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Research Note

New record of three species of the family Warnowiaceae (Dinophyceae) in the Gulf of California

Nuevos registros de tres especies de la familia Warnowiaceae (Dinophyceae) en el Golfo de California

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Abstract.- The naked marine dinoflagellates *Proterythropsis vigilans, Nematodinium armatum,* and *Nematodinium torpedo* are reported for the first time in the Gulf of California. The first and the third species are also recorded for the first time on the Pacific coast of Mexico. They were recorded during winter-spring in seawater at 20-26.5°C. *Nematodinium armatum* was the most frequent species. *Proterythropsis vigilans* was less frequent. The 3 species were found in phytoplankton net samples. No quantitative data are reported here. A short description is given for each species, and micrographs provide information about the main morphological characteristics.

Key words: Dinoflagellates, *Nematodinium, Proterythropsis,* Mexico

Introduction

Dinoflagellates are unicellular protists that exhibit great diversity of form and are one of the major constituents among marine phytoplankton. Dinoflagellates represent a major lineage of unicellular eukaryotes with unparalleled diversity and complexity in morphological features, molecular processes, nutritional modes, and symbioses with distantly related organisms (Taylor 1987, Hackett *et al*. 2004, Taylor *et al.* 2008). Because they are difficult to identify, unarmored dinoflagellates species have been poorly studied in Mexico (Hernández-Becerril & Bravo-Sierra 2004). Recent studies of this group have demonstrated that they are important component of the phytoplankton community, particularly during winter-spring (Gárate-Lizárraga *et al*. 2009, 2010, 2011; Gárate-Lizárraga 2011). Warnowiid dinoflagellates are among the more remarkable eukaryotes because they are unarmored phagotrophic dinoflagellates that possess highly elaborate ultrastructural systems: pistons, nematocysts, and ocelloids (Hoppenrath *et al*. 2009a); they have been poorly studied in the seas adjacent to Mexico (Gárate-Lizárraga *et al*. 2009, 2010). According to Fensome *et al.* (1993) and Hoppenrath *et al*. (2009a), the family Warnowiaceae comprises 5 genera: *Warnowia*, *Nematodinium*, *Proterythropsis*, *Greuetodinium*, and *Erythropsidinium*. Other genera such as *Nematopsides* and *Protopsis* are taxa of doubtful validity (Hoppenrath *et al*. 2009a). To more clearly outline the taxonomic challenges associated with understanding warnowiid diversity, Hoppenrath *et al*. (2009a) summarized the morphological features described for each genus. Nematocysts, ejectile organelles, have only been found in *Nematodinium* and *Proterythropsis*. This report describes the presence of *Proterythropsis vigilans* Marshall 1925, *Nematodinium armatum* (Dogiel 1906) Kofoid *et* Swezy 1921, and *N. torpedo* (Dogiel 1906) Kofoid *et* Swezy 1921 for the first time in the Gulf of California.

Materials and Methods

Bahía de La Paz is the largest bay on the peninsular side of the Gulf of California. The bay constantly exchanges water with the Gulf of California via a northern and a southern opening (Gómez-Valdés *et al*. 2003). The northern mouth is wide and deep (up to 300 m), while the southern mouth is straight and shallow and associated with a shallow basin about 10 m in depth and a large lagoon, the Ensenada de La Paz, connected to Bahía de La Paz by a narrow inlet (1.2-km wide and 4-km long) having an average depth of 7 m. A sampling station was located at 24.21°N, 110.31°W in the shallow basin of the southernmost region of the bay. Forty-two surveys for collecting phytoplankton samples were done at a sampling station in Bahía de La Paz, Baja California Sur, Mexico from January 2009 to April 2012 (Fig. 1, Table 1). Surface horizontal tows and vertical hauls (15 m depth) were carried out with a 20 µm mesh net with a mouth of 50 cm in diameter. Each net sample was immediately preserved with

Table 1. Occurrence and temperature data of *Proterythropsis vigilans*, *Nematodinium armatum*, and *Nematodinium torpedo* in 16 samplings performed in the Bahía de La Paz from January 2009 to April 2012 / Datos de ocurrencia y temperatura de *Proterythropsis vigilans, Nematodinium armatum* y *Nematodinium torpedo* en 16 muestreos realizados en la Bahía de La Paz de enero de 2009 a abril de 2012

