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Trachelomonas from temporary ponds on Menorca Island.

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ABSTRACT

***Trachelomonas* from temporary ponds on Menorca Island.**

A taxonomic study of the genus *Trachelomonas* (Euglenophyta) was undertaken in temporary ponds on Menorca Island, Spain, called "*basses*". Among the 13 studied ponds, this genus was found in only one, called Verda B. A light and scanning electron microscope study of the samples taken from this body enabled the identification of 28 taxa, 26 of which were documented by SEM micrographs, including different views and details of the lorica. All the taxa described are reported as new for this region, while 18 are new for Spain. *T. granulosa* var. *crenulatocollis*, *T. nigra*, *T. pseudofelix*, and *T. raciborskii* var. *nova* f. *minor* are examined and photographed by SEM for the first time. *T. duquei* var. *minor* is proposed as a new variety.

KEYWORDS: Adiversity, *Trachelomonas*, temporary ponds, Menorca Island, Spain.

RESUMEN

Trachelomonas *de estanques temporales de la isla de Menorca.*

Un estudio taxonómico sobre el género Trachelomonas *(Euglenophyta) fue realizado en estanques temporales de la isla de Menorca, España, llamados "*basses*". De los 13 cuerpos estudiados, este género fue encontrado sólo en uno de ellos, llamado Verda B. El análisis con microscopio óptico y electrónico de barrido de las muestras tomadas de este estanque nos permitió la identificación de 28 taxones, de los cuales 26 se ilustran con micrografías MEB, incluyéndose diferentes vistas y detalles de las lóricas. El total de las entidades descritas constituyen nuevas citas para esta región y 18 lo son para España.* T. granulosa *var.* crenulatocollis*,* T. nigra*,* T. pseudofelix *y* T. raciborskii *var.* nova *f.* minor *son examinados y fotografiados con MEB por primera vez.* T. duquei *var.* minor *es propuesta como una nueva variedad.*

PALABRAS CLAVES*: diversidad,* Trachelomonas*, estanques temporales, Isla de Menorca, España.*

INTRODUCTION

Menorca, a small island located in the northern part of the Balearic Islands, Spain, was declared a biosphere reserve by UNESCO in 1993. It is characterized by a great biodiversity. Almost all the Mediterranean habitats are represented in its 702 km^2, including temporary ponds, called "*basses*" in the local terminology, which are characterized by their small extension and shallow

waters. As they are scarce, fragile, and unique, they were considered of priority interest for conservation (The Habitats Directive, 9243CEE). These ponds are dependent on rain for water; however, the shortage of water makes them dynamic, variable ecosystems, which defines them as biodiversity hotspots. The geology of the island substrates introduces spatial diversity in the chemical characteristics of these bodies of water (Pretus et al., 2010), a determining factor for organisms that colonize them, particularly for the most sensitive ones, therefore they have the highest value as an environmental indicator. Their small size, shallowness, and abundance make temporary ponds vulnerable ecosystems, suffering the threat of human, who are altering their conservation status to the point of causing them to disappear.

At present, only three scientific studies have focused on microalgae from Menorca. In the first one, 13 taxa were recorded within the Flagellophyceae group (currently Euglenophyceae), but no data were included from temporary ponds (Margalef, 1952). The second one was dedicated to the total phytoplankton from Menorca temporary ponds (Moyá & Conforti, 2009); while, in the third one, the research was carried out on cyanobacteria, microalgae, and testaceous amoebas (Moyá, 2013).

This algae taxonomic study shows that one of these ponds, called "bassa" Verda B presents favorable conditions for the growth of euglenoids, mainly species belonging to the genus Trachelomonas. Our study exhibits a large number of taxa of this genus, much higher than the figure found in previous papers, not only for Menorca but for the Balearic archipelago. During the analysis of the "bassa" Verda B samples, 28 taxa belonging to *Trachelomonas* were determined, 27 of which were examined for their lorica ultrastructure and photographed in detail by scanning electron microscopy (SEM). *T. volvocinopsis*, meanwhile, was only observed under an optical microscope. Before our study, only three taxa belonging to *Trachelomonas* had been described, *T. abrupta* var. *abrupta*, *T. intermedia*, and *T. pulcherrima* var. *ovalis* (Margalef, 1952). This study has significantly increased the knowledge of this genus not only on this island but also in Spain, including extensive and detailed SEM documentations. Further, our results demonstrate the ultrastructural variability between the taxa examined.

METHODS

Phytoplankton samples were collected by a 20 µm pore diameter net during the spring of 2006, 2007, 2008, and 2014 in 13 temporary ponds around Menorca. The samples were kept cool during transport to the laboratory and taxonomic studies were carried out. The first sample analysis was made using an inverted microscope and then an optical microscope. As a result of these studies, specimens of *Trachelomonas* were only found in the samples from the "*bassa*" Verda B, so only these materials were analyzed for this study. Taxonomic identifications and measurements were made using living material.

For SEM observations, materials were fixed with 4 % formaldehyde, washed with bi-distilled water, filtered through Millipore filters (0.20 µm pore), and air-dried. Filter pieces were attached on stubs to be subsequently coated with gold/palladium. Specimens were examined and photographed by means of a HITACHI S-3400N SEM in the Electron Microscopy Service of the University of the Balearic Islands, Mallorca, Spain.

Specialized literature was consulted for taxa identification: Deflandre, 1926; Balech, 1944; Conrad & Van Meel, 1952, Couté & Iltis, 1981, Couté & Thérézien, 1985, Huber¬-Pestalozzi, 1955, Popova, 1966, Starmach, 1983, Tell & Conforti, 1986, Rino & Pereira, 1989-1990, Conforti, 1993, 1999, 2010, Conforti & Tell, 1986, Conforti & Joo, 1994, Conforti & Nudelman,1994, Conforti & Pérez, 2000, Conforti & Ruiz, 2001, Wolowski & Hindák, 2004, 2005, Wolowski & Grabowska, 2007, Wolowski & Walne, 2007, Da et al., 2009, Ciugulea & Triemer, 2010, and Garduño Solorzano et al., 2011.

The characteristics used to delimit the different taxa were size and shape of the lorica, pore with or without a collar, and type of ornamentation. Additionally, comments on relevant morphological features, distribution characteristics, and the list of papers where their ultrastructure

was previously studied were added for each taxon.

