

ARTICLE

Microbial filaments in stromatolites and laminites of Balbuena III Sequence (Maastrichtian/Danian) of Yacoraite Formation in Metán-Alemania Sub-basin, Salta region, Argentina, and its palaeoenvironmental significance

Filamentos microbianos em estromatólitos e laminitos da Sequência Balbuena III (Maastrichtiano/Daniano) da Formação Yacoraite na Sub-bacia Metán-Alemania, na região de Salta, Argentina, e seus significados paleoambientais

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RESUMO: A Bacia de Salta localiza-se no noroeste argentino e sua origem está relacionada a um rifte intracontinental, formado durante o Cretáceo e início do Paleógeno. Subdivide-se em quatro sub-bacias (Lomas de Olmedo a leste; Sey a oeste; Tres Cruces a norte; e Metán-Alemania a sul) que foram preenchidas por sedimentos dos Subgrupos Pirgua (fase sin-rifte), Balbuena e Santa Bárbara (fase *sag*). O Subgrupo ou Supersequência Balbuena é dividido em quatro sequências: Balbuena I, II, III e IV, da base para o topo. O presente estudo identificou filamentos microbianos em imagens geradas pelo microscópio eletrônico de varredura em amostras de estromatólitos e laminitos da Sequência Balbuena III (Maastrichtiano/Daniano) da Formação Yacoraite, na Sub-bacia Metán-Alemania, região do dique Cabra Corral, distrito de Coronel Moldes, Argentina. A ocorrência desses filamentos, identificados em calcários formados em períodos de clima árido e depositados no intervalo de lago fechado da sequência em estudo, indica a influência de atividade biológica na formação dessas rochas. A identificação dos filamentos microbianos, preservados a partir de estruturas de cianobactérias, auxilia a interpretação paleoambiental, uma vez que a ocorrência desses microorganismos é limitada à zona fótica e é comum a ambientes estressantes. Os laminitos da área de estudo foram interpretados como formados em planícies lamosas supralito-

ABSTRACT: Salta Basin is located in the northwest of Argentina and its origin is related to an intracontinental rift which was formed during the Cretaceous and early Paleogene. It is subdivided into four sub-basins (Lomas de Olmedo – east; Sey – west; Tres Cruces – north; and Metán-Alemania – south) that were filled by sediments from the Pirgua (synrift phase), Balbuena and Santa Bárbara (both in *sag* phase) Subgroups. The Subgroup or Supersequence Balbuena is divided into four sequences: Balbuena I, II, III and IV, from bottom to top. This research identified microbial filaments in scanning electron microscope images from stromatolites and laminites samples of Balbuena III Sequence (Maastrichtian/Danian), Yacoraite Formation, in the Metán-Alemania Sub-basin, situated in Cabra Corral dam region, district of Coronel Moldes, Argentina. The occurrence of these filaments, identified in limestones formed in dry weather periods and deposited in the closed lake interval of the studied sequence, indicates the influence of biological activity in the formation of these rocks. The identification of microbial filaments, preserved from cyanobacteria structures, supports the palaeoenvironmental interpretation, since the occurrence of these microorganisms is limited to the photic zone and is common to stressful environments. The laminites of the studied area have been interpreted as formed in supralittoral mudflats in the vadose zone,

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râneas na zona vadosa, enquanto os estromatólitos se formaram em ambiente sublitorâneo, estando constantemente submersos. Além dos microbialitos, ocorrem na Sequência Balbuena III fácies carbonáticas, siliciclásticas e mistas, depositadas em ambiente lacustre.

PALAVRAS-CHAVE: ambiente lacustre; sedimentologia; MEV.

while the stromatolites were formed in sublittoral environment, being constantly submerged. In addition to the microbialites, the Balbuena III Sequence consists of carbonates, siliciclastic and mixed facies which have been deposited in a lacustrine environment.

KEYWORDS: lacustrine environment; sedimentology; SEM.

INTRODUCTION

The Basin of Salta Group (Turner 1959), also called Salta Basin (Marquillas *et al.* 2005), extends from north-western Argentina, which comprises 70% of its territory, to the south of Bolivia and western Paraguay, totaling an approximate area of 150,000 km² (Del Papa & Salfity 1999). Its origin is related to an intracontinental rift of the South American plate in response to the Gondwana opening process during the Patagonidic orogenic cycle

(Keidel 1921). This process began during the Cretaceous and lasted until the end of Eocene, interrupted by the Andean orogeny (Hernandez & Echavarria 2009). Structural features (Biannucci *et al.* 1981, Salfity 1979, Salfity & Marquillas 1994) enable the division of the Salta Basin in four sub-basins (Fig. 1): Lomas de Olmedo, to east; Sey, to west; Tres Cruces, to north; and Metán-Alemania, to south, which were filled by sediments of Pirgua (synrift phase), Balbuena and Santa Bárbara (both in sag phase) subgroups.