Sampling dates	Surface net samples	Vertical net samples	T (°C)
23/01/09	*N. armatum* (1)	*N. torpedo* (2)	20
		N. armatum (1)	
19/02/09	*N. torpedo* (2)	*N. armatum* (1)	21
26/02/09	*N. armatum* (2)	*N. torpedo* (1)	21
05/03/09	*N. armatum* (1)	*N. torpedo* (1)	22
	P. vigilans (1)	*P. vigilans* (3)	
17/03/09	*N. armatum* (2)	*N. torpedo* (1)	22.5
24/06/10	*P. vigilans* (3)	*P. vigilans* (1)	25
25/06/10	*N. armatum* (1)	*N. torpedo* (1)	25
29/07/10	*N. armatum* (1)	*N. armatum* (5)	26.5
13/12/10	*N. armatum* (1)	*N. armatum* (2)	21
23/01/11	*N. armatum* (2)	*N. armatum* (2)	20
28/04/11	*N. armatum* (1)	*N. armatum* (5)	23
		N. torpedo (2)	
25/05/11	*N. armatum* (1)	*N. armatum* (1)	24
12/12/11	*N. armatum* (2)	*N. armatum* (3)	21.5
26/01/12	*N. armatum* (1)	*P. vigilans* (2)	20
24/02/12	*N. armatum* (2)	*N. armatum* (3)	22
19/04/12	*N. armatum* (3)	*N. armatum* (4)	24
		N. torpedo (2)	

acid Lugol's solution. Other subsamples were taken for live phytoplankton observations. Cell counts were made in 5 mL settling chambers under an inverted phase contrast microscope (Hasle 1978). Sea-surface temperature was recorded using a bucket thermometer. Microscopic images were taken under a compound microscope.

Results and Discussion

Three species of the family Warnowiaceae were found in Bahía de La Paz from January 2009 through April 2012: *Proterythropsis vigilans* (Fig. 1. A-B), *Nematodinium armatum* (Fig. 1. C-D) and *Nematodinium torpedo* (Fig.1. H-K). These species were very scarce and were observed only in net phytoplankton samples (Table 1). Therefore, no quantitative data are presented. Ten specimens of *Proterythropsis vigilans* were observed in the net phytoplankton samples collected on 5 March 2009, 24 June 2010, and 26 January 2012. *Proterythropsis vigilans* cells are small and ovoid (Fig. 1 A-B), with a left-handed spiral girdle making one and a half turns continuing onto a posterior ventral tentacle or prod (Fig. 1 A), but it is immobile and called a 'posterior extension'. A compact posterior ocelloid is present, containing a red pigment mass with yellow core and pyriform lens with concentric rings. The nucleus is large and anterior. Cells are 35-63 µm long and 20-35 µm wide. The specimens swim very quickly in a straight line. Some specimens of *P. vigilans* have 2-3 extremely large nematocysts with a reduced or retracted posterior extension (Fig. 1B). There are few records of *Proterythropsis vigilans* in the literature. This species was described using specimens from Millport (Marshall 1925) and it is distributed elsewhere in the Clyde area and Liverpool of the United Kingdom (Lebour 1925, Dodge 1982). It was also reported for the Eurasian Arctic