STUDY AREA

Bassa Verda B is one of a group of small temporary freshwater ecosystems on Menorca, the Balearic Islands, Western Mediterranean, whose location is 40°3'9"N 3°56'6"E, see map (Fig. 1).

The water comes directly from rain, and its permanence in the bassa is variable, but in general does not exceed three months between October and April. Its maximum depth is 52 cm and the maximum surface area when full is 307 m^2. Within the limnological classification made by Pretus et al. (2010), Bassa Verda B is characterized by the presence of organic and inorganic particles, clay, and silt in suspension. In the different analyses carried out, the water has always had a low mineral content, with concentrations of Ca, Mg, and SO_4 less than 100 mg/L, and conductivity not exceeding 1000 µS/cm, while the pH is slightly basic with little buffering capacity, see Table 1.

This body presents a high diversity of eukaryotic phytoplankton, with representatives of different groups of algae in addition to *Trachelomonas*. There are many belonging to the Zygnematophyceae *Cosmarium* and *Staurastrum*, diatoms of the genera *Fragilaria*, *Pinnularia*, *Rhopalodia*, and *Stauroneis*, and the Dinophyceae *Peridinium* and *Peridiniopsis*. It is interesting to note the presence of Chrysophyceae cysts formed by silica, an important difference with the loricas of *Trachelomonas* which are rich in iron, an element present in water associated with organic matter (Moyà 2010).

Table 1. Average values of different chemical parameters analyzed in the basses studied. Ordered from highest to lowest iron concentration from Petrus et al. (2010). *Valores medios de diferentes parámetros químicos analizados en los estanques temporales estudiados (basses). Ordenados de mayor a menor concentración de hierro a partir de Petrus et al. (2010).*

Bassa	Fe µm	pH	Ca^{++} mg/l	Conductividad µS/cm
Es Mal Lloc	71.3	6.94	26.65	660
Algaiarens	29.8	6.68	16.03	1.210
Ses Pallisses	27.0	7.16	28.46	1,290
Penyes Egipte	12.5	6.22	7.61	350
Verda B	5.2	7.37	66.93	720
Curniola	5.1	7.35	11.62	330
Sa Mesquida	3.0	7.78	139.87	4.300
Ets Armaris	1.4	7.48	87.77	5.640
Verda A	0.8	7.70	73.34	1.200
Es Molinet	0.0	7.45	-	1.530
Torre Llafuda	0.0	8.39	25.65	440
Clot des Guix	0.0	7.60	435.20	2.660
Cos des Síndic	0.0	7.84	1118.00	47.000

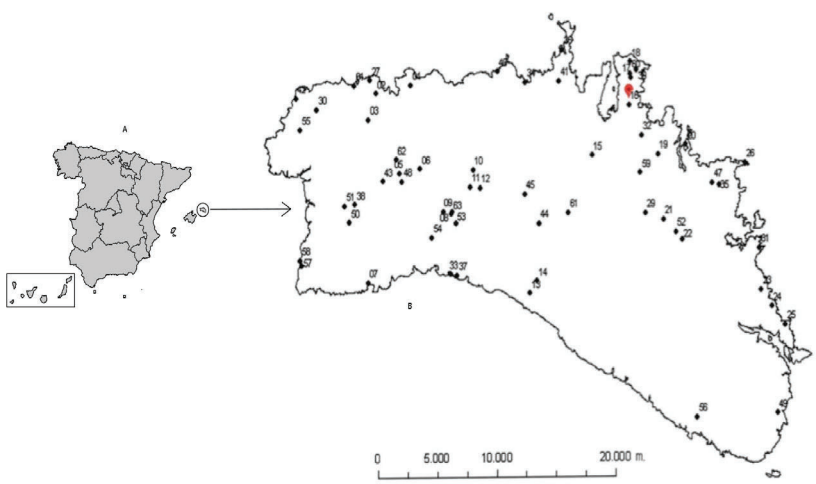

Figure 1. A) Map of Spain; B) Menorca Island, Spain, locating with the number 16 the Bassa Verda. A) Mapa de España B) Isla de Menorca, España, localizando con el número 16 la Bassa Verda.

RESULTS

As a result of this study, 28 taxa of *Trachelomonas* were described in alphabetical order, 18 of which were new recordings for Spain, indicated by an asterisk (*) before their denomination. Figures 2 to 42 show the characteristics of each species grouped according to Deflandre (1926).

Genus *Trachelomonas* Ehrenberg 1833 emend. Deflandre 1926

T. abrupta var. *arcuata* (Playf.) Defl. Figure 33

Loricas 18-19 µm long, 18-19 µm wide; smaller than those cited in the bibliography, broadly ovoid, curved sides, posterior end rounded. Surface densely and finely punctuate (300-340/100 µm^2). Pore surrounded by an annular thickening 2-3 µm diam. Cosmopolitan. In Spain: Sierra Nevada, Granada (De La Rosa & Sánchez Castillo, 1991).

T. abrupta var. *minor* Defl. Figures 34-35

Lorica 20-22 µm long, 11-13 µm wide; cylindrical with the sides slightly curved to parallel, the posterior end somewhat acuminate. Surface with irregularly distributed punctuations, rather separate from each other (110-160/100 µm^2), ornamented with closely distributed warts. Pore with an annular thickening surrounded by a crown of conical spines. Cosmopolitan. Cited in Spain sec. Cobelas,1984; Sierra Nevada, Granada (De La Rosa & Sánchez Castillo, 1991).

* *T. bacillifera* var. *minima* Playf. Figures 29-30

Lorica 21-23 µm long, 18-20 µm wide; ovoid to broadly ellipsoid. Surface punctuate (120-150/100 µm^2), ornate with rod-like obtuse spines (0.5-1.5 µm long), irregularly distributed (90-110/100 µm^2). Pore surrounded by an annular thickening 3-3.5 µm diam. Cosmopolitan.

* *T. curta* Da Cunha em. Defl. Figure 4

Lorica 15-21 µm long, 17-23 µm diam.; subspherical. Surface smooth. Pore 2.5-3 µm diam., surrounded by a fine annular thickening. Cosmopolitan.

