



## ***Cyclostephanos salsae* and *Placoneis patagonica*, two new diatoms (Bacillariophyta) from Laguna Cháltel in southern Patagonia, Argentina**

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With 42 figures and 1 table

**Abstract:** Two new diatom species, *Cyclostephanos salsae* and *Placoneis patagonica* found in plankton, superficial sediments and quaternary lacustrine sediment samples from Laguna Cháltel, Southern Argentina, are described. The detailed morphology of these two taxa is examined using both light (LM) and scanning electron microscopy (SEM). The main morphological features that distinguish *C. salsae* sp. nov. are the raised central area without fultoportulae and with scattered areolae, and the presence of some interstriae bifurcated on the valve mantle, bearing one fultoportulae in one of the forks. The variable, sometimes asymmetrical outline and the irregularly spaced central striae are the main morphological features of *P. patagonica* sp. nov. The relationships of these two new taxa with morphologically similar species and their ecological affinities are discussed.

**Key words:** Bacillariophyta, centric diatoms, crater lake, Holocene, Paleolimnology, sediments, pennate diatoms, Santa Cruz Province, South America

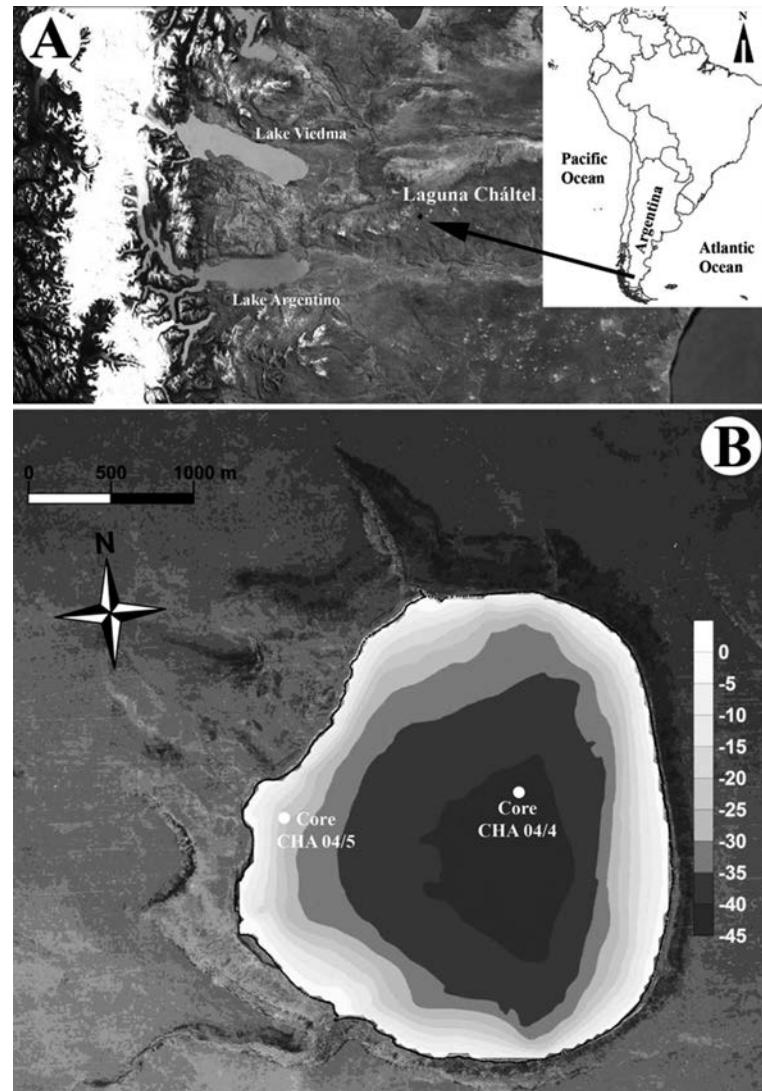
Received on 20th September 2016, revised manuscript accepted on 29th March 2017

### **Introduction**

Patagonia is a wide geographical region (1,060,631 km<sup>2</sup>) located at the southernmost end of South America, covering territories of Chile, at the west, and Argentina, at the east of the Andes (Fig. 1). In Argentina, it extends over the provinces of La Pampa, Neuquén, Río Negro, Chubut, Santa Cruz, and Tierra del Fuego.

As a result of a rain shadow effect of the Andes, Argentinian Patagonia exhibits a strong rainfall gradient in the W-E direction with rainfall exceeding 2000 mm/year in the Andean region to less than 200 mm/year in the area close to the Atlantic Ocean (Paruelo et al. 1998).