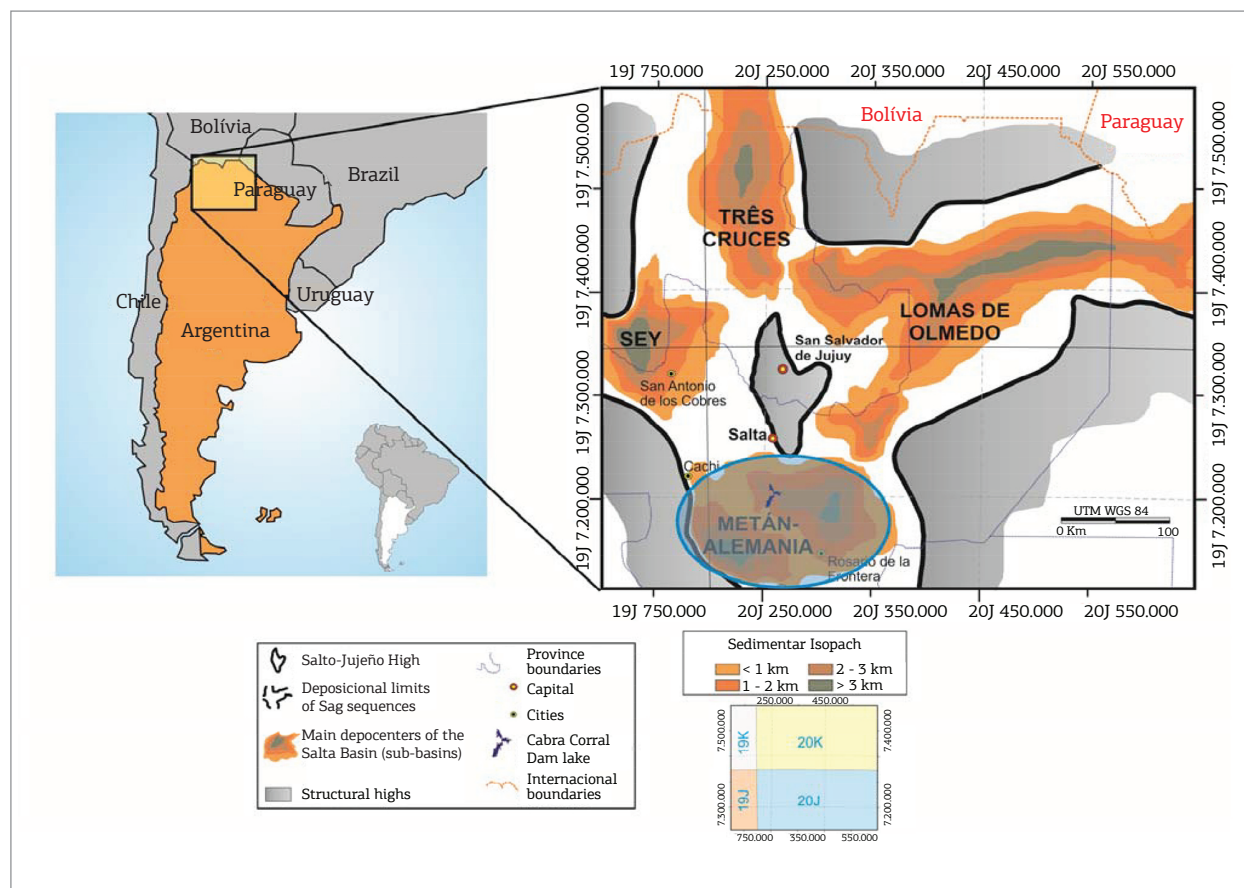


Figure 1. Location of Salta Basin and its sub-basins. In evidence (blue circle) is the Metán-Alemania Sub-basin, target of this study. Modified from Bento-Freire (2012).

In the Metán-Alemania Sub-basin, the Balbuena Subgroup is formed by the Lecho and Yacoraite formations. The Lecho Formation consists of quartz-arkosean sandstones deposited in eolic-fluvial environment. Carbonate facies associated with pelitic facies occur sporadically. Their deposits are tabular with 150 m average thickness (Salfity 1979). The Yacoraite Formation comprises tabular deposits formed in lacustrine environment with a maximum of 200 m thick (Hernandez *et al.* 1999), in which occurs carbonate, siliciclastic and mixed (with carbonate and siliciclastic components) sedimentary rocks and volcanoclastic rocks (Bento-Freire 2012, Gomes 2013, Pedrinha 2014, Roemers-Oliveira 2014).

In the Sequence Stratigraphy context, the Lecho and Yacoraite formations are equivalent to Balbuena Supersequence, which can be subdivided into four sequences, interpreted as third order, from bottom to top: Balbuena I, Balbuena II, Balbuena III and Balbuena IV (Hernandez *et al.* 1999).

The Balbuena III Sequence, target of this study, according to Roemers-Oliveira (2014), comprises siliciclastic, carbonate and mixed sedimentary rocks deposited in lacustrine environment during Maastrichtian/Danian, where climate was the main controlling agent of the lake base-level, determining the predominant type of sedimentation. The alternation of wet and dry periods caused variations in the hydrological regime of the basin. In wet weather moments, the basin behaved as an open lake, in which were deposited siliciclastic and mixed facies, and in periods of dry weather, as a closed lake, in which formed the carbonate facies. Among the carbonate facies found in the sequence studied are stromatolites and laminates, where the microbial filaments were identified.

The objective of this research was to use the features identified as a tool for sedimentological characterization of microbialites from the studied area, contributing to palaeo-environmental interpretation and facies distribution.

