



## FIRST RECORD OF *Erythropsidinium agile* (GYMNODINIALES: WARNOWIACEAE) IN THE MEXICAN PACIFIC

**Primer registro de *Erythropsidinium agile* (Gymnodiniales: Warnowiaceae) en el Pacífico Mexicano**

**RESUMEN.** Se registra por primera vez *Erythropsidinium agile*, un dinoflagelado de la Familia Warnowiaceae para el Pacífico Mexicano, dentro de Bahía de La Paz (Golfo de California). Se observaron 26 ejemplares de *E. agile*, principalmente en muestras de fitoplancton de red para el período de estudio (Junio, 2006 a Junio, 2010). En muestras de botella se estimaron densidades entre 80 y 1000 cél. L<sup>-1</sup>. Los ejemplares de *E. agile* mostraron gran variación en forma, tamaño y coloración; se presentaron principalmente en el período invierno-prIMAVERA, cuando la columna del agua está homogénea, a temperaturas entre 19 y 22°C y rica en nutrientes.

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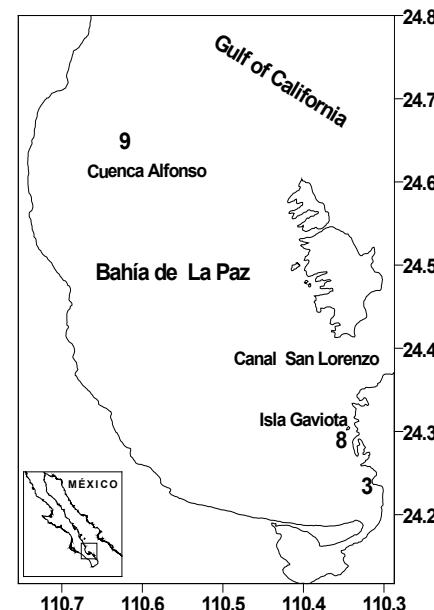
Along the Mexican Pacific coast dinoflagellates have been studied from floristic and taxonomic points of view. Recently, red tide-forming dinoflagellates have received more attention (Gárate-Lizárraga *et al.*, 2004; 2006, 2008; Alonso-Rodríguez & Ochoa, 2004). Naked dinoflagellates have been less studied, and their contribution to productivity of phytoplankton may have been underestimated (Hernández-Becerril & Bravo-Sierra, 2004; Okolodkov & Gárate-Lizárraga, 2006). Naked dinoflagellates have been recorded and currently the list of species is growing because there are more observations on live phytoplankton (Gárate-Lizárraga & Verdugo-Díaz, 2007; Gárate-Lizárraga *et al.*, 2007, 2009, 2010). Naked dinoflagellates are normally deformed or destroyed by sampling nets, fixative agents, and during storage with traditional preservation solutions used in routine phytoplankton sampling. Hence, using live phytoplankton renders an opportunity for studies of naked dinoflagellates species.

Warnowiid dinoflagellates are among the more remarkable eukaryotes because they possess highly elaborate ultrastructural systems: pistons, nemato-cysts, and ocelloids (Hoppenrath *et al.*, 2009). They have been poorly studied in the seas adjacent to Mexico (Gárate-Lizárraga *et al.*, 2009). According to Fensome *et al.* (1993) and Hoppenrath *et al.* (2009), this family has five genera: *Nematodinium* Kofoid

et Swezy, 1921, *Proterthyropsis* Kofoid et Swezy, 1921, *Warnowia* Lindemann, 1928, *Greuetodinium* Loeblich III, 1980, and *Erythropsidinium* P.C. Silva, 1960. Ten species of *Erythropsidinium* have been described from warm and temperate seas. However, a taxonomical study based on the changes in structure, position, and coloration of the ocelloid in the course of the cell division or individual development revealed that some species had different morphotypes (Elbrächter, 1979). At present the valid species currently considered to belong to this genus are: *E. agile* (Hertwig, 1884) P.C. Silva, 1960, *E. cochlea* (Schütt, 1895) P.C. Silva, 1960, *E. extrudens* (Kofoid et Swezy, 1921) P.C. Silva, 1960, and *E. minus* (Kofoid et Swezy, 1921) P.C. Silva, 1960. For the Mexican Pacific, the naked dinoflagellate *Erythropsidinium agile* is reported here for the first time, and information about its morphology and ecology is also provided.