Argentinian Patagonia is promising for biodiversity studies since it has been relatively unaffected by human activities. The region has numerous waterbodies that have been little explored, especially because many areas are remote and inaccessible, which implied serious logistic problems for sample collection. The first algal studies for the region were published in the mid 1900s (Frenguelli 1923–1924, 1933, 1936, 1939, 1941, 1942, 1951, 1953, Guarrera & Kühnemann 1949, Seckt 1950–1956) mainly as floristic works. Studies of the last two decades



**Fig. 1.** Study area. In grey, the Argentinian Patagonia.

also include ecological aspects showing structure and dynamics of algal communities, as well as the neo- and paleolimnology of lakes and ponds (e.g., Izaguirre 1993, Izaguirre et al. 1990, Izaguirre & Saad 2014, Guarrera & Echenique 1992, Díaz et al. 2000, Tell et al. 2011, Mayr et al. 2005 and Zolitschka et al. 2006). The coverage of the different regions of Patagonia is not uniform. For the Santa Cruz Province, there are a few publications; some of them do not mention the diatoms (e.g., Seckt 1931, 1950–1956, Guarrera & Kuhnemann 1949 and Tell et al. 2011), whereas others only concentrate on this group of algae (e.g., Krasske 1949, Luchini 1975, 1976, Maidana 1999, Maidana & Round 1999, Maidana et al. 2005, Messyasz et al. 2007 and Echazú & Maidana 2010).

The austral extreme of Argentinian Patagonia has been little explored regarding algal diversity in the numerous aquatic habitats available, but in the last 20 years, several neo- and paleolimnological projects were conducted in the region, which allowed the study of modern and fossil diatoms in shallow and deep lakes, wetlands and bogs previously unexplored (Echazú 2012, Izaguirre et al. 2015). In particular, the international and interdisciplinary projects South Argentinian Lakes Archives and Modeling (SALSA, Zolitschka et al. 2004) and Potrok Aike maar Lake Sediment Archive Drilling Project (PASADO, Zolitschka et al. 2006a) yielded new genera, such as *Corbellia* Maidana & Round (1999), and species such as *Thalassiosira patagonica* Maidana (1999), and *Cymbella gravida* Recasens & Maidana (2013).

One of the goals of the SALSA project was to perform high-resolution analysis and interpret the information provided by various proxies, including diatoms. As part of this project we have undertaken studies of various lakes in the Province of Santa Cruz, Argentina. When analyzing samples from Laguna Cháltel collected from water, surface sediments, and sediments recovered from two cores drilled at the center of the lake, unknown representatives of the genera *Cyclostephanos* Round and *Placoneis* Mereschk. were observed. The morphology of both taxa was analyzed using LM and SEM. The peculiar characteristics of the two taxa allow us to consider them as new to science.

## Material and methods

### Study area

Laguna Cháltel is a crater lake located at 49°58' S, 71°07' W in the Pampa Alta volcanic plateau, Santa Cruz Province, southern Patagonia, Argentina (Fig. 1). In some maps this lake is called 'Laguna Azul', however, local people do not use that name. To avoid any confusion with the multitude of other lakes bearing that name we prefer to use the name Laguna Cháltel as 'cháltel' is an expression of the native Tehuelche group, meaning 'bluish' (Baleta 1999). The lake is located in the semiarid steppe environment of southern Patagonia at about 800 m a.s.l. on the Cenozoic Pampa Alta volcanic plateau. Laguna Cháltel is a typical maar lake with steep ca. 40 m high subaerial crater walls, a mean diameter of 2.6 km and a maximum depth of 41 m. There are several apparently permanent inflows fed by springs at the crater walls entering the lake via three deeply incised canyons. The environmental history of this lake has been studied by Ohlendorf et al. (2014) through a multiproxy approach using diatoms along with other biological, sedimentological and geochemical data to investigate the hydrological development of Laguna Cháltel and the paleoenvironmental history of southeastern Patagonia during the last ca. 4700 cal BP.

### Methods

Studied samples came from (a) two sediment cores, one obtained from the center of the lake (CHA 04/4; 58 cm in length) and the other located 20 m from the shore (CHA 04/5; 98 cm in length), (b) surface sediments of the littoral zone of the lake and (c) a 1L water sample, obtained in the middle of the lake with a PVC bottle. All samples were collected in February 2004 and preserved *in situ* with 4% formalin.

For diatom analyses, core CHA 04/4 was subsampled every 4 cm while core CHA 04/5 every 16 cm. Part of each subsample was processed following standard methods for diatom analysis (Battarbee 1986). An aliquot of each sample was dried at 80 °C. The sample was oxidized with H<sub>2</sub>O<sub>2</sub> (30%, 100 Vol.) and heated in a microwave oven for 2 min at maximum power in order to eliminate organic material. The samples were rinsed repeatedly until neutrality with distilled water. Permanent slides were mounted using Naphrax®.