STUDIED AREA

The studied area is located in the Metán-Alemania Sub-basin, around Cabra Corral dam region at Coronel Moldes district, Salta Province, Argentina. Access to the studied area from the city of Salta, capital of the province, is by highway RA-51 until the junction with highway RA-68, that heads toward south, passing through the towns of Cerrillos, La Merced and El Carril before it reaches Coronel Moldes district. From this site, to the Cabra Corral Dam region is accessed by the provincial road 47 to east, which is a 10 km drive. All the way is done by paved roads, in a total distance

of 65 km. From the dam, the farthest points were accessed by back roads (Fig. 2).

METHODS

The identification of microbial filaments in stromatolites and laminites from Balbuena III Sequence occurred from samples collected in the studied area. For the selection of the samples, 14 vertical stratigraphic sections were described in a high-resolution scale (1:40) according to the method presented in Miall (2000). After its selection, the samples were analyzed by optical microscope and scanning electron microscope (SEM). To assist in mineralogical analyses, alizarin and potassium ferricyanide solution was applied on thin sections for identification and differentiation between calcite and dolomite. After applying the solution, dolomite remains colorless whereas calcite (and their polymorphs) is stained, becoming pink to pale pink (Dickson 1966). For preparation of the samples for SEM analysis, rock fragments were used with an average size of 1 cm³ covered with gold film after dehumidification in kiln. The creation of fresh surfaces aimed to avoid contamination effects or modification of the material surface before analysis. High magnification and resolution images were generated in the SEM, with embossed surfaces (three-dimensionality) formed from the electron beams emission created from heating of a tungsten capillary (Welton 1984). Further, the device coupled to SEM, energy dispersive spectrometry (EDS), was used to identify the semiquantitative chemical composition of the analyzed material.

GEOLOGICAL SETTINGS OF THE BALBUENA III SEQUENCE

The Balbuena III Sequence (BIIS) at studied area comprises siliciclastic (mudstones, siltstones, sandstones and heterolites), carbonate (calclutites, calcarenites, calcirudites, stromatolites and laminites) and mixed (marls and hybrid sandstones) sedimentary rocks and volcanoclastic rocks (tuff) from Yacoraite Formation of the Balbuena Supersequence or Subgroup (Salta Basin) deposited in lacustrine environment (Roemers-Oliveira 2014).

The BIIS differs from the other sequences of Balbuena Supersequence by presenting predominance of siliciclastic facies, especially pelitic. There is a large proportion of clay and silty rocks, so that such a sequence is common in the literature to be referred to as "Silt-clay Section" (Hernandez *et al.* 1999).