* *T. duquei* Conf. & Nud. var. *minor* nov. var. Figure 25

A varietate minoribus dimensionibus differt. Lorica 21-22 µm long., 18-19 µm lat. In Verda *lacu*, Menorca, Hispania.
Lorica 21-22 µm long, 18-19 µm wide; broadly ovate with regularly arched sides. Pore surrounded by an annular thickening 3-3.5 µm diam. Surface strongly punctuate (300-350/100 µm²), ornamented with short conical spines (0.5-1 µm long), sparsely located (50-65/100 µm²). *T. duquei* was described by SEM in materials from the USA (Wolowski & Walne, 2007) but their specimens showed dimensions more like this new variety than those of that type.

* *T. globularis* (Aw.) Lemm. Figures 7-8

Lorica 17-19 µm diam.; spherical. Surface punctuate (200-220/100 µm^2), totally covered in short, thick, regularly distributed spines (12-20/100 µm²). Cosmopolitan.

We considered that the specimen shown by Da et al. (2009) also corresponded to this taxon; they described it as *T. globularis* var. *punctata*, but that variety has the lorica covered by few dispersed spines.

* *T. granulosa* var. *crenulatocollis* (Szab.) Hub.-Pest. Figures 36-37

Lorica 25-29 µm long, 17-19 µm wide; ellipsoid. Surface strongly punctuate (90-110/100 µm²), very irregular, ornamented with granules (40-50/100 µm²) and small, irregularly distributed papillae. Pore surrounded by a cylindrical collar (2-3 x 4-4.5 µm) with conical spines at the tip, which diverge distally. Cosmopolitan. This taxon is studied by SEM for the first time.

* *T. hirta* Da Cunha. Figures 18-19

Lorica 19-21 µm long, 16-18 µm wide; broadly ellipsoid with round to subspherical ends. Surface reddish to deep brown, strongly punctuate (60-

90/100 µm²). They showed long spines (1.5-2.5 µm long) which were irregularly located (6-10/100 µm²) and presented a particular shape: conical and sharply tapered towards the tip. Pore surrounded by an annular thickening (2-2.5 µm diam.). Only recorded in America: Argentina, Brazil, Colombia, and the USA.

T. hispida f. *hispida* (Perty) Stein *em*. Defl. Figures 14-15

Lorica 23-26 µm long, 19-21 µm wide; broad ellipsoid. Surface deep to reddish-brown, finely punctuate (180-200/100 µm²), ornamented with short conical spines (0.8-1 µm long), scattered distribution (36-50/100 µm²). Pore surrounded by an annular thickening. Cosmopolitan. In Spain: Barcelona (Margalef, 1948 sec. Cobelas 1984), Granada (Sánchez Castillo, 1988, De La Rosa & Sánchez Castillo, 1991).

* *T. hispida* f. *minor* Bourr.

Lorica 17-18 µm long, 13.5-14 µm wide; the dimensions of our specimens were smaller than those described with SEM by Conforti & Nudelman (1994), 21-23 x 16-18 µm, broad ellipsoid. Surface yellowish- to reddish-brown, finely punctuate (100-110/100 µm²), ornamented with short pointed conical spines (0.5-1 µm), regularly distributed (70-85/100 µm²). Widespread.

This species was studied by SEM, but we consider that their photos do not have a good enough quality to be published.

T. intermedia Dang. Figure 9

Lorica 18-21 µm long, 15-17 µm wide; broadly ellipsoid. Surface densely punctuate (250-300/100 µm²). Pore (2-2.5 µm diam.) surrounded by a fine annular thickening. Cosmopolitan. In Spain: Barcelona (Margalef, 1948), Mallorca (Margalef, 1952).

T. nigra Swir. Figure 13

Lorica 21-22 µm long, 17-18 µm wide; broadly ellipsoid. Surface irregularly punctuate (65-70 /100 µm²) and slightly papillate, especially at the posterior end. Pore (2.5-3 µm diam.) surrounded by an annular thickening and small papillae. Thailand, Europe. In Spain: Granada (De La Rosa & Sánchez Castillo, 1991).

* *Trachelomonas planctonica* var. *oblonga* Drez. Figures 38-39

Lorica 25-27 µm long, 17-18 µm wide, oblong. Surface punctuate (100-150/100 µm²). Pore surrounded by a short conical truncate collar 2-3 x 4-5 µm, irregularly denticulate at the tip (1-1.5 µm long). Cosmopolitan.

* *Trachelomonas planctonica* var. *planctonica* f. *ornata* (Skv.) Popova. Figures 40-41

Lorica 23-25 µm long, 16-18 µm wide; surface punctuate (95-120/100 µm²). Pore surrounded by a short conical truncate collar (1-3 x 4-5 µm), with conical spines around the distal end (1-3 µm long). This variety differs from the type by their surface ornamented with very short conical to rod-like obtuse, very sparsely distributed, spines (210-310 nµm). Widespread.

* *T. pseudofelix* Defl. Figure 12

Lorica 15-17 µm diam.; spherical to subspherical. Surface coarse, punctuate, ornamented with small papillae and different types of irregularly located granulations. Pore (2-3 µm diam.) surrounded by flattened particles. Argentina, China, France, Holland.

This is the first SEM study of the species. Wolowski & Grabowska (2007) showed a SEM photo but did not analyze the ultrastructure.

* *T. raciborskii* var. *nova* f. *minor* Hort. Figure 20

Lorica 20-21 µm long, 17-18 µm wide; broadly ellipsoid. Wall strong, densely scrobiculate, with one punctuation at the bottom (120-160/100 µm²), ornate with robust, short, conical spines (0.5-1.5 µm long) arranged only around the posterior end. Besides this, very small, irregularly distributed papillae could be observed on the rest of the surface. Apical pore without a collar. Hungary, Argentina.

This is the first SEM study of this forma.

* *T. robusta* Swir. *em*. Defl. Figures 16-17

Lorica 18-22 µm long, 13-20 µm wide; ellipsoid to broad ellipsoid. Surface strongly and irregulary punctuate (190-210/100 µm²), ornamented with short (1-1.5 µm long), thick, regularly distributed conical spines (40-55/100 µm²). Pore surrounded by an annular thickening (3-4 µm diam.). Some of the studied specimens showed a smaller diameter than those reported by other authors. Besides this, some of these presented more densely spiny ornamentation than those observed in Da et al., 2009. Cosmopolitan.