The study area comprised Bahía de La Paz in the Gulf of California (Sampling stations: 3, 8, and 9, Fig. 1). Sampling station 3 is located at 24°21' N; 110°31' W offshore of the PEMEX storage facility in the shallow basin of the southern end of Bahía de La Paz. Sampling station 8 is located at 24°18.39' N, 110°20.80' W near Isla Gaviota. Sampling station 9 is located at Cuenca Alfonso (24°39' N, 110°36' W).

Thirty six phytoplankton samples were collected



**Figure 1.** Sampling stations in the Bahía de La Paz. 3, PEMEX; 8, off Isla Gaviota; 9, Cuenca Alfonso.

monthly at sampling station 3 in Bahía de La Paz from January 2009 to June 2010 (Fig. 1) with surface and vertical (from the 15 m depth) tows using a 50 cm diameter, 20 µm mesh hand net. A portion of each tow was immediately fixed with acid Lugol solution and later preserved in 4% formalin. Other sub-samples were taken for live phytoplankton observations. Sixteen surface water samples were also collected for identification and cell counting. At Station 8, sea water samples for phytoplankton analysis were collected at 0, 15 and 30 m depth with van Dorn bottles from June 2006 to May 2007 (Fig. 1). At Station 9, six vertical net hauls were conducted from a 60 m depth to the surface from February through June 2010 (Fig. 1). Sea water temperature was recorded in sampling stations 3 and 8 using a bucket thermometer. Temperature and salinity at Station 9 (Cuenca Alfonso) were registered with a data recorder (SeaBird 19 CTD). Cell counts were made in 2 ml (St. 3) and 25 ml (St. 8) settling chambers under an inverted phase-contrast Zeiss microscope. An Olympus CH2 compound microscope was used for cells measurements. A SONY Cyber-shot camera (8.1 MP) was used for image capture. Images 5, 9, 13 and 17 were taken using a Leica microscope.

#### **Systematic account of *Erythropsidinium agile* (Hertwig, 1884) P.C. Silva, 1960:**

Division: Dinoflagellata (Butschli, 1885) Fensome *et al.*, 1993; Subdivision: Dinokaryota Fensome *et al.*, 1993; Class: Dinophyceae Pascher, 1914; Subclass: Gymnodiniphycidae Fensome *et al.*, 1993; Order: Gymnodiniales Apstein, 1909; Family: Warnowiaceae Lindemann, 1928.

Basynonym: *Erythropsis agilis* Hertwig, 1855, p. 204-212, Pl. 6.

Synonymous: *Erythropsis cornuta* (Schütt, 1895) Kofoid et Swezy, 1921, p. 489-491, Pl. 12 fig. 129, textfig. SS, 1; *E. hispida* Kofoid et Swezy, 1921, p. 499-501, Pl. 12 fig. 127, textfig. SS, 2; *E. labrum* Kofoid et Swezy, 1921, p. 501-503, Pl. 12 fig. 132, textfig. SS, 9; *E. pavillardi* Kofoid et Swezy, 1921, p. 505-507, Pl. 12 fig. 133, textfig. SS, 5; *E. richardi* Kofoid et Swezy, 1921, p. 508-510, Pl. 12 fig. 134, textfig. SS, 10; *E. scarlatina* Kofoid et Swezy, 1921, p. 510-512, Pl. 12 fig. 128, textfig. RR, 2, SS, 4; *Erythropsidinium cornutum* (Schütt) P. C. Silva 1960, p. 23; *E. hispidum* (Kofoid et Swezy, 1921) P. C. Silva 1960, p. 23; *E. labrum* (Kofoid et Swezy, 1921) P. C. Silva 1960, p. 23; *E. pavillardi* (Kofoid et Swezy, 1921) P. C. Silva 1960, p. 23; *E. richardi* (Kofoid et Swezy, 1921) P. C. Silva 1960, p. 23; *E. scarlatinum* (Kofoid et Swezy, 1921) P. C. Silva 1960, p. 23.