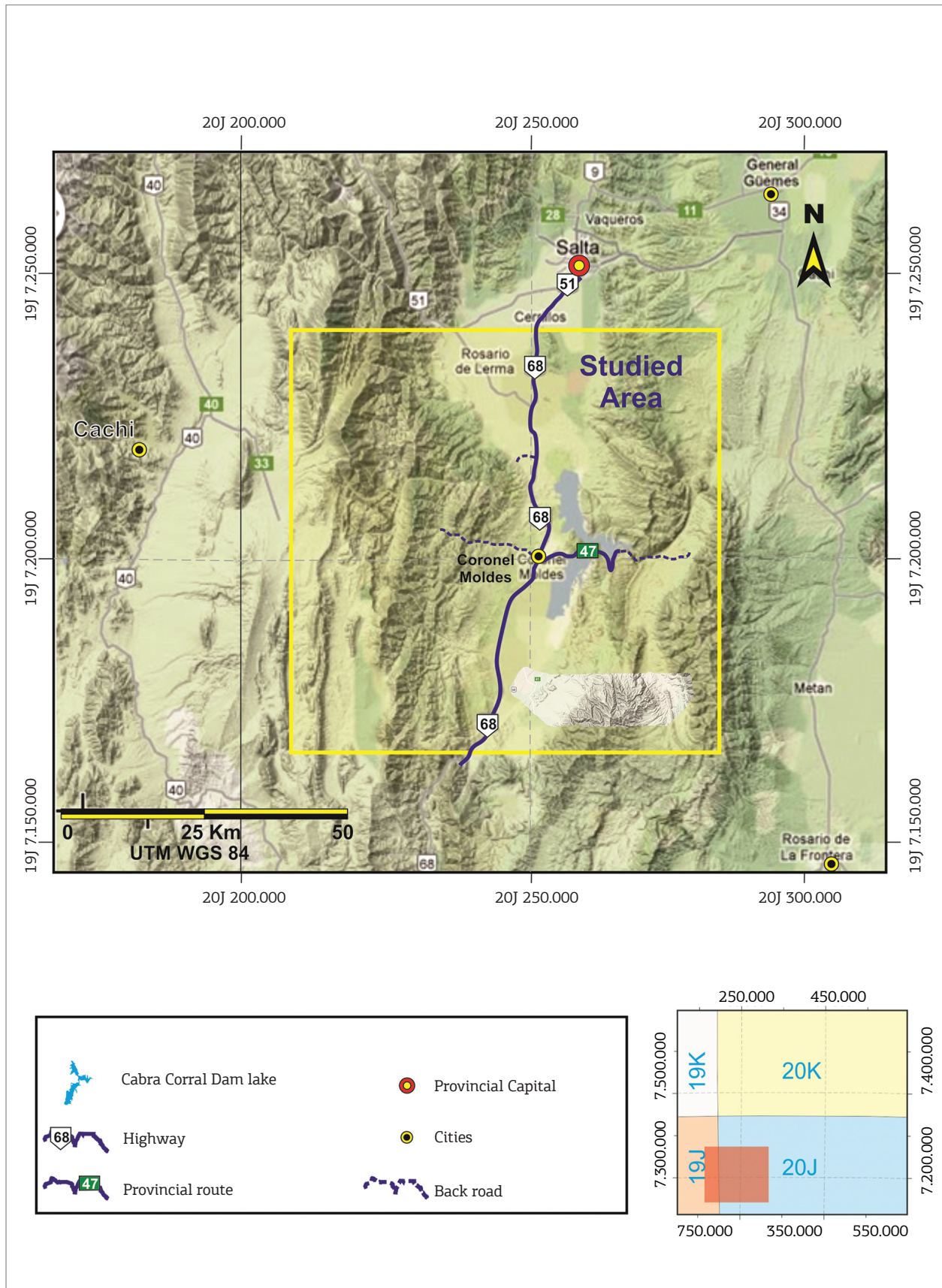


Figure 2. Access map to studied area (yellow square) adjacent to the Cabra Corral Dam, Salta Province, Argentina. Topographic map from Google® Maps Brazil. Modified from Bento-Freire (2012).

The BIIS thicknesses in the measured sections ranged from 30 to 35 m and, in general, on a single outcrop it is possible to have all the sequence exposed. Due to the Andean tectonic, the strata are tilted with an average slope of 30° to 40° in relation to its deposition (horizontal).

At the studied area, the BIIS is relatively easy to recognize in the field, since it has marker beds that facilitate its identification and the correlation between different outcrops. In the bottom of the sequence, there is a red siltstone bed of up to 3 m thick, in which, according to stable isotopes studies ($\delta^{18}\text{O}$ and $\delta^{13}\text{C}$) from Marquillas *et al.* (2003, 2007), it represents the K/Pg (Cretaceous/Paleogene) boundary (red dashed line shown in Fig. 3A). Around the middle of the BIIS occurs a columnar stromatolite bed that is also an excellent marker (Fig. 3B). Another marker bed is at the top of the section (in fact already in Balbuena IV Sequence – by Hernández *et al.* 1999), where there is a limestone bed between mudstones. In this bed, typical features of dissolution by ascending fluids can be found (observe inverted “teardrop” pore shape created by dissolution in Fig. 3C – yellow arrow). In a few centimeters above this bed a tuff layer 15 to 20

cm thick is frequently found in this stratigraphic position (Fig. 3C).

An important characteristic of the BIIS is the variation of predominant facies. From the bottom to approximately 2/3 of the section occurs predominantly siliciclastic facies, while in the upper 1/3, the carbonate facies predominate (Figs. 4 and 5). This variation in the prevalence of facies reflects changes in hydrologic and climatic regimes that directly influenced the deposition of the lake. The siliciclastic facies were formed at open lake hydrologic regime during periods of wet weather, while carbonate facies were originated at a closed lake system during periods of dry weather (Roemers-Oliveira 2014).

Figure 4 shows the vertical stratigraphic type-section of BIIS, with rock types, primary and secondary sedimentary structures and major components observed. Figure 5 presents the patterns of total and spectral natural gamma ray logs collected from the type-section at the sequence under study. In the type-section, the response of the spectral range for thorium consistently shows the variation of predominance of facies: the siliciclastic facies in the lower portion of BIIS shows highest values than the carbonatic facies.

