoptosis in the copepod *Calanus helgolandicus* and in sea urchin *Paracentrotus lividus* was demonstrated experimentally (Romano *et al*., 2003). Lettieri *et al.* (2015) showed that decadienal can affect the expression level of genes involved in stress response and developmental processes. In our second set of experiments, diatoms released a lot more decadienal than dinoflagellates, thus this might be a reason why diatoms have a greater inhibitory effect on copepods than dinoflagellates.

Conclusion

Our results underline the importance that marine phytoplankton is not a passive participant in biotic interactions, and that they can actively alter their own susceptibility to attackers by releasing aldehydes, acting as a form of population control in the ocean.

Acknowledgements

This study was supported by Chinese marine industry research special funds for public welfare projects (201505001), Shandong province key research and development plan, China (2018GSF117012) and open fund from Shandong Yellow River delta ecological environment key Laboratory, China (2016KFJJ01).

We thank Professor Herwig Stibor and Tine Hohmann for the interesting discussion and suggestions during the writing of this paper.

References

Adolph, S., S.A. Poulet and G. Pohnert, 2003. Synthesis and biological activity of α, β, γ, δ-unsaturated aldehydes from diatoms. *Tetrahedron,* 59: 3003–3008

Amin, R.M., M. Koski, U. Båmstedt and C. Vidoudez, 2011. Strain-related physiological and behavioral effects of *Skeletonema marinoi* on three common planktonic copepods. *Mar. Biol.,* 158: 1965–1980

Ban, S., C. Burns, C. Castel, E. Christou, R. Escribano, F.F. Umani, S. Gasparini and F.G. Ruiz, 1997. The paradox of diatom-copepod interactions. *Mar. Ecol. Progr. Ser*., 157: 287–293

Brett, M. and D. Müller-Navarra, 1997. The role of highly unsaturated fatty acids in aquatic foodweb processes. *Freshwater Biol*., 38: 483–499

Brugnano, C., A. Granata, L. Guglielmo, R. Minutoli, G. Zagami and A. Ianora, 2016. The deleterious effect of diatoms on the biomass and growth of early stages of their copepod grazers. *J. Exp. Mar. Biol. Ecol.,* 476: 41–49

Carstensen, J., R. Klais and J.E. Cloern, 2015. Phytoplankton blooms in estuarine and coastal waters: Seasonal patterns and key species. *Estuar. Coast. Shelf Sci.,* 162: 98–109

Gao, G.D., X.H. Wang and X.W. Bao, 2014. Land reclamation and its impact on tidal dynamics in Jiaozhou Bay, Qingdao, China. *Estuar. Coast. Shelf Sci*., 151: 285–294

Grima, E.M., J.A.S. Pkrez, F.G. Camacho, A.R. Medina, A.G. Gimknez and D.L. Alonso, 1995. The production of polyunsaturated fatty acids by microalgae: From strain selection to product purification. *Process Biochem.,* 30: 711–719

Guillard, R.R.L., 1975. *Culture of Phytoplankton for Feeding Marine Invertebrates.* Culture of Marine Invertebrate Animals. Springer, USA

Ianora, A., A. Miralto and G. Romano, 2012. Antipredatory defensive role of planktonic marine natural products. *In: Handbook of Marine Natural Products,* pp: 711–748. Fattorusso, E., W.H. Gerwick and O. Taglialatela-Scafati (Eds.). Springer, The Netherlands

Ianora, A., M.G. Bentley, G.S. Caldwell, R. Casotti, A.D. Cembella, J. Engström-Öst and D. Vaiciute, 2011. The relevance of marine chemical ecology to plankton and ecosystem function: an emerging field. *Mar. Drugs,* 9: 1625–1648

Ianora, A., A. Miralto, S.A. Poulet, Y. Carotenuto, I. Buttino, G. Romano, R. Casotti, G. Pohnert, T. Wichard, L. Colucci-D'Amato, G. Terraggano and V. Smetacek, 2004. Aldehyde suppression of copepod recruitment in blooms of a ubiquitous planktonic diatom. *Nature*, 429: 403–407

Ianora, A., S. Poulet and A. Miralto, 2003. The effects of diatoms on copepod reproduction: A review. *Phycologia*, 42: 351–363

Lavrentyev, P., G. Franze, J. Pierson and K. Stoecker, 2015. The effect of dissolved polyunsaturated aldehydes on microzooplankton growth rates in the chesapeake bay and atlantic coastal waters. *Mar. Drugs,* 13: 2834–2856

Lettieri, A., R. Esposito, A. Ianora and A. Spagnuolo, 2015. *Ciona intestinalis* as a marine model system to study some key developmental genes targeted by the diatom-derived aldehyde decadienal. *Mar. Drugs*, 13: 1451–1465

Li, J., S. Sun, C.L. Li, Z. Zhang and Z.C. Tao, 2010. Effects of single and mixed diatom diets on the reproduction of copepod *Calanus sinicus*. *Acta Hydrochim. Hydrobiol.,* 34: 117–125

Li, J., S. Sun, C.L. Li, Z. Zhang and X.M. Pu, 2008. Effects of different diets on the reproduction and naupliar development of the copepod *Acartia bifilosa*. *J. Exp. Mar. Biol. Ecol*., 355: 95–102