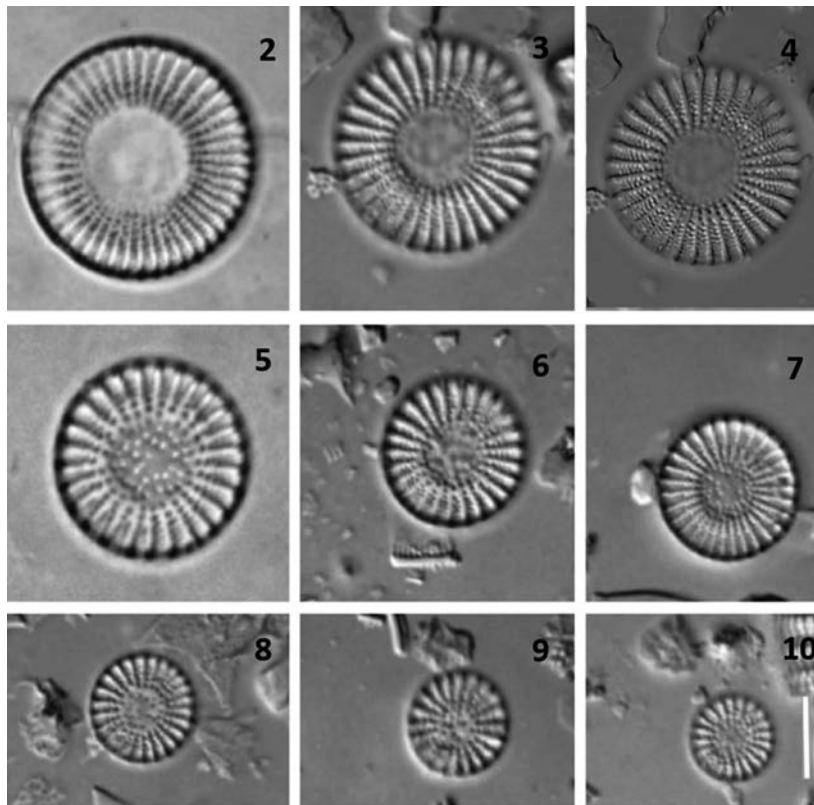
Light micrographs were captured using a Reichert-Jung Polivar binocular optical microscope equipped with a PlanApo 100X, NA 1.32, immersion objective and DIC optics and a Sony Cyber-Shot, digital camera.

For SEM observations, aliquots of the cleaned material were dried on aluminum stubs at room temperature before being coated with gold and examined using a Philips XL series 30 electron microscope from the Electron Microscopy Service, Museo Argentino de Ciencias Naturales (Buenos Aires, Argentina).

Absolute abundances were calculated using the microspheres method (Battarbee & Kneen 1982), and to calculate the relative abundances, at least 400 valves per slide were counted. The relative abundances of the most abundant diatom species (> 3% in at least one sample) were plotted using the computer program TILIA and TGView 2.0.2; (Grimm 1993, 2004).

Terminology used in the descriptions was based on Round et al. (1990), Håkansson (2002), Cox (1987, 2003) and Houk et al. (2014).

Water temperature (12.3 °C), pH (8.92), dissolved oxygen (8.9 mg L<sup>-1</sup>), and electric conductivity (786 µS cm<sup>-1</sup>) were measured in the field with a Universal Pocket Meter (Multi 340i, WTW).