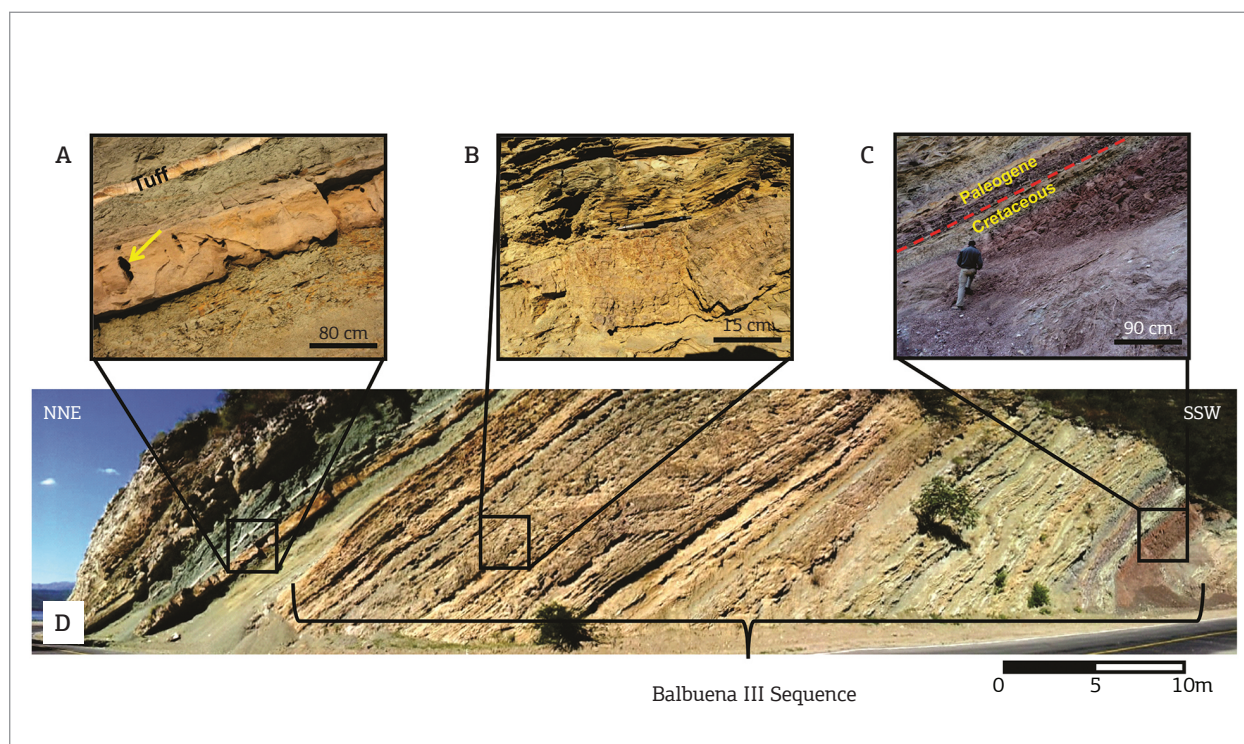


Figure 3. Marker beds of Balbuena III Sequence: (A) red siltstone bed with K/Pg (Cretaceous/Paleogene) boundary (red dashed line); (B) columnar stromatolite; and (C) typical features of dissolution by ascending fluids (yellow arrow) in limestone and tuff layer. Assado outcrop (13.5 km, provincial route 47, coordinates: 20J 0260922/7202232).

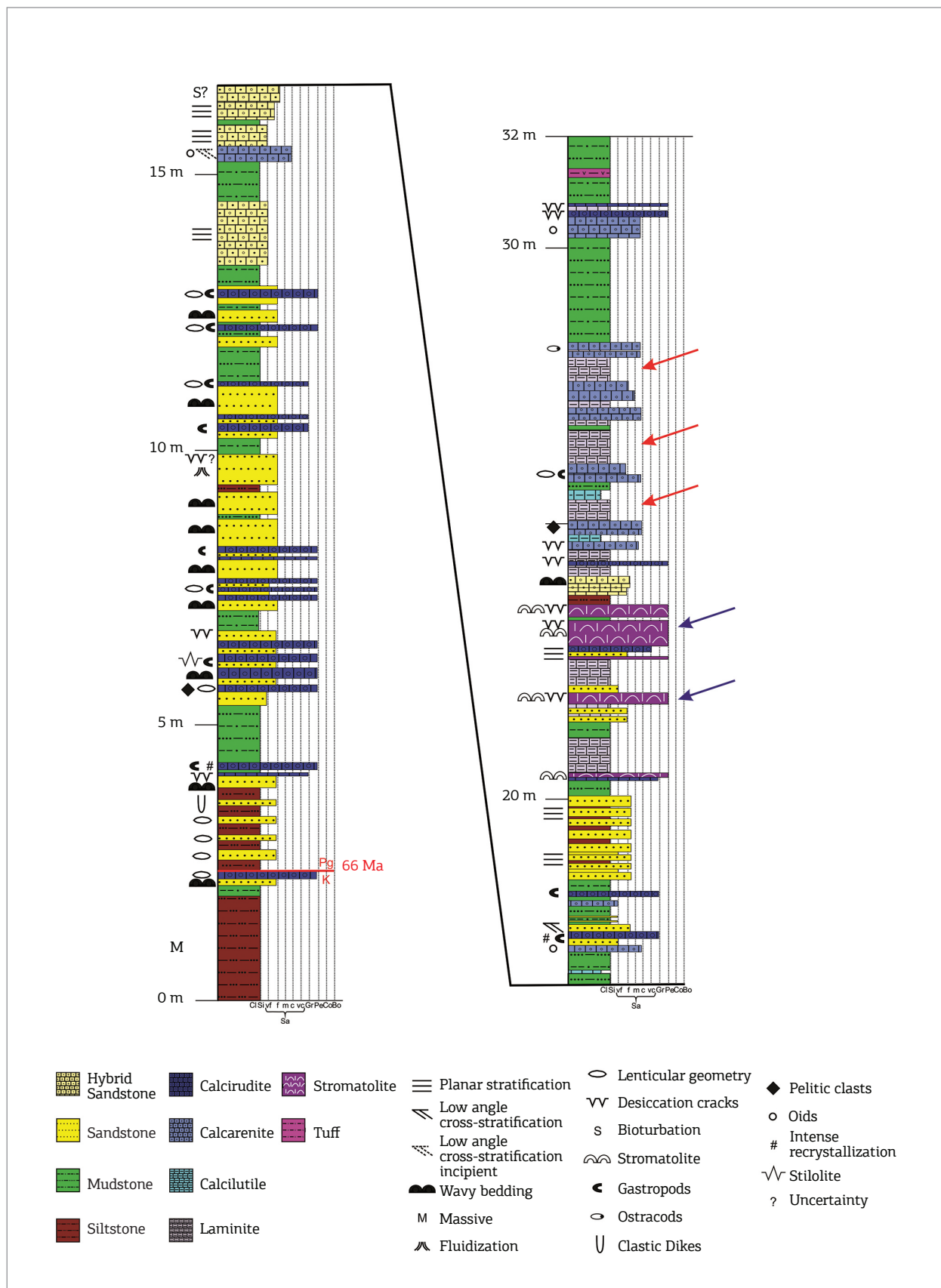


Figure 4. Balbuena III Sequence type-section (Vapumas outcrop – coordinates: 20J 0260670/7202127). Note the K/Pg boundary (66 Ma) in the base of the section (red line). Red arrows indicate the occurrence of laminites and blue arrows of stromatolites. Grain size classification by Wentworth (1922).

