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Influence of UVB radiation on the lethal and sublethal toxicity of dispersed crude oil to planktonic copepod nauplii

Rodrigo Almeda [a, b, *], Tracy E. Harvey [a], Tara L. Connelly [a, c], Sarah Baca [a, d], Edward J. Buskey [a]

[a] Marine Science Institute, University of Texas at Austin, Port Aransas, 78373 TX, USA
[b] Centre for Ocean Life, Technical University of Denmark, Denmark
[c] Department of Ocean Sciences, Memorial University of Newfoundland, St. John's NL A1C 2S7, Canada
[d] College of Sciences, University of Texas at El Paso, El Paso, 79902 TX, USA

HIGHLIGHTS

- Dispersed crude oil and dispersant Corexit 9500 are highly toxic to copepod nauplii.
- UVB radiation substantially increases the toxicity of crude oil to copepod nauplii.
- Dispersed crude oil cause sublethal effects on copepod nauplii, such as reduced growth, development and swimming activity.

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ABSTRACT

Toxic effects of petroleum to marine zooplankton have been generally investigated using dissolved petroleum hydrocarbons and in the absence of sunlight. In this study, we determined the influence of natural ultraviolet B (UVB) radiation on the lethal and sublethal toxicity of dispersed crude oil to naupliar stages of the planktonic copepods *Acartia tonsa*, *Temora turbinata* and *Pseudodiaptomus pelagicus*. Low concentrations of dispersed crude oil (1 μL L^{-1}) caused a significant reduction in survival, growth and swimming activity of copepod nauplii after 48 h of exposure. UVB radiation increased toxicity of dispersed crude oil by 1.3–3.8 times, depending on the experiment and measured variables. Ingestion of crude oil droplets may increase photoenhanced toxicity of crude oil to copepod nauplii by enhancing photosensitization. Photoenhanced sublethal toxicity was significantly higher when *T. turbinata* nauplii were exposed to dispersant-treated oil than crude oil alone, suggesting that chemical dispersion of crude oil may promote photoenhanced toxicity to marine zooplankton. Our results demonstrate that acute exposure to concentrations of dispersed crude oil and dispersant (Corexit 9500) commonly found in the sea after oil spills are highly toxic to copepod nauplii and that natural levels of UVB radiation substantially increase the toxicity of crude oil to these planktonic organisms. Overall, this study emphasizes the importance of considering sunlight in petroleum toxicological studies and models to better estimate the impact of crude oil spills on marine zooplankton.

1. Introduction

After a marine oil spill, crude oil is affected by a variety of abiotic and biotic processes that determine the fate and impact of petroleum pollution in marine ecosystems (National Research Council, 2003). One of the major abiotic factors affecting spilled crude oil is sunlight, mainly ultraviolet (UV) radiation (Garrett et al., 1998; Lee, 2003; Guipeng et al., 2006; Fathalla, 2007). UV radiation not only promotes photochemical degradation of crude oil (e.g. photo-oxidation) (Garrett et al., 1998; Lee, 2003), but may also increase the toxicity of petroleum pollution to marine organisms (Boese et al., 1997; Pelletier et al., 1997; Barron et al., 2003; Duesterloh et al., 2003; Almeda et al., 2013a). Some crude oil compounds, such as alkenes and polycyclic aromatic hydrocarbons (PAHs), strongly absorb UV light, inducing photo-enhanced toxicity of crude oil (i.e.

* Corresponding author. Centre for Ocean Life, Technical University of Denmark, National Institute for Aquatic Resources, Kavalergården 6, Charlottenlund 2920, Denmark.
E-mail addresses: roal@aqua.dtu.dk, ralmeda2010@live.com (R. Almeda).

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increased crude oil toxicity in the presence of light) (Landrum et al., 1987; Kagan et al., 1990; Arfsten et al., 1996; Nicodem et al., 1997). Photo-enhanced toxicity may be caused by (1) photosensitization, where bioaccumulated petroleum compounds act as photoreceptors and transfer light energy to other surrounding biomolecules generating reactive oxygen species that cause cell damage, and/or (2) photomodification, where petroleum hydrocarbons are chemically transformed into more toxic compounds (Landrum et al., 1987; Kagan et al., 1990; Arfsten et al., 1996; Nicodem et al., 1997). However, most studies that have investigated the toxicity of crude oil were performed under artificial, fluorescent light, disregarding the influence of UV radiation on the toxicity of crude oil to marine organisms (Arfsten et al., 1996).

Among marine organisms, copepods are the dominant component of mesozooplankton communities (Longhurst, 1985; Humes, 1994). Planktonic copepods play a key role in pelagic systems, mediating the transfer of matter from primary producers to higher trophic levels and contributing to marine biochemical cycles (Banse, 1995; Alcaraz et al., 2010). Postembryonic development of planktonic copepods generally includes 11 larval stages, six naupliar stages (designated as nauplius I to nauplius VI, NI to NVI) and five copepodid stages (namely copepodid I to copepodid V, CI to CV) (Ferrari and Dahms, 2007). Copepod nauplii are considered the most abundant forms of metazoans on the planet (Björnberg, 1986; Fryer, 1986) and the main prey for many fish larvae (Last, 1980) contributing to the recruitment of commercially important fish stocks (Castonguay et al., 2008). However, despite the importance of copepods in marine systems, copepod nauplii are frequently neglected from environmental research (Björnberg, 1986), especially among eco-toxicological studies. In fact, we know little about the lethal and sublethal effects of dispersed crude oil and photenhanced toxicity on planktonic copepod nauplii, which is essential for understanding the impact of oil spills on copepod population dynamics and marine planktonic food webs.