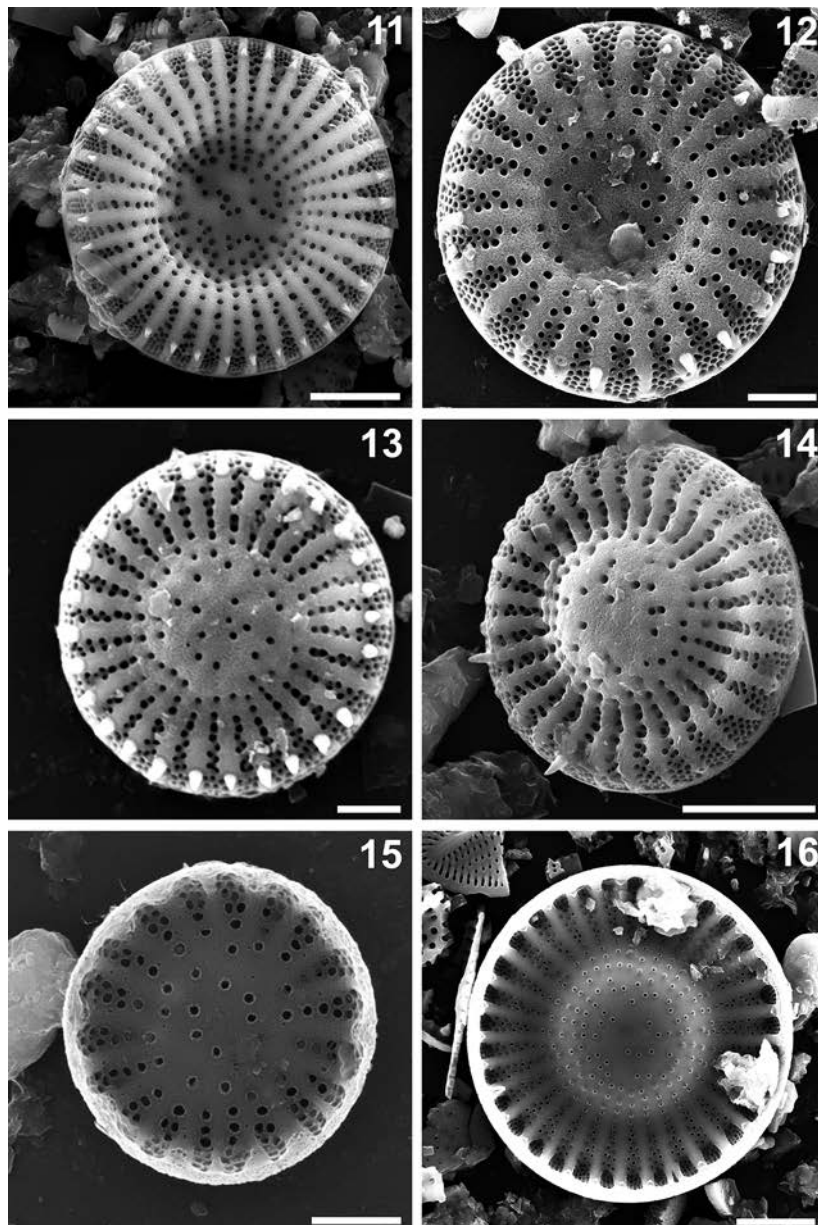


**Figs 2–10.** *Cyclostephanos salsae* sp. nov. LM. (slide CHA 04/4 6–7 cm). Variability in valve diameter; stria density, and arrangement of the central areolae. Scale: 10 µm.

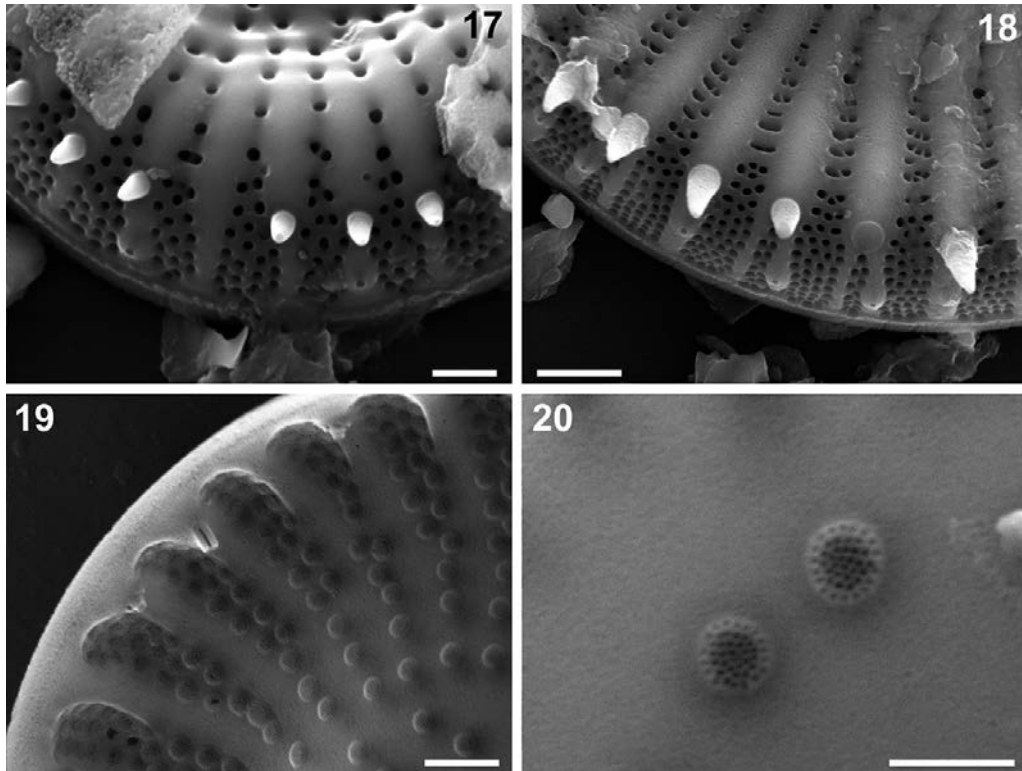
**Observations**

*Cyclostephanos salsae* Maidana & Aponte sp. nov. Figs 2–10 (LM); 11–20 (SEM)

*Description:* Cells solitary, shortly cylindrical, perivalvar axis shorter than diameter. Valves circular, central portion concave or convex. Striae radial, fasciculated at the periphery, unordered and scarce in central region; 2–3 series of areolae at valve face-mantle junction and in sets of



**Figs 11–16.** *Cyclostephanos salsae* sp. nov. SEM. (slide CHA 04/4 6–7 cm). Figs 11–14. External views; Figs 15–16. Internal views. Scale bars represent 10  $\mu$ m.



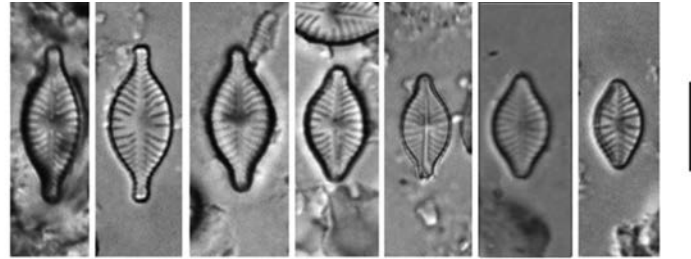
**Figs 17–20.** *Cyclostephanos salsae* sp. nov. SEM. (slide CHA 04/4 6–7 cm). Figs 17–18. Valve mantle in external view. Fig. 17. Valve with no forked interstriae. Fig. 18. Valve with forked interstriae; note the external opening of the fuloportulae in one of the branches (arrows). Figs 19–20. Internal views. Fig. 19. Detail of the valve margin. Note the openings of the fuloportulae (arrows) and the rimoportula (arrowhead). Fig. 20. Detail of the cribra. Scale bar represents 1  $\mu$ m.