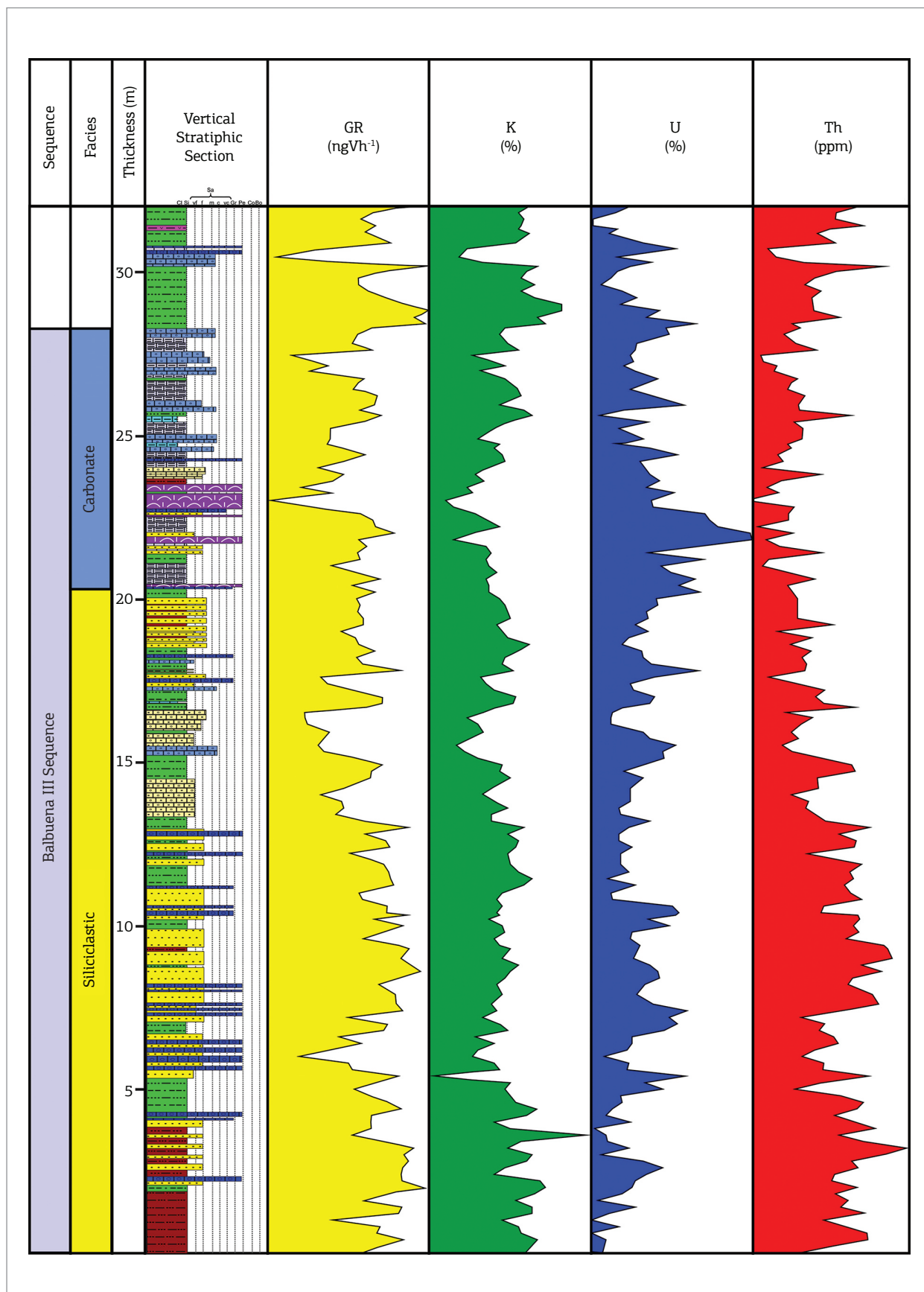


Figure 5. Patterns of total and spectral natural gamma ray logs to the Balbuena III Sequence type-section. Symbols of lithotypes according to Fig. 4.

RESULTS AND DISCUSSION

Lakes are dynamic systems and very susceptible to changes in relation to the climate, water chemical, bathymetry and tectonic settings, resulting in significant variations of litho and biofacies, and their lateral and vertical distribution patterns (Flügel 2010).