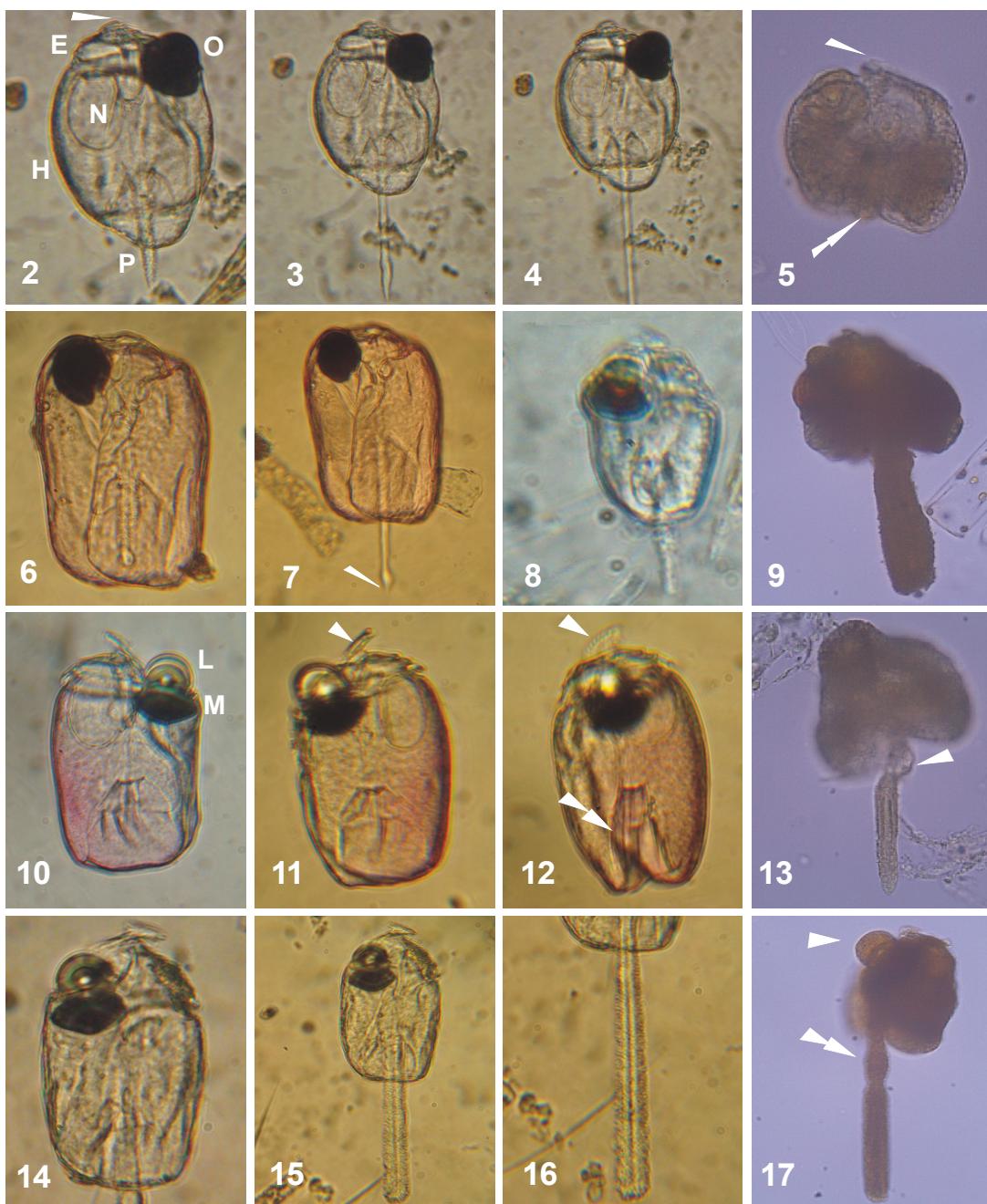
The species of the Warnowiaceae family have been poorly studied and particularly so in Mexico, where only one species of *Nematodinium* and three species of *Warnowia* have been reported (Gárate-Lizárraga *et al.*, 2007; 2009). *Erythropsidinium agile*

is a member of Warnowiaceae, which is a gymnodinioid dinoflagellate characterized by one or several elaborate eyespots, a light-receiving organelle (ocelloid), which is located at the left side of the cell seen in ventral view (Fig. 2). This species has an episome that is shorter (less than 1/4 the cell size) than the hyposome (Fig. 2). The sulcus of *E. agile* extends almost straight from near the apex to the antapex, with a small apical horn above its proximal end. The ventral recess is a deep excavation opening ventro-posteriorly and sheltering the basal end of the piston (Fig. 12). The cingulum originates at the proximal end of the sulcus and descends in a left spiral of about 1.5 turns around the cell. The narrow groove originates from the base of the apical horn (Takayama, 1985). The nucleus is ellipsoidal and usually located at the right side of the cell (Fig. 2).