4–5 areolae on valve mantle, with parallel or quincunx arrangement (Figs 2–10; 11–14). Areolae internally occluded by domed cribra (Fig. 20).

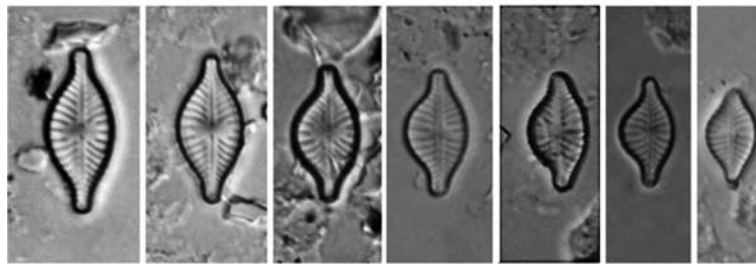
Prominent interstriae almost always with a spine at valve-mantle junction. Interstriae continue to valve mantle where they sometimes bifurcate (Figs 17–18).

Central area lacking fuloportulae; marginal fuloportulae located in mantle, every 1–2, exceptionally 3, interstriae. In case of bifurcate interstriae, marginal fuloportulae located in one branch (Fig. 18). External fuloportula opening located in small conical protrusion (Figs 17–18), 2 satellite pores seen in inner view (Fig. 19). One sessile rimoportula located in one non-bifurcated interstria, frequently lacking a spine (Figs 12). External rimoportula opening not distinguished from fuloportulae openings. Degree of disorganization and number of areolae in central region not related to valve size (Figs 5, 10). Dimensions: diameter 7.3–36.3 ( $21.4 \pm 4.4$ ;  $n = 56$ )  $\mu$ m; interfascicles 3.0–9.0 ( $4.4 \pm 0.2$ ;  $n = 56$ ) in 10  $\mu$ m; marginal areolae (2) 3–5 in 1  $\mu$ m ( $n = 10$ ).

*Holotype*: Population in slide CHA 04/4 6–7 cm (LPC 15217), deposited in the Herbarium of the Divisi3n Ficol3gía “Dr. Sebasti3n A. Guarrera” Herbario Museo de Ciencias Naturales, La Plata.



21-27



28-34

**Figs 21–34.** *Placoneis patagonica* sp. nov. LM. (slide CHA 04/5 76 cm). Variability in valve size and symmetry. Figs 21–27. Symmetrical valves. Figs 28–34. Asymmetrical valves. Scale bar represent 10  $\mu$ m.

Partially illustrated here in LM as Figs 5–7. Leg. SALSA project (<http://www.salsa.uni-bremen.de/home.html>), 2004.

*Isotype*: BA 49154 Herbario de Plantas Celulares, Museo Argentino de Ciencias Naturales “Bernardino Rivadavia”.

*Type locality*: Laguna Cháltel (49° 57.6' S, 71°06.5' W), Province of Santa Cruz, Argentina.

*Etymology*: Dedicated to the members of the project “South Argentinian Lake Sediment Archives and Modelling” (SALSA) who collected and studied samples in which this new species was encountered.