* *T. rugulosa* var. steinii Defl. Figures 5-6

Lorica 14.5-15-5 µm diam.; spherical. Surface ornamented densely with slightly anastomosing ribs which radiate from the pore. Pore 1.5-2 µm diam. Colombia, Mexico, Europe, Korea.

Trachelomonas scabra Playf. Figure 42

Lorica 19-21 µm long, 16-18 µm wide; with a very irregular shape. Surface thick, coarse, with numerous adhering exogenous particles on its surface, which increase its diameter and change its external contour. Pore (4.5-5 µm diam) surrounded by agglutinated particles. Widespread. Cited in Spain sec. Cobelas, 1984; Sierra Nevada, Granada (as *Strombomonas scabra* (Playf.) Tell & Conf. by De La Rosa & Sánchez Castillo, 1991).

T. superba var. *superba* Swir. em. Defl. Figures 21-22

Lorica 33-38 µm long, 28-32 µm wide, broadly ellipsoid. Surface densely punctuate (350-400/100 µm²), ornamented by numerous conical spines (1-2.5 µm long), scattered distribution (20-25/100 µm²). Pore (4-5 µm diam.) surrounded by a crown of 8-10 conical spines. Cosmopolitan. Cited in Spain sec. Cobelas, 1984.

* *T. superba* var. *swirenkiana* Defl. Figures 23-24

Lorica 35-40 µm long, 27-31 µm wide; broadly ellipsoid. Surface yellowish to deep brown, finely and densely punctuate (300-350/100 µm²), ornamented by scattered conical spines (30-45/100 µm²) which are longer around the posterior end (2.5-4.5 µm long). Pore (3.5-4 µm diam.) surrounded by a crown of 12-14 conical spines. Argentina, Bolivia, Europe.

* *T. sydneyensis* var. *minima* Playf. Figures 26-27

Lorica 28-30 µm long, 18-20.5 µm wide. Surface strongly punctuate (300-320/100 µm²), ornamented by scattered conical spines (40-45/100 µm²). In addition to this, some specimens showed numerous papillae (not shown). Pore (3.5-4 µm diam.) surrounded by a crown (Fig. 26) of divergent conical spines (1-1.5 µm long). Widespread.

* *T. sydneyensis* var. *obesa* Playf. Figure 28

Lorica 33-37 µm long, 28.5-30.5 µm wide. Surface densely punctuate (250-300/100 µm²), ornamented by conical spines (1-1.5 µm long), scattered distribution (40-50/100 µm²). Pore (6.5-7 µm diam.) surrounded by a short neck (1-1.5 µm long) ornamented by divergent conical spines (0.5-1 µm long). It is the first SEM study of this variety. Australia.

* *T. verrucosa* Stokes f. *irregularis* Defl. Figures 10-11

Lorica 10-11 µm long, 9.5-10 µm diam.; spherical to subspherical. Surface not punctuate, showing a fine network of folds ornamented by very small granules irregularly distributed on their crests. Pore 1.5-2 µm diam., surrounded by an annular thickening. Widespread.

T. volvocina var. *volvocina* Ehr. Figure 2

Lorica 5-17 µm diam.; spherical. Surface smooth, deep reddish brown. Pore 1.5-2 µm diam., surrounded by a fine annular thickening. Cosmopolitan. In Spain: Barcelona, Gerona (Margalef, 1948 sec. Cobelas, 1984).

T. volvocina var. *punctata* Playf. Figure 3

Lorica 13-15 µm diam.; spherical. This variety differs from the type because of the lorica punctuate. Pore 2-2.5 µm diam. Cosmopolitan. In Spain:

Gerona (Margalef, 1948 sec. Cobelas 1984).

T. volvocinopsis Swir.

Lorica 15-20 µm diam.; spherical. This species was only observed by optical microscopy. It is likely that some of those identified by SEM, such as *T. volvocina*, belong to this species, since both showed identical lorica ultrastructure. These taxa are only distinguished by their chloroplasts: *T. volvocinopsis* presents numerous ones without a pyrenoid; while *T. volvocina* shows only two, each one with a diplopyrenoid. Cosmopolitan. Cited in Spain sec. Cobelas, 1984; Tablas de Daimiel (Rojo et al., 1999).

* *T. zorensis* Lef. Figures 31-32

Lorica 15-20 µm long, 14.5-16 µm wide; broadly ellipsoid, posterior end rounded. Surface reddish-brown, strongly, and densely scrobiculate (150-200/100 µm²), with one punctuation at the bottom. Pore surrounded by an annular thickening (2-3 µm diam.). This species was studied previously by SEM by Conforti & Tell (1989) but the lorica of our specimens showed smaller dimensions than those examined by these authors (24-28 µm long, 20.5-22 µm wide), and were coincident with those given by Huber-Pestalozzi (1955). Argentina, Europe.

DISCUSSION

As a result of the analysis of the material from the sampled ponds, a great diversity of different genera of microalgae could be observed, but among these 13 studied bodies only one – called *bassa* Verda B – presented a wide variety of species of Euglenophyta. Their ability to move freely in aquatic environments and their capacity to live heterotrophically confers this group advantages in shallow waters, where they can capture nutrients present in deeper layers and then migrate to the euphotic zone (Giani et al., 1999). These microalgae are commonly found in reservoirs (Wołowski & Grabowska, 2007) and not in large lakes or bodies with running water.

The available information (Pretus et al., 2010) suggests that the chemical heterogeneity of the geological substrate of each one, drainage area, and human influence are the determining factors of their differences. These authors said that *bassa* Verda B belongs to the pond type with a low mineral content, and neutral or slightly basic clear water. They found a spectrum of cellular types, and phyletic and ecological diversities that are unique among ponds and even in overall water ecosystems in the Balearic Islands. According to the physical-chemical parameters of this body, it best fits a dystrophic ecosystem model, as its characteristics are most favorable for euglenoid development. This "*bassa*" is rich in dissolved organic carbon, has low dissolved O_2 content, and high values of total phosphorus and ammonium. The elevated organic matter content could explain the high development of euglenoids, which usually increase their abundance in these kinds of environments (Lackey, 1968; Munawar, 1972; Conforti, 1991; Reynolds et al., 2002; Rosowski, 2003; Wolowski & Hindák, 2004). The particular characteristics of this body would be decisive for the presence of a large number of species of *Trachelomonas*, a genus considered a typical indicator of a high content of organic matter (Sládecek, 1973), especially associated with elevated levels of ammonium (Alves-da-Silva et al., 2008). Additionally, "*bassa*" Verda B presented a high concentration of iron, which is fundamental for *Trachelomonas* lorica development.