Hydrologically, lakes can be classified as open or closed lakes (Flügel 2010, Martínez & Meléndez 2010, Neumann *et al.* 2008, Platt & Wright 1991, Tucker & Wright 1990). Open lakes, commonly associated with humid climates, have relatively stable shorelines once they have permanent tributaries and the rivers and rain inputs are in equilibrium with evaporation. The clastic material from river influx dominates, but some also present chemical and biogenic precipitations. On the other hand, closed lakes have moving shorelines, do not have stable tributaries and how they are common in regions with semiarid warm and hot climates, they have negative water and chemical balances.

Due to the high ionic concentration of calcium carbonate associated with the limited fluvial sediments input, the carbonate facies are formed predominantly within close lakes. Therefore, stromatolites and laminites, target of this study, were interpreted as formed in arid climate periods in a hydrologically closed lake.

Microbial filaments identified within microbialites samples in the studied area indicate these rocks are organosedimentary deposits originated at littoral zone in shallow waters (photic zone) in which the bioherms and/or biostromes extend over large areas since they could be traced for tens of kilometers in the basin.

In this study, stromatolite was adopted as presented by Riding (1991, 1999, 2000), corresponding to a laminated deposit originated from benthic microbial communities. This definition corresponds to an update of the original concept, proposed by Kalkowsky (1908), which says stromatolite corresponds to organic laminated deposit.

Fine-grained (silt/clay) carbonate rocks formed by recurrence of laminae with wavy surfaces are named as crenulated laminites as presented by Terra *et al.* (2010). According to these authors, these structures are indicative of microbial origin.

Following, short macro and microscopic descriptions for each of these lithotypes, in which microbial filaments were identified, are presented.

Columnar stromatolites

The stromatolites from BIIIS are columnar, forming biostromes, since they are laterally continuous. They are also excellent marker beds, once they occur restricted to

certain stratigraphic horizons. In all the outcrops studied, at least two stromatolites intervals are identified, with thicknesses ranging from 20 to 50 cm (Figs. 6A and 6B). The lower stratum (Fig. 6C) is characterized by tabular stromatolites, corresponding to the type-LLH (Laterally linked hemispheroids) from Logan *et al.* (1964) classification. In the upper stratum, the stromatolites are preferably domical (Fig. 6D), corresponding to type-SH (vertically stacked hemispheroids) from the same classification. Rarely the stromatolites can be identified as bioherms, which are lenticular up to 50 cm long and 15 cm thick (at build-up axis) (Figs. 6E and 6F).

Macroscopically, both LLH and SH stromatolites types are laminated with columnar internal structure (Figs. 7A and B). In plan view, they present cerebriform aspect (Fig. 7C). Desiccation cracks at the top of build-ups are common (Fig. 7D).

In thin section (Fig. 8), both stromatolite types (LLH and SH) have very similar features. They are laminated into convex columnar structures up to 5 mm in length (Figs. 8A and 8B), alternating millimeter intervals of micrite and fine sand-sized peloids (Figs. 8C and 8D) and intervals with calcite fascicular crystals up to 1 mm in length (yellow arrows in Figs. 8E and 8F). Rarely there are quartz grains. High growth-framework primary porosity type (Figs. 8E and 8F) from Choquette & Pray (1970) porosity classification is observed in these rocks.

Crenulated laminites

The laminites (Fig. 9) may occur associated with carbonate muddy or stromatolitic facies. In some cases, the lamination is discontinued/brecciated by effects of sub-aerial exposure. Thin sections show that the laminites have calcite lamellar crystals (defined by alizarin staining) intercalated with micrite in crenulated laminations (wavy) (Fig. 9B). This lithotype is quartz-rich and may have up to 25% of angular to rounded very fine sand/silt-sized quartz grains. Very fine sand-sized ostracods carapaces, normally disarticulated, and submillimeter particles of muscovite crystals occur as accessories (<5%). The occurrence of ostracods disjointed shields indicates they were transported and deposited away from their living environment. The laminites have low-porosity, fenestral type (according to Choquette & Pray 1970 porosity classification).

Microbial filaments

Microbial filaments were identified in stromatolites and laminites from Balbuena III Sequence by semiquantitative analysis of the chemical composition of the material analyzed by EDS (which indicated that the filaments are partially

replaced by calcite) and comparison of SEM images with literature examples (*e.g.* Folk 1993).

Based on the depositional environment context, facies associations, associated biota, EDS data and comparison to literature examples, the filaments found were interpreted as

EPS preserved by permineralization. The EPS (Extracellular Polymeric Substance) is the main organic product of the cyanobacteria.

According to Golubic and Seong-Joo (1999), the record fossil of cyanobacteria is one of the oldest in