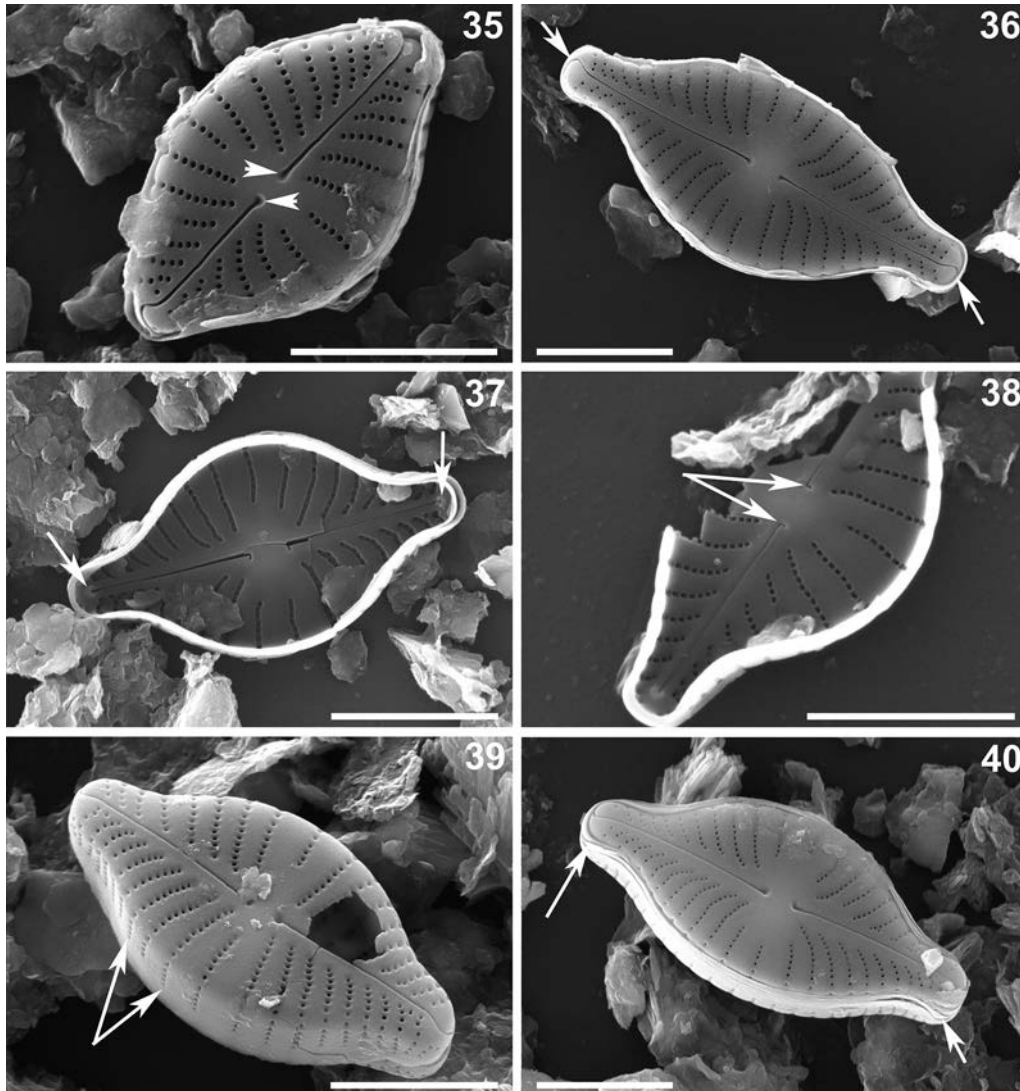
The general appearance of the valves of *C. salsae* presents some similarity with *C. dubius* (Fricke) Round in Theriot et al. (Theriot et al. 1987, Håkansson 2002), but the former differs by the absence of fultoportulae in the central area, by having fewer seriated fascicles in the valve face/mantle junction, and by the number and irregular arrangement of the areolae in the central area (Table 1).

*Cyclostephanos patagonicus* Guerrero & Echenique was described for western Patagonia in Argentina (Guerrero & Echenique 2002) and was later found in sediments of Laguna Potrok Aike in southern Santa Cruz Province (Wille et al. 2007) and Puyehue lake in Chile (quoted as *Cyclostephanos* sp. in Sterken et al. 2008). This species also has the areolae irregularly arranged in the central region, but they are denser than in *C. salsae*. Both species lack valve face fultoportulae, but in *C. salsae* there are no pores as the one observed in *C. patagonicus* (Table 1; see Guerrero & Echenique 2002, figs 14, 15).

**Table 1.** Comparison of morphological and morphometrical features of *Cyclostephanos salsae* to morphologically related species.

	<i>C. salsae</i>	<i>C. mansfeldensis</i>	<i>C. dubius</i>	<i>C. patagonicus</i>	<i>C. tholiformis</i>
	this study	Houk et al. (2014)	Houk et al. (2014)	Guerrero & Echenique (2002)	Stoermer et al. (1987)
Diameter ( $\mu\text{m}$ )	7.3–36.3	10–45	3–35	19–30	7–12
Fascicles (in 10 $\mu\text{m}$ )	3–9	5–8	8–12	5–8	12–13
Rows of areolae/fascicle at the valve/ mantle junction	(2) 3–5	(2) 3–4	(2) 3–4	3–4	3
Rows of areolae/fascicle at the mantle	4–5	Increasing in number towards the valve margin (not shown in figs.)	3–4	3–5	2–3
Location of marginal fulcportulae	Every 1–2 interfascicles	Every 1 interfascicle	Every 2–3 interfascicles	Every 1–2 interfascicles	Every 3–4 interfascicles
Number of central fulcportulae	0	Probably 0	1 or more	0	1
Striae bifurcation	Sometimes	No	No	Sometimes	Yes





**Figs 35–40.** *Placoneis patagonica* sp. nov. SEM. (slide CHA 04/5 76 cm). Fig. 35. External view. Arrow heads show depressions at raphe proximal ends. Fig. 36. External view. Arrows mark the hoe-shaped distal ends of the raphe reaching the valve mantle and curved in the same direction. Fig. 37. Internal view showing helictoglossa (arrows). Fig. 38. Broken valve in internal view. Arrows mark the proximal raphe ends bent in the same direction. Fig. 39. External view showing valve mantle and striae that are continuous from valve face to mantle. Fig. 40. External view with arrows showing the rather slender girdle bands. Scale bars represent 5  $\mu$ m.