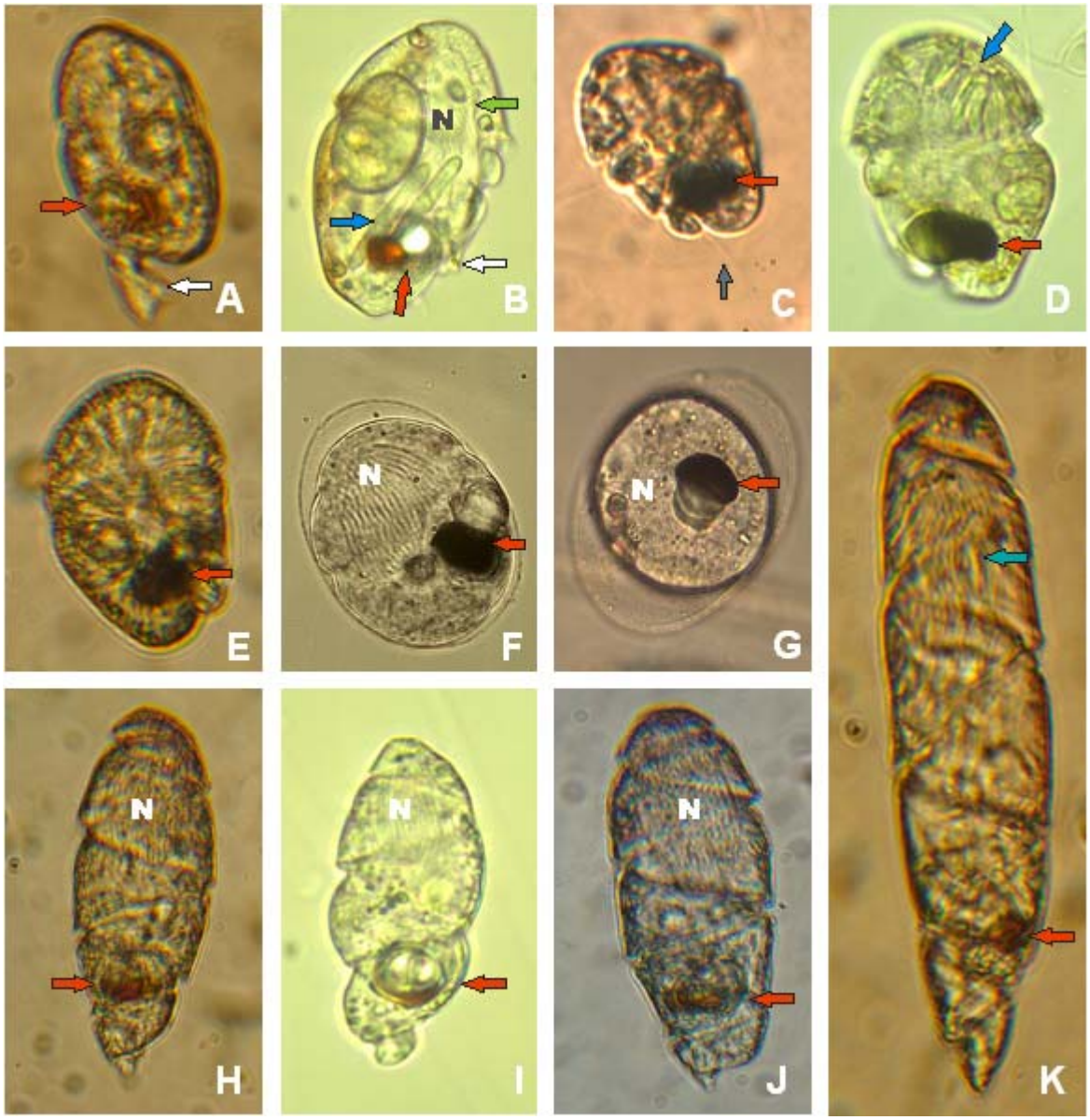

Figure 1: A-K. Microphotographs of 3 species in Bahía de La Paz belonging to the family Warnowiaceae. A. Specimen of *Proterythropsis vigilans* showing the ocelloid (red arrow), and the posterior extension (white arrow). B. Another specimen of *Proterythropsis vigilans* showing the ocelloid (red arrow), two extremely large nematocysts (blue arrow), the nucleus (N), several green structures (green arrow), and the posterior extension retracted (white arrow). C-E. Three different specimens of *Nematodinium armatum*. C. Specimen of *Nematodinium armatum* showing the ocelloid (red arrow), and the flagellum (gray arrow). D. Specimen of *Nematodinium armatum* showing the ocelloid (red arrow) and nematocysts (blue arrow). F-G. Pellicle or temporary cysts of *Nematodinium* (?) showing the ocelloid and the nucleus (N). H-K. Different specimens of *Nematodinium torpedo*; red arrows indicate the ocelloid and N the nucleus / A-K. Microfotografías de 3 especies diferentes de la familia Warnowiaceae encontrados en la Bahía de La Paz. A. Espécimen de *Proterythropsis vigilans* mostrando el oceloide (flecha roja) y el proceso de extensión (flecha blanca). B. Otro espécimen de *Proterythropsis vigilans* mostrando el oceloide (flecha roja), dos nematocistos extremadamente grandes (flecha azul), el núcleo (N), algunas estructuras verdes (flecha verde) y el proceso de extensión retraído (flecha blanca). C-E. Tres diferentes especímenes de *Nematodinium armatum*. C. Espécimen de *Nematodinium armatum* mostrando el oceloide (flecha roja) y el flagelo (flecha gris). D. Espécimen de *Nematodinium armatum* mostrando el oceloide (flecha roja) y los nematocistos (flecha azul). F-G. Quistes peliculares o temporales de *Nematodinium* (?) mostrando el oceloide y el núcleo. H-K. Diferentes especímenes of *Nematodinium torpedo*; la flecha roja indica el oceloide y N el núcleo

(Okolodkov 1997), in the Kattegat in the North Sea (Hansen & Larsen 1992), and in the coastal waters of Japan[1]. It was found in the mouth of the St. Lawrence River and Gulf of St. Lawrence, Canada (Bérard-Therriault *et al*. 1999). More recently, *P. vigilans* has been recorded in the Helgoland and Sylt islands, the North Sea (Hoppenrath *et al*. 2009b). This finding of *P. vigilans* represents the first record of this species not only in the Gulf of California, but also in the whole Mexican Pacific.