CONCLUSIONS

From this study, we can clearly conclude that *bassa* Verda B presents very appropriate conditions for the development of the *Trachelomonas* species. Among the 28 taxa described in this paper, 27 were photographed by SEM, while *T. volvocinopsis* was only observed under optical microscope. *T. duquei* var. *minor* was proposed as a new variety. Four taxa were examined and photographed by SEM for the first time ever: *T. granulosa* var. *crenulatocollis*, *T. pseudofelix*, *T. raciborskii* var. *nova* f. *minor*, and *T. sydneyensis* var. *obesa*. Most of the described taxa have a widespread or cosmopolitan distribution and are commonly cited, such as *T. hispida* var. *hispida*, *T. robusta*, *T. volvocina* var. *volvocina*, and *T. volvocinopsis*. Others are more rarely recorded, *T. nigra* (Thailand, Europe), *T pseudofelix* (Argentina, China, France, Holland), *T. raciborskii* var. *nova* f. *minor* (Argentina, Hungary). Finally, *Trachelomonas hirta* had only been recorded in America (Argentina, Brazil, Colombia, and the USA) and *T. sydneyensis*

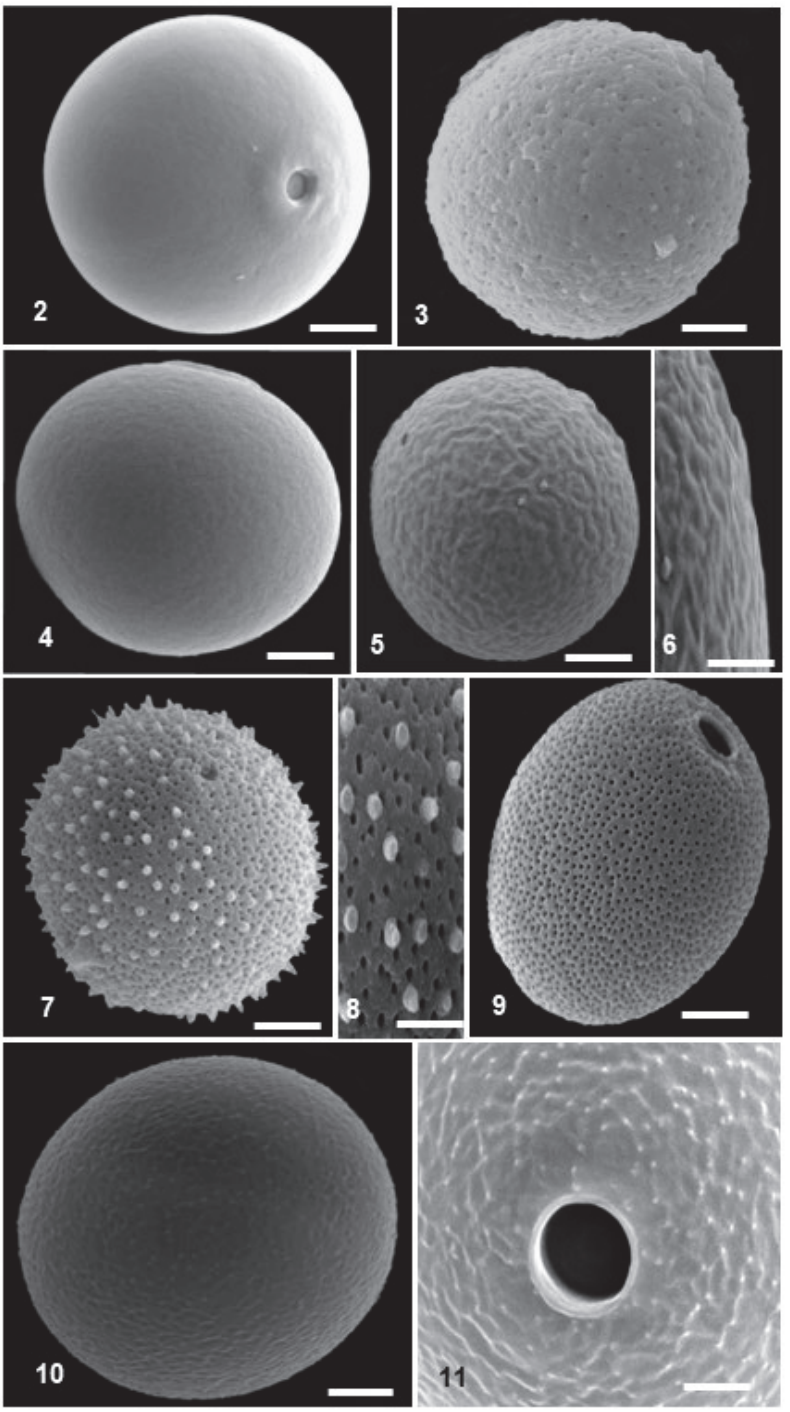

Figure 2-11. (2) *T. volvocina* var. *volvocina*; (3) *T. volvocina* var. *punctata*; (4) *T. curta*; (5-6) *T. rugulosa* var. *steinii*, (5) general view, (6) detail of the lorica surface; (7-8) *T. globularis*, (7) general view, (8) detail of the lorica surface; (9) *T. intermedia*; (10-11) *T. verrucosa* f. *irregularis*, (10) general view, (11) detail of the lorica surface. *(2)* T. volvocina var. volvocina*. (3)* T. volvocina *var.* punctata*. (4)* T. curta*. (5-6)* T. rugulosa var. steinii*; (5) vista general, (6) detalle de la superficie de la lórica. (7-8)* T. globularis*; (7) vista general, (8) detalle de la superficie de la lórica. (9)* T. intermedia*. (10-11)* T. verrucosa *f.* irregularis*; (10) vista general, (11) detalle de la superficie de la lórica.* Scale bars: 2-5, 7, 9-10 = 10 µm; 6, 8 = 5 µm, 11 = 4 µm.