*Cyclostephanos patagonicus*, *C. tholiformis* Stoermer et al. (Stoermer et al. 1987), *C. damasii* (Hustedt) Round (Stoermer & Håkansson 1983), and *C. salsae* can have bifurcated interstriae in the mantle, but in the first two species marginal fultoportulae are located before the bifurcation (Table 1, see Guerrero & Echenique 2002, fig. 11 and Håkansson 2002, figs 219, 220). *Cyclostephanos tholiformis* has profusely branched interstriae, with only one row of areolae between

the banches. *Cyclostephanos mansfeldensis* (Fricke) Houk et al. (Houk et al. 2014), a species living in brackish reservoirs, has a similar unordered central area, but the interstriae are not bifurcated in the mantle (Houk et al. 2014: 55; plates 184 and 185).

*Cyclostephanos salsae* was found in Laguna Cháltel in modern water samples (relative abundance 63.9%), associated with *Discostella glomerata* (Bachmann) Houk & Klee (29.9%), and *Cocconeis euglypta* Ehrenberg (3.2%). In superficial sediment samples (diatom abundance  $22 \times 10^6$  valves  $\text{cm}^{-3}$ ), *C. salsae* represents 56.9% and it is associated with *D. glomerata* (13.4%), and *C. euglypta* (8.3%).

*Cyclostephanos salsae* appeared in the uppermost 12 cm of core CHA 04/4 (Fig. 41) with relative abundances ranging between 0.08% (11 cm depth, 1665 years BP) and 83.9% (1 cm depth, 68 cal years BP) and in the uppermost 16 cm of the core CHA 04/5 (from 2500 to 177 cal years BP, Fig. 42). In these samples, *C. salsae* was found associated with *D. glomerata* (only present at 1 cm depth; 13.5%), *C. euglypta* (4–29%) and *Karayevia clevei* Bukthiyarova & Round (0–7.8%).

*Cyclostephanos salsae* is also present, although at lower percentages, in surface sediment samples from the lake shore, where it coexists with *Thalassiosira patagonica*, a species also found in other lakes in the region, with alkaline waters and high electrical conductivity (i.e.  $2900 \mu\text{S cm}^{-1}$  in Laguna Potrok Aike; Wille et al. 2007), and a new centric diatom described in this number of Nova Hedwigia as a new genus and species (Maidana et al. 2017: 63–72), that was provisionally called *Hyalodiscus* sp. by Markgraf et al. 2003. This latter diatom was also found as fossil in Cardiel Lake and was not recorded from modern samples.

On the basis of the ecological preferences of the associated diatom species (Hofmann et al. 2011, Van Dam et al. 1994) and the main water chemistry characteristics of the lake, it is thought that this new diatom also prefers slightly oligotrophic and alkaline waters.

***Placoneis patagonica* Maidana sp. nov.** Figs 21–34 (LM), 35–40 (SEM)

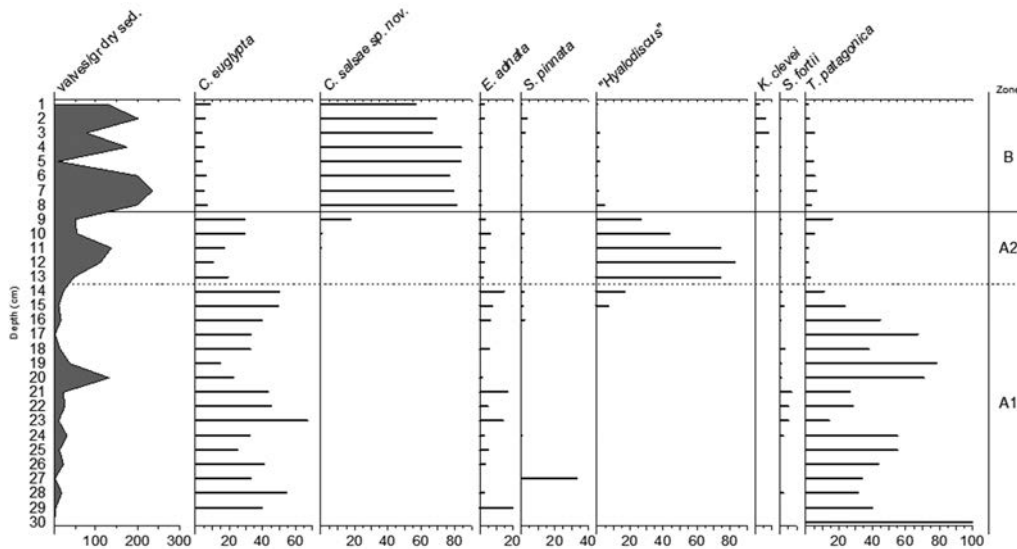
**Description:** Valves lanceolate to broadly lanceolate usually asymmetric about the apical axis, ends subrostrate in smaller specimens and rostrate to rostrate-capitate (Figs 21–34) in larger ones. Axial area narrow, linear; central area large in larger specimens, enclosed by irregularly shortened striae. Raphe filiform, somewhat sinuous. Externally, hoe-shaped, curved distal raphe fissures reaching the valve mantle (Figs 35, 36); proximal raphe fissures end in small pores in shallow depression (Figs 35). Internally, distal raphe fissures ending in conspicuous helictoglossa (Fig. 37), proximal raphe ends bend in the same direction at  $90^\circ$  (Fig. 38). Striae radiate throughout valve, not interrupted at the valve face-mantle junction and reaching valve edge (Figs 35, 36, 39, 40). Striae separated at valve center, denser toward poles (Figs 35, 36, 39, 40). Areolae small and circular in external view (Figs 35, 39), slightly quadrangular in internal view (Fig 37, 38), bearing props. Girdle bands delicate and slender (Fig. 40). Dimensions ( $n = 34$ ): length  $9.5\text{--}18.3 \mu\text{m}$  ( $14.5 \pm 2.6 \mu\text{m}$ ), width  $5.2\text{--}7.8 \mu\text{m}$  ( $6.5 \pm 0.6 \mu\text{m}$ ), striae in  $10 \mu\text{m}$   $12\text{--}18$  ( $15.1 \pm 1.5$ ), areolae in  $1 \mu\text{m}$   $5\text{--}6$ .