In 16 samplings, 49 specimens of *Nematodinium armatum* were found (Fig. 1 C-E, Table 1). The specimens fit well with the diagnosis and description of Kofoid & Swezy (1921). This is summarized as: cells with the epicone larger than the hypocone; medium-sized species; body elliptical with torsion; cingulum makes 1.5 turns; sulcus extending from near to the apex spirally in descending left spiral of about 1.2 turns; large and ovoid nucleus is located at the anterior end of the cell. Few yellow chloroplasts are present in some specimens, scattered throughout the cell, which coincide with those photosynthetic specimens of *N. armatun* reported by Hoppenrath *et al*. (2009). The ocelloid is composed of a single lens near to the melanosome with concentric rings and is posteriorly located (Fig. 1. C-E); nematocysts lie in a group of 8-12 near the nucleus (Fig. 1D); cells range from 75-105 µm length and 43-65 µm width. This species has 3 different size ranges, one in each of three geographic regions; it is suspected that there is homonomy (Steidinger & Tangen 1997). *Nematodinium armatum* is rarely reported on both sides of the Pacific Ocean. This species was found in net phytoplankton samples (80 m vertical net haul) collected 6 and 4 nautical miles (nm) off La Jolla, California on 20 and 27 July 1917 (Kofoid & Swezy 1921). Four pellicle cysts (or temporary cysts), probably of *Nematodinium*, were found in net phytoplankton samples (Fig. 1. F-G). This species is common in the Mediterranean Sea, from where it was described (Dogiel 1906). It has also been reported off Plymouth (United Kingdom), Italy, the German Bight at Helgoland and Sylt, and the Oslo Fjord (Dodge 1982, Hoppenrath *et al*. 2009b). In addition, it has been reported in coastal waters of Japan (Takayama 1984, Ono 1990a), River Derwent (Tasmania), and Port Phillip (Victoria, Australia) (Hallegraeff *et al*. 2010), and in Great Pond, Barnegat Bay (New Jersey), coastal New Jersey (Hulburt 1957), in the Gulf of Mexico (Steidinger *et al*. 2009), and in Bahía Magdalena, on the west coast of Baja California peninsula (Gárate-Lizárraga *et al*. 2007). This finding of *N. armatum* represents the first record of this species in the Gulf of California.

Twelve specimens of *Nematodinium torpedo* were found in net phytoplankton samples collected on 23 January 2009 to 19 April 2012 (Table 1). It is a large species with elongated, fusiform body (Fig. 1. H-K); the girdle is a descending left spiral of 2.25 turns; the sulcus has nearly 2 turns, extending almost from apex to antapex. The ocelloid with its red melanosome is situated on the left side, above the junction of girdle and sulcus (Fig. H-K). The nucleus is elongate in the anterior part of the body and is about half as broad as long and shows clearly visible chromatin strands. Many nematocysts were observed in 2 irregular bands in the region of the nucleus (Fig. 1K). No chloroplasts were observed. Cells are 80-102 µm long and ~25-42 µm wide. The color of the cytoplasm was grayish-green, sometimes reddish. There are few records of *Nematodinium torpedo* in the literature. This species was first described using one individual taken in a net vertical haul (from 100 m to the surface) on 24 June 1907, 2 nm off La Jolla, California, in surface temperature of about 16.5°C (Kofoid & Swezy 1921). This species has also been recorded in Australia (Wood 1963), in the northwestern African upwelling region (Elbrächter 1979), in Japan (Ono 1990b), and in the Gulf of Mexico (Steidinger *et al*. 2009). This finding of *N. torpedo* represents the first record of this species not only in the Gulf of California, but also in the Mexican Pacific.

Monitoring live microalgae species that form red tides has been done since 2000 in Bahía de La Paz. Live phytoplankton samples allowed us to identify many naked dinoflagellates (Gárate-Lizárraga *et al*. 2004, 2009, 2011). Based on our findings, the ecological importance of this group has been underestimated because the fixative solutions used to preserve the samples destroy important morphological characteristics of the cells. Gárate-Lizárraga *et al*. (2009, 2010, 2011, this study) reported that naked dinoflagellates are an important component of phytoplankton during upwelling conditions observed during winter–spring in Bahía de La Paz.

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[1]< http://www.geocities.jp/takayama_haruyoshi/HAB_Art/Nematopsides_vigilans.html>

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