The piston (prod, dart, or tentacle) is capable of incessant rhythmic contractions (Greuet, 1967). The piston is only present in two warnowiid genera, *Erythropsidinium* and *Greuetodinium* (Greuet, 1987). The piston projects from a postero-ventral invagination in the hyposome (Kofoid & Swezy, 1917; Greuet, 1970; 1977; Hoppenrath *et al.*, 2009). Different cells of *E. agile* show the form and size variation of the piston (Figs. 2-17). The cells presented in Figs. 2-9 and 13-17 show the piston in different phases of elongation. The distal end of the piston may show a terminal stylet. Contrasting with the results found by Gómez (2008), we observed only one specimen of *E. agile* with a very small stylet (Fig. 7). The length and thickness of the piston were highly variable among the specimens (Figs. 2-17). According to the authors mentioned immediately above, the piston of *Erythropsidinium* species causes movement. Our observations of live cells of *E. agile* agree with the findings of Hoppenrath & Saldarriaga (2008) that *E. agile* cells move more quickly than the movement of the piston because there are small phytoplankton species surrounding the cells; however, no feeding behavior was observed in this study.

**Abundance, morphological characteristics, and cell size of *E. agile*:** The specimens examined match previous description for this species. Fourteen live specimens of *E. agile* were found in net phytoplankton samples from Bahía de La Paz obtained between January and May 2009. In bottle samples collected in February, March, and May 2009 densities ranged from 500 to 1000 cells L<sup>-1</sup>. No cells of *E. agile* were found from June 2009 to June 2010. At the Sampling station 8, a density of 80 cells L<sup>-1</sup> was found only in September 2006 at a depth of 15 m. Twelve cells were observed in phytoplankton net samples collected from February through June 2010 at Station 9.

**Cell size:** ranges from 80 to 140 µm in lenght (excluding piston) and from 40 to 60 µm in width. Variation in size, shape, and color was clearly visible among the specimens (Figs. 2-17). Most of the differences are developmental stages of cells between



**Figures 2–17.** Microphotographs of *Erythropsidinium agile*. The same specimen (Figs. 2–4) showing the piston (P) in different phases of elongation. Fig. 2 shows the ocelloid (O) in the left half of the cell (ventral view), the nucleus (N) in the right half of the cell, the episoma (E), and the hyposoma (H). The arrowhead in Fig. 2 shows the apical horn. Double arrowhead in Fig. 5 shows a retracted piston; single arrowhead shows the lanceolate apical extension of the cell. Figs. 6 and 7 show two red cells in dorsal view. At the end of the piston, arrowhead in Fig. 7 shows a very thin stylet. Fig. 8 a cell of *Erythropsidinium agile* recorded in surface water samples collected in May. Figs. 10–12 show square-like cells with a well-developed ocelloid (L= lens; M= melanoma). Fig. 12 shows the piston retracted into the cell. Arrowheads in these figures show the lanceolate apical extension of the cells. Arrowhead in Fig. 13 shows a papillae in the piston. Figs. 14–16 are a translucent specimen of *Erythropsidinium agile* with a well-developed ocelloid and piston. Figs. 5, 9, 13, and 17 are cells collected in Cuenca Alfonso. These specimens present a well-developed piston. Fig. 17 shows the ocelloid; double arrowhead indicates a thickening or papillae in the piston.