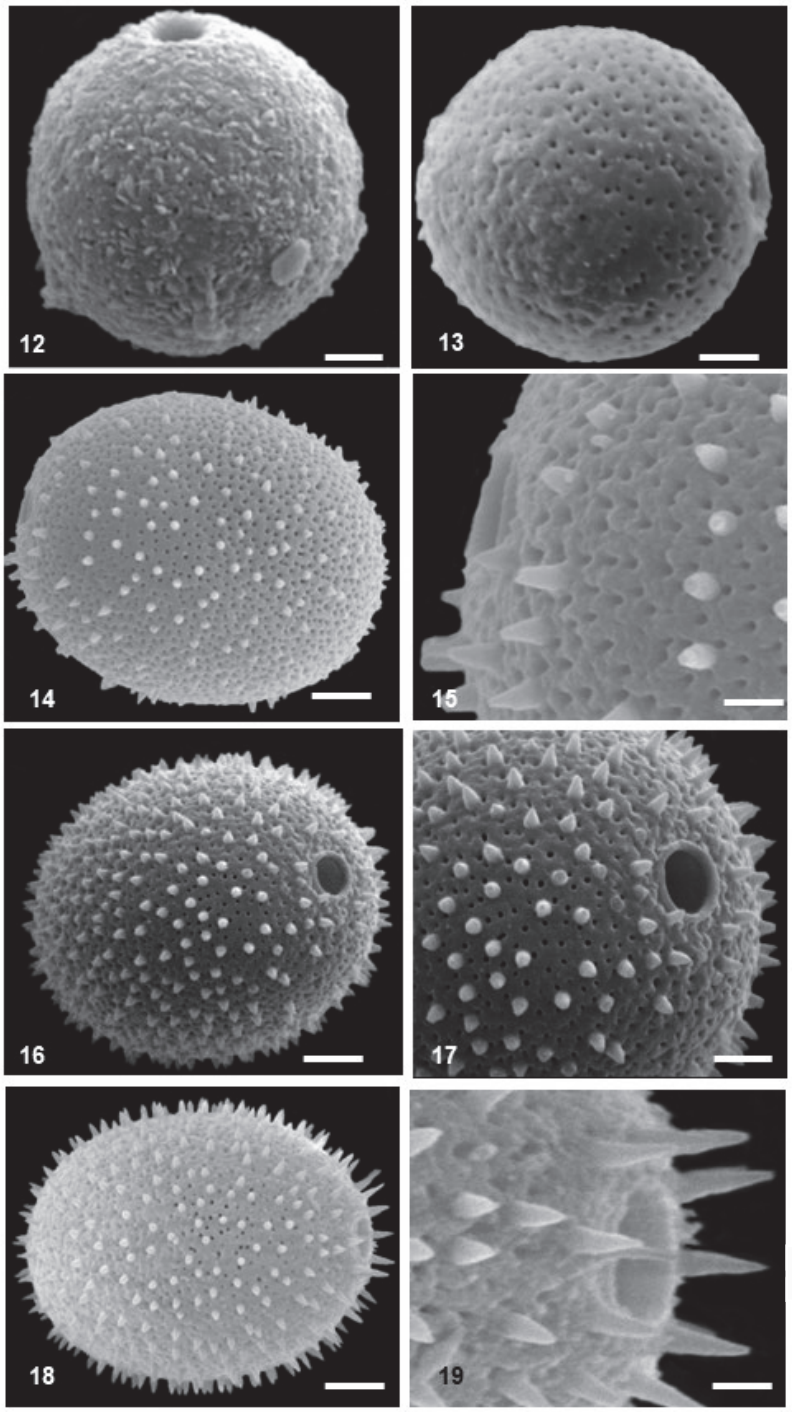

Figure 12-19. (12) *T. pseudofelix,* (13) *T. nigra,* (14-15) *T. hispida* f. *hispida,* (14) general view, (15) detail of the lorica surface, (16-17) *T. robusta*, (16) general view, (17) detail of the lorica surface, (18-19) *T. hirta*, (18) general view, (19) detail of the pore and lorica surface. *(12)* T. pseudofelix, *(13)* T. nigra*, (14-15)* T. hispida *f*. hispida, *(14) vista general, (15) detalle de la superficie de la lórica, (16-17)* T. robusta*, (16) vista general, (17) detalle de la superficie de la lórica, (18-19)* T. hirta*, (18) vista general, (19) Detalle del poro y de la superficie de la lórica.* Scale bars: 12-14, 16, 17 = 10 µm; 15, 17, 19 = 5 µm.