During a marine crude oil spill, zooplankton are exposed to crude oil droplets generated by natural mixing or/and application of chemical dispersants (Forrester, 1971; Caneveri, 1978; Lichtenthaler and Daling, 1985; Delvigne and Sweeney, 1988). Dispersed crude oil droplets (1–100 μm) are frequently within the zooplankton prey size spectra (Hansen et al., 1984) and may be ingested by copepods and other planktonic organisms (Conover, 1971; Mackie et al., 1978; Gyllenburg, 1981; Lee et al., 2012; Almeda et al., 2014a, b, c). A recent study found that both feeding-current and ambush feeder copepod nauplii ingested dispersed crude oil (Almeda et al., 2014a). However, most toxicological studies on zooplankton have been conducted using the crude oil water soluble fraction (WSF), or certain mixed or individual petroleum hydrocarbons (Corner et al., 1976; Harris et al., 1977; Berdugo et al., 1997; Bejarano et al., 2006; Jiang et al., 2010, 2012) overlooking the influence of ingestion of crude oil droplets on the toxicity of crude oil to planktonic copepods. Similarly, photoenhanced toxicity of petroleum to marine animals has been mainly investigated using dissolved petroleum hydrocarbons (Boese et al., 1997; Pelletier et al., 1997; Barron et al., 2003) whereas the information on phototoxicity of crude oil to zooplankton is limited. Also, chemical dispersants (e.g. Corexit 9500A) used to treat oil spills, which can be toxic to planktonic organisms, particularly to larval stages (George-Ares and Clark, 2000; Goodbody-Gringley et al., 2013; Almeda et al., 2014b, d), may influence the photo-ehanhced toxicity of crude oil to aquatic animals (Barron et al., 2003). As far as we know, combined effects of chemical dispersants and UV radiation on crude oil toxicity to planktonic copepod nauplii have not been previously investigated.

Photoenhanced toxicity studies with crude oil or PAHs have usually been conducted using UVA and UVB simultaneously and, therefore, the relative contribution of different UV regions of the light spectrum to photoenhanced toxicity of petroleum is not well known (Boese et al., 1997; Pelletier et al., 1997; Barron et al., 2003; Duesterloh et al., 2003). In a previous study (Almeda et al., 2013a), we estimated the influence of UV/sunlight exposure on the toxicity of dispersed crude oil to natural copepod assemblages using 3 different light regimes: 1) the ambient full solar radiation spectrum (PAR + UVA + UVB), 2) the ambient full spectrum without UVB (PAR + UVA) and 3) no light (i.e. complete darkness). We found that PAR + UVA radiation did not significantly enhance the toxicity of dispersed crude oil to mesozooplankton after 48 h of exposure compared to dark incubations and that photo-enhanced toxicity of dispersed oil was due mainly to UVB exposure (Almeda et al., 2013a). Thus, although UVA or other light spectrum regions can enhance toxicity of petroleum hydrocarbons (Diamond et al., 2000; Barron et al., 2003), our previous results and those of other studies (Huovinen et al., 2001; Barron et al., 2003) suggest that UVB plays the dominant role in determining the magnitude of photoenhanced toxicity of crude oil to zooplankton. Understanding the effect of UVB radiation on the phototoxicty of crude oil is of particular interest because levels of UVB radiation have increased at the Earth's surface as a result of the stratospheric ozone depletion during the last decades (Madronich et al., 1998; McKenzie et al., 2007).

In the present study, we aim to 1) determine the toxicity of mechanically and/or chemically dispersed crude oil to copepod nauplii and 2) measure the influence of natural UVB radiation levels on toxicity of dispersed crude oil to copepod nauplii. To that end, we estimated the effects of dispersed crude oil on survival, growth rates and swimming behavior of copepod nauplli after 48 of exposure with and without natural UVB radiation. We used naupliar stages of the calanoid planktonic copepods *Temora turbinata*, *Acartia tonsa*, and *Pseudodiaptomus pelagicus* (Fig. 1). These species belong to some of the most representative copepod genera in estuarine and coastal subtropical and temperate waters, including the Gulf of Mexico, where these species are among the most common mesozooplankton (Razouls, 2005–2013). This study is particularly valuable for increasing our understanding of the potential impact of oil spills on zooplankton communities in the Gulf of Mexico, a region with a high risk for oil spills due to the intense oil production and transportation.

2. Material and methods

2.1. Experimental organisms

Zooplankton samples were collected from the Aransas Ship Channel near the University of Texas Marine Science Institute or from a nearby channel in Corpus Christi Bay (Port Aransas, Texas) using a plankton net (150 μm mesh, 50 cm diameter). Plankton samples from the Corpus Christi Bay channel were collected by towing the plankton net through the surface water, whereas samples from the Aransas Ship Channel were collected from surface waters by tying a plankton net to the University of Texas Marine Science Institute pier and allowing it to stream with the tidal current for approximately 5–10 min. Specimens of *A. tonsa* and *P. pelagicus* were isolated from samples collected in the Corpus Christi Bay Channel in July 2013. *T. turbinata* were isolated from zooplankton samples taken in September/October 2014 from the Aransas Ship Channel during flood tides from the Gulf of Mexico. Contents of collection buckets (cod ends) were poured in a cooler containing *in situ* unfiltered seawater until returning to the laboratory, where samples were lightly aerated.

Once in the laboratory, aliquots of the zooplankton samples were examined under a dissecting microscope and copepod adult stages of *T. turbinata*, *A. tonsa*, and *P. pelagicus* were identified and