**Holotype:** Population in slide CHA 04/5 76 cm (LPC 15219) deposited in the Herbarium of the División Ficología “Dr. Sebastián A. Guarrera”, Herbario Museo de La Plata. Partially illustrated here in LM as Figs 21–37. Leg. SALSA project (<http://www.salsa.uni-bremen.de/home.html>), 2004.

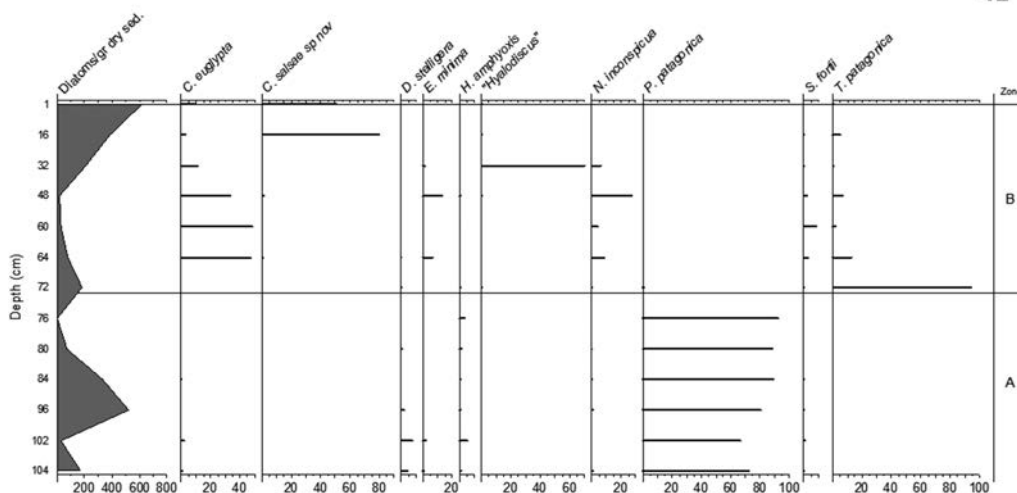
**Isotype:** BA 49156. Herbario de Plantas Celulares, Museo Argentino de Ciencias Naturales “Bernardino Rivadavia”.

**Type locality:** Laguna Cháltel ( $49^\circ 57.6' \text{ S}$ ,  $71^\circ 06.5' \text{ W}$ ), Province of Santa Cruz, Argentina.

The genus *Placoneis* Mereschkowski, a name resurrected by Cox (1987), is poorly known in Argentina. According to the catalogue of Vouilloud (2003), only 8 of the 31 species listed in Cox (2003) have been reported in Argentina (6 of them as *Navicula* species).



41



42

**Figs 41–42.** Relative abundances of diatoms (> 3%) in the studied samples. Fig. 41. Core CHA 04/4. Fig. 42. Core CHA 04/5.

It was not possible to view living cells, and thus we did not observe the chloroplasts typical of the genus (formed by two cross-shaped plates lying against each of the valves and connected by a column containing the pyrenoid). Nevertheless, like all species of the genus, *P. patagonica* has small and circular areolae in external view, and quadrangular in the internal view.

Species bearing comparable features to those of the new species presented herein have not been found so far. The lanceolate valves, usually asymmetric about the apical axis, with rostrate ends and radiate striae, more coarsely spaced at the center, are the most conspicuous features

distinguishing this taxon at the LM level. Besides these features, the lack of a stigma stands out in SEM view.

*Placoneis patagonica* was the dominant species between 72 cm (ca. 3500 y BP) and the bottom (ca. 3900 y BP) of core CHA 04/5, and was not found in water, surface sediment samples or those of core CHA 04/4. In the core section where *P. patagonica* was found, only *Discostella stelligera* (Cleve & Grunow) Houk & Klee reached relative abundances higher than 3% (Fig. 42). According to this, no much can be said about the ecological preferences of this species.

### Acknowledgements

The authors wish to thank SALSA team members for their cooperation along this study and to Prof. M. Brusa-Topham for her assistance with the English version of the manuscript.

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