Miralto, A., G. Barone, G. Romano, S.A. Poulet, A. Ianora, G.L. Russo, I. Buttino, G. Mazzarella, M. Laabir, M. Cabrini and M.G. Giacobbe, 1999. The insidious effect of diatoms on copepod reproduction. *Nature,* 402: 173–176

Mundy, C.J., M. Gosselin, Y. Gratton, K. Brown, V. Galindo, K. Campbell, M. Levasseur, D. Barber, T. Papakyriakou and S. Bélanger, 2014. Role of environmental factors on phytoplankton bloom initiation under landfast sea ice in Resolute Passage, Canada. *Mar. Ecol. Progr. Ser*., 497: 39–49

Nahon, S., F. Charles, F. Lantoine, G. Vétion, K. Escoubeyrou, M. Desmalades and A.M. Pruski, 2010. Ultraviolet radiation negatively affects growth and food quality of the pelagic diatom *Skeletonema costatum*. *J. Exp. Mar. Biol. Ecol*., 383: 164–170

Niehoff, B., 2004. The effect of food limitation on gonad development and egg production of the planktonic copepod *Calanus finmarchicus*. *J. Exp. Mar. Biol. Ecol*., 307: 237–259

Pezzolesi, L., S. Pichierri, C. Samorì, C. Totti and R. Pistocchi, 2017. PUFAs and PUAs production in three benthic diatoms from the northern adriatic sea. *Phytochemistry*, 142: 85–91

Pichierri, S., L. Pezzolesi, S. Vanucci, C. Totti and R. Pistocchi, 2016. Inhibitory effect of polyunsaturated aldehydes (PUAs) on the growth of the toxic benthic dinoflagellate *Ostreopsis* cf. *ovata*. *Aquat. Toxicol.,* 179: 125–133

Pohnert, G., 2010. Diatom/copepod interactions in plankton: the indirect chemical defense of unicellular algae. *ChemBiochem*, 6: 946–959

Pohnert, G., 2002. Phospholipase A2 activity triggers the wound-activated chemical defense in the diatom *Thalassiosira rotula*. *Plant Physiol.,* 129: 103–111

Pohnert, G., 2000. Wound-activated chemical defense in unicellular planktonic algae. *Angew. Chem. Intl. Ed.,* 39: 4352–4354

Poulet, S.A., A. Cueff, T. Wichard, J. Marchetti, C. Dancie and G. Pohnert, 2007. Influence of diatoms on copepod reproduction. iii. consequences of abnormal oocyte maturation on reproductive factors in *Calanus helgolandicus*. *Mar. Biol*., 152: 415–428

Ribalet, F., M. Bastianini, C. Vidoudez, F. Acri, J. Berges, A. Ianora, A. Miraito, G. Pohnert, G. Romano, T. Wichard and R. Casotti, 2014. Phytoplankton cell lysis associated with polyunsaturated aldehyde release in the northern adriatic sea. *PLoS One*, 9: e85947

Ribalet, F., T. Wichard, G. Pohnert, A. Ianora, A. Miralto and R. Casotti, 2007. Age and nutrient limitation enhance polyunsaturated aldehyde production in marine diatoms. *Phytochemistry*, 68: 2059–2067

Romano, G., M. Costantini, I. Buttino, A. Ianora and A. Palumbo, 2011. Nitric oxide mediates the stress response induced by diatom aldehydes in the sea urchin *Paracentrotus lividus*. *PLoS One,* 6: e25980

Romano, G., G.L. Russo, I. Buttino, A. Ianora and A. Miralto, 2003. A marine diatom-derived aldehyde induces apoptosis in copepod and sea urchin embryos. *J. Exp. Biol*., 206: 3487–3494

Strom, S.L., 2008. Microbial ecology of ocean biogeochemistry: a community perspective. *Science,* 320: 1043–1045

Vehmaa, A., P. Larsson, C. Vidoudez, G. Pohnert, M. Reinikainen and J. Engström-Öst, 2011. How will increased dinoflagellate: diatom ratios affect copepod egg production? — a case study from the baltic sea. *J. Exp. Mar. Biol. Ecol.,* 401: 134–140

Vidoudez, C. and G. Pohnert, 2008. Growth phase-specific release of polyunsaturated aldehydes by the diatom *Skeletonema marinoi*. *J. Plankton Res*., 30: 1305–1313

Wichard, T., A. Gerecht, M. Boersma, S.A. Poulet, K. Wiltshire and G. Pohnert, 2007. Lipid and fatty acid composition of diatoms revisited: rapid wound-activated change of food quality parameters influences herbivorous copepod reproductive success. *ChemBiochem*, 8: 1146–1153

Wichard, T., S.A. Poulet, C. Halsbandlenk, A. Albaina, R. Harris, D. Liu and G. Pohnert, 2005. Survey of the chemical defence potential of diatoms: screening of fifty species for α, β, γ, δ-unsaturated aldehydes. *J. Chem. Ecol*., 31: 949–958

Yool, A. and T. Tyrrell, 2003. Role of diatoms in regulating the ocean's silicon cycle. *Glob. Biogeochem. Cycl*., 17: 1–21

Yuan, Y., Y.U. Zhiming, X. Song and X. Cao, 2017. Temporal and spatial characteristics of harmful algal blooms in qingdao waters, China. *Chin. J. Oceanol. Limn*., 35: 1–15

[Received 17 Apr 2019; Accepted 13 May 2019; Published (online) 20 Aug 2019]