subsequent cell divisions. Two specimens collected in January 23, 2009 were ovoid and translucent; these had a brownish-black ocelloid, and a very large piston (Figs. 2 and 3). They moved in circles with the piston moving very quickly. The contour of the cell in the specimens had an anterior lamella or horn (Figs. 2, 3, 4, 6, and 7). In some specimens, the lamella was pointed and emerged from a peduncle with a lanceolate shape (Figs. 10–12 and 14–16). Chloroplasts and food vacuoles were not observed. The seven cells of *E. agile* found in the two collections in February (19 and 26) 2009 were square-like, rather than round pink colored, contained a brownish-black ocelloid, and the piston moved very quickly (Figs. 6–8). A specimen collected in March 19, 2009 was observed for ~2 h; there was no movement of its piston. Three specimens found in April 14, 2009 were pink-colored in part and translucent in part (Figs. 10–12) and were observed for ~1.5 h without movement of their pistons. In these cells, the base of the ocelloid was black (melanoma) and the upper part was translucent, resembling a lens. Two cells observed in May 14, 2009 were similar in shape to those collected in April; the ocelloid was also similar in shape, but their bodies were colorless and had a well-developed piston (Figs. 14–16). A single cell was found in May 22, 2009 (Fig. 8). The cells fixed in Lugol solution were easily recognizable. Figures 5, 9, 13, and 17 are cells collected in February 5, March 19, April 19, and May 21, 2010 respectively, at Station 9. The specimens found in March 8, and June 19 in this sampling station is not illustrated.

**Worldwide distribution of *E. agile*:** According to previous records, this species seems to have a cosmopolitan distribution. First reports are based on the specimens from the eastern Atlantic Ocean, particularly, the Mediterranean Sea, in the Italian and French coastal waters (Hertwig, 1885; Schütt, 1895; Pavillard, 1905; Fauré-Fremiet, 1914; Greuet, 1970; 1973). More recent records in that area were made by Delgado (1990) and Margalef (1995) in the Spanish Mediterranean coastal waters, and by Gómez *et al.* (2009) in Marseille, France. Some specimens from NW Africa are connected with upwellings (Margalef, 1975; Elbrächter, 1979). More recently, *E. agile* was reported near the Canary Islands (Gil-Rodríguez *et al.*, 2003) and in the Neretva River delta, Croatia (Jasprica & Hafner, 2005). For the western Atlantic Ocean, specimens have been reported from the Gulf Stream off Ft. Pierce, Florida (Hoppenrath *et al.*, 2009) and in the South Atlantic in the estuary of Rio de La Plata (Kogan, 2005). In the eastern Pacific Ocean, specimens have been collected off California (Kofoid & Sewzy, 1921; Kimor & Reid, 1989). In the western Pacific, specimens were collected near Japan (Takayama, 1985). Gómez (2008) reports findings in the vicinity of the Kuroshio and Oyashio currents, off the Philippines in the Celebes, Sulu, and the South China Sea, and in the SE Pacific Ocean (Chile and Peru). In general, the species

is broadly distributed in all the regions of the open Pacific Ocean from 34°N to 33°S, except for the cold waters of the Subarctic Oyashio Current.

**Distribution in the Mexican Pacific:** Our finding of *E. agile* represents the first record of this species not only in the Gulf of California, but in the Mexican Pacific.

**Ecological notes:** In general, *E. agile* occurred at temperatures ranging between 19 °C and 28 °C. At Cuenca Alfonso, samples were collected when water was 22.5–25.6 °C and the salinity was 34.75–35 psu. At Sampling station 8, samples were collected when water was 28 °C (September). Gómez (2008) reports that water temperature ranged from 15 °C to 30 °C, but this does not seem to be an important factors favoring the presence of this species. He mentions that the highest abundances of *E. agile* are found in the transition between the warm Kuroshio Current (18–22 °C) and the highly productive continental slope off southern Japan.

We found the highest density of specimens in water ranging from 19 °C to 22 °C when the water-column was homogeneous and during the periods of upwelling. Elbrächter (1979) and Margalef (1975) found this species off NW African in the areas of upwelling region. Gárate-Lizárraga *et al.* (2009; 2010) reported that heterotrophic dinoflagellates had become an important component of phytoplankton during upwelling conditions in Bahía de La Paz.

Monitoring microphytoplankton by studying live specimens as well as Lugol-fixed samples has been performed since 2000 in Bahía de La Paz (Gárate-Lizárraga *et al.*, 2004). Since that time *E. agile* has been observed at the times listed in this report from water samples collected at the surface and at 15 m depth. At Cuenca Alfonso, we cannot say with certainty the depth where *E. agile* was collected because vertical net hauls started from 60 m. Gómez (2008) states that nearly all the specimens of *E. agile* were collected in the upper 90 m, above the deep chlorophyll maxima because this species inhabits the illuminated layer where its light-receptor is more efficient. Any further investigations of the vertical distribution of naked dinoflagellates and their ecological significance will require sampling at deeper waters.

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