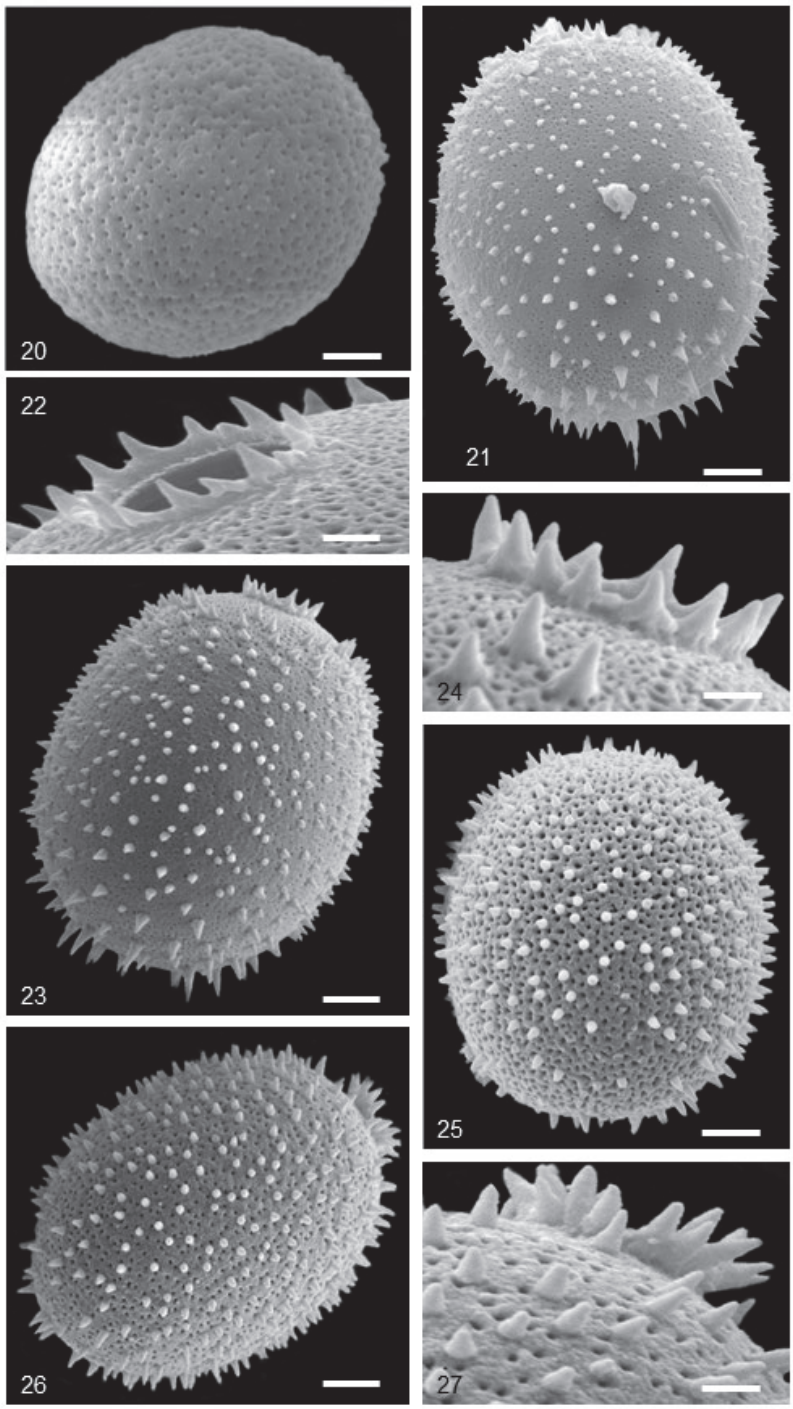

Figure 20-27. (20) *T. raciborskii* var. *nova* f. *minor*; (21-22) *T. superba* var. *superba*, (21) general view, (22) detail of pore ornamentation; (23-24) *T. superba* var. *swirenkiana*, (23) general view, (24) detail of pore ornamentation; (25) *T. duquei* var. *minor*; (26-27) *T. sydneyensis* var. *minima*, (26) general view, (27) detail of pore ornamentation. *(20)* T. raciborskii *var.* nova *f.* minor. *(21-22)* T. superba *var.* superba*; (21) vista general, 22. detalle de la ornamentación del poro. (23-24)* T. superba *var.* swirenkiana*; 23. vista general, 24. detalle de la ornamentación del poro. 25.* T. duquei *var.* minor. *(26-27)* T. sydneyensis *var.* minima*; 26. vista general, 27. detalle de la ornamentación del poro.* Scale bars: 23, 25 = 20 µm; 20, 22, 26 = 10 µm; 21. 24, 27 = 5 µm.

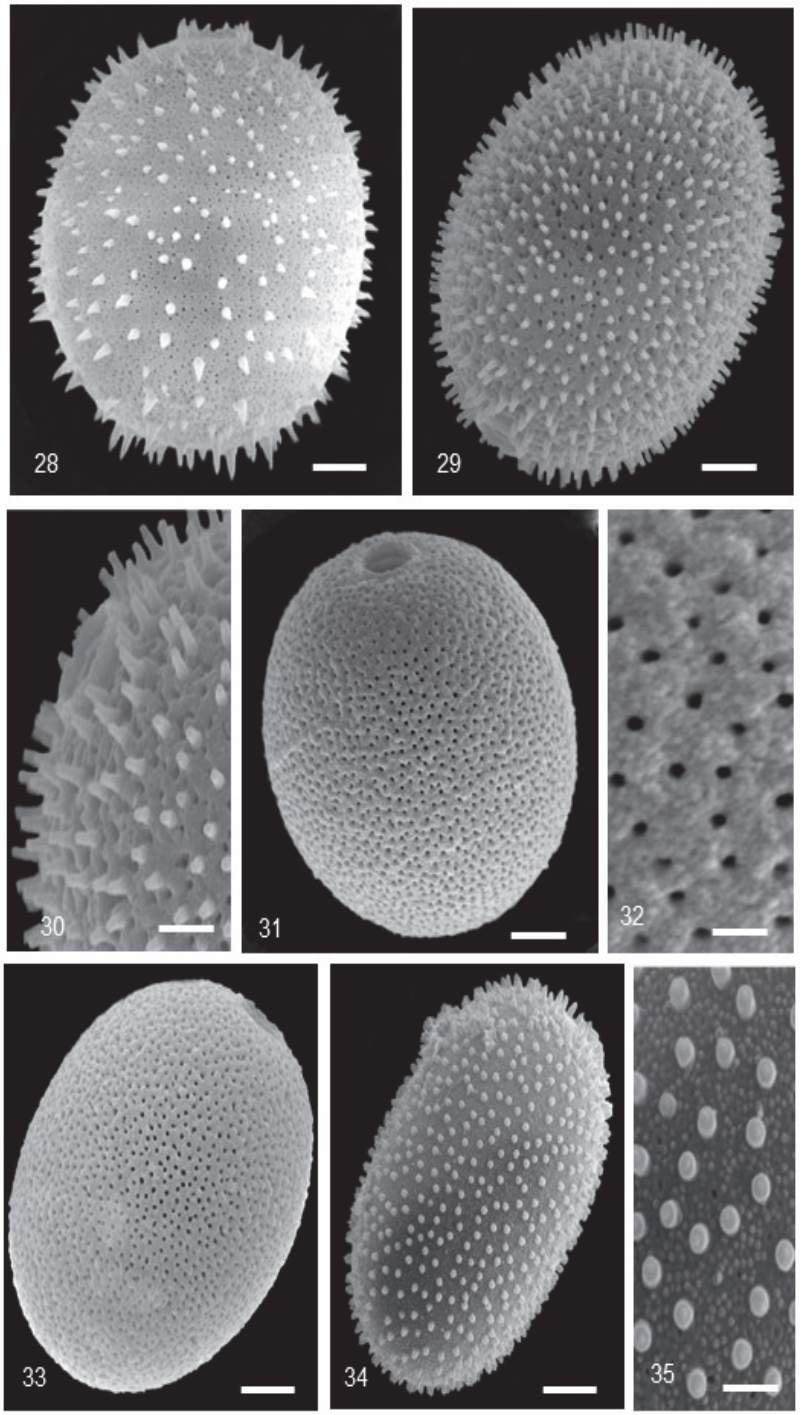

Figure 28-35. (28) *T. sydneyensis* var. *obesa*; (29-30) *T. bacillifera* var. *minor*, (29) general view, (30) detail of pore ornamentation; (31-32) *T. zorensis*, (31) general view, (32) detail of the lorica surface; (33) *T. abrupta* var. *arcuata*; (34-35) *T. abrupta* var. *minor*, (34) general view, (35) detail of the lorica surface. *(28)* T. sydneyensis *var.* obesa*. (29-30)* T. bacillifera *var.* minor*; (29) vista general, (30) detalle de la ornamentación del poro. (31-32)* T. zorensis*; (31) vista general, (32) detalle de la superficie de la lórica. (33)* T. abrupta *var.* arcuata*. (34-35)* T. abrupta *var.* minor*; (34) vista general, (35) detalle de la superficie de la lórica.* Scale bars: 28 = 20 µm; 29, 31, 33, 34 = 10 µm; 30, 32 = 5 µm; 35 = 3 µm.

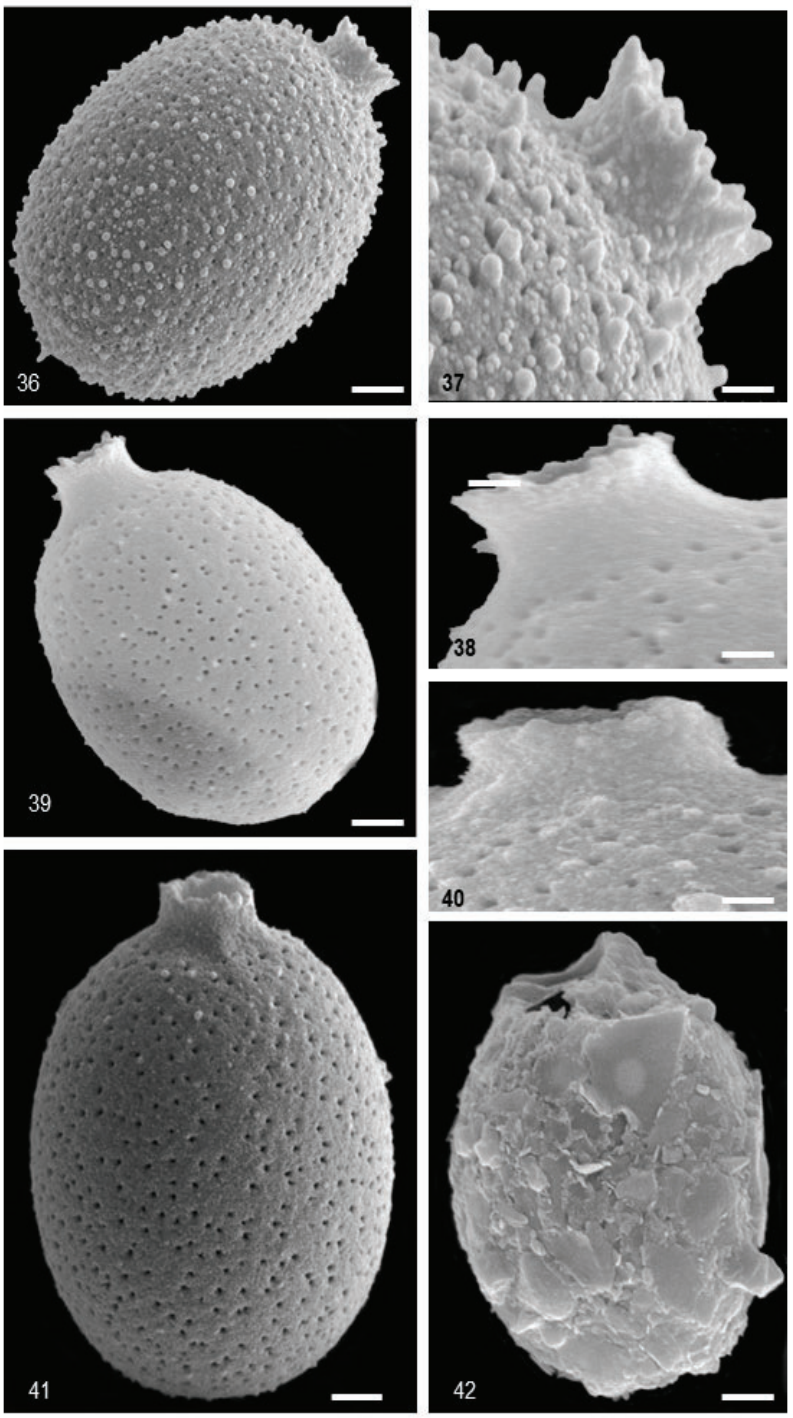

Figure 36-42. (36-37) *T. granulosa* var. *crenulatocollis*, (36) general view, (37) detail of the collar and lorica surface; (38-39) *T. planctonica* var. *oblonga*, (38) detail of the collar, (39) general view; (40-41) *T. planctonica* var. *planctonica* f. *ornata*, (40) detail of the collar, (41) general view; (42). *T. scabra*. *(36-37)* T. granulosa *var.* crenulatocollis*; (36) vista general, (37) detalle del collar y de la superficie de la lorica. (38-39)* T. planctonica *var.* oblonga*; (38) detalle del collar, (39) vista general. (40-41)* T. planctonica *var.* planctonica *f.* ornata*; (40) detalle del collar, (41) vista general. (42)* T. scabra. Scale bars: 36, 37, 41, 42 = 10 µm; 37 = 4 µm; 38 = 3 µm; 40 = 5 µm.

var. *obesa* only in Australia, so this is the first time that these species have been cited for Europe. All the taxa described in this work are cited in Menorca for the first time, while 18 of them are new recordings for Spain.

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