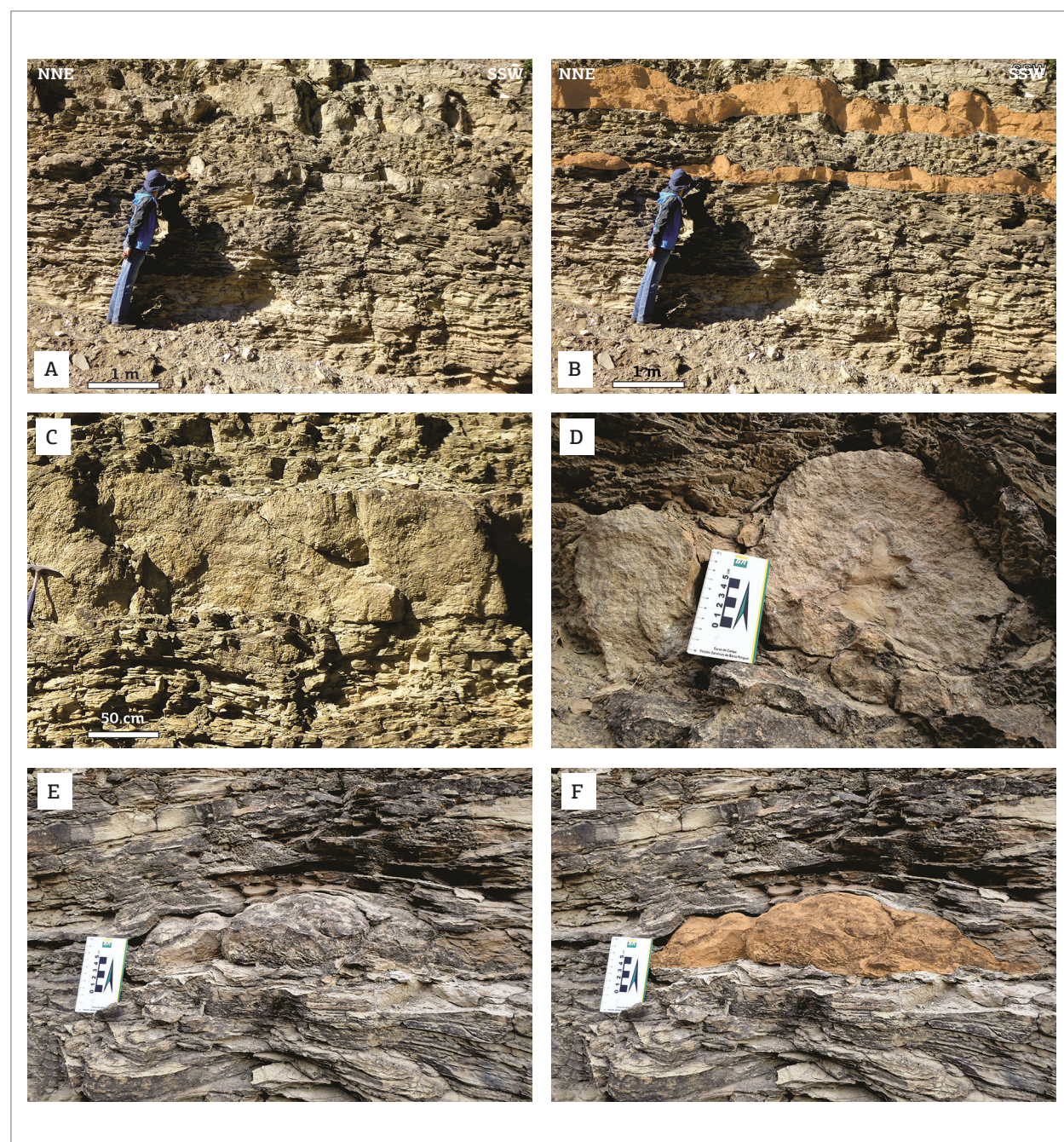


Figure 6. Stromatolites from Balbuena III Sequence at Lomito outcrop (coordinates: 20J 0262343/7201424). (A) Biostromes of stromatolites – original photo; (B) biostromes of stromatolites – interpreted levels; (C) type-LLH stromatolite classified by Logan *et al.* (1964); (D) type-SH stromatolite by Logan *et al.* (1964) classification; (E) stromatolitic bioherm between crenulated laminites – original photo; (F) stromatolitic bioherm – lens detached.

geological record, possibly reaching back to 3.5 Ga ago. In comparative studies with cyanobacteria in modern environments, the general trend in the group evolution is gradually increasing complexity and diversity through geological time, although their morphological features have been maintained.

Figures 10 and 11 show images obtained from Scanning Electron Microscope from columnar stromatolite (Fig. 10) and crenulated laminite (Fig. 11).

Figure 10 shows that the microbial filaments occur amongst calcite rhombohedral crystals (composition verified by EDS) and clay minerals. Figure 11 presents SEM images of crenulated laminite with calcite crystals (micrite) ranging from 1 to 20 microns in length. It is also observed the occurrence of microbial filaments partially replaced by calcite (Figs. 11B and 11D). Some examples of similar structures interpreted as filamentous organic matter/EPS remains

in microbialites from Brazil can be found in Bahniuk *et al.* (2015) and Catto (2015) and within stromatolites from England in Perri *et al.* (2013).

Table 1 provides three semiquantitative results of chemical composition (wt%) obtained by EDS in stromatolite (numbers 1 and 2 – Figs. 10A and 10C) and laminite (number 3 – Fig. 11D). Analyzing the results, it is observed that microbial filaments have relatively high carbon content, indicative of organic origin, but also with considerable content of calcium, magnesium and oxygen, showing that filaments were partly calcitized. The partial replacement by calcite proves that it is a fossil material. Values for aluminum, silicon, potassium and iron may correspond to influences of adjacent materials to the analysis spots such as clay minerals and quartz, affecting the EDS results. Figure 12 shows spectral profiles for each of the three points analyzed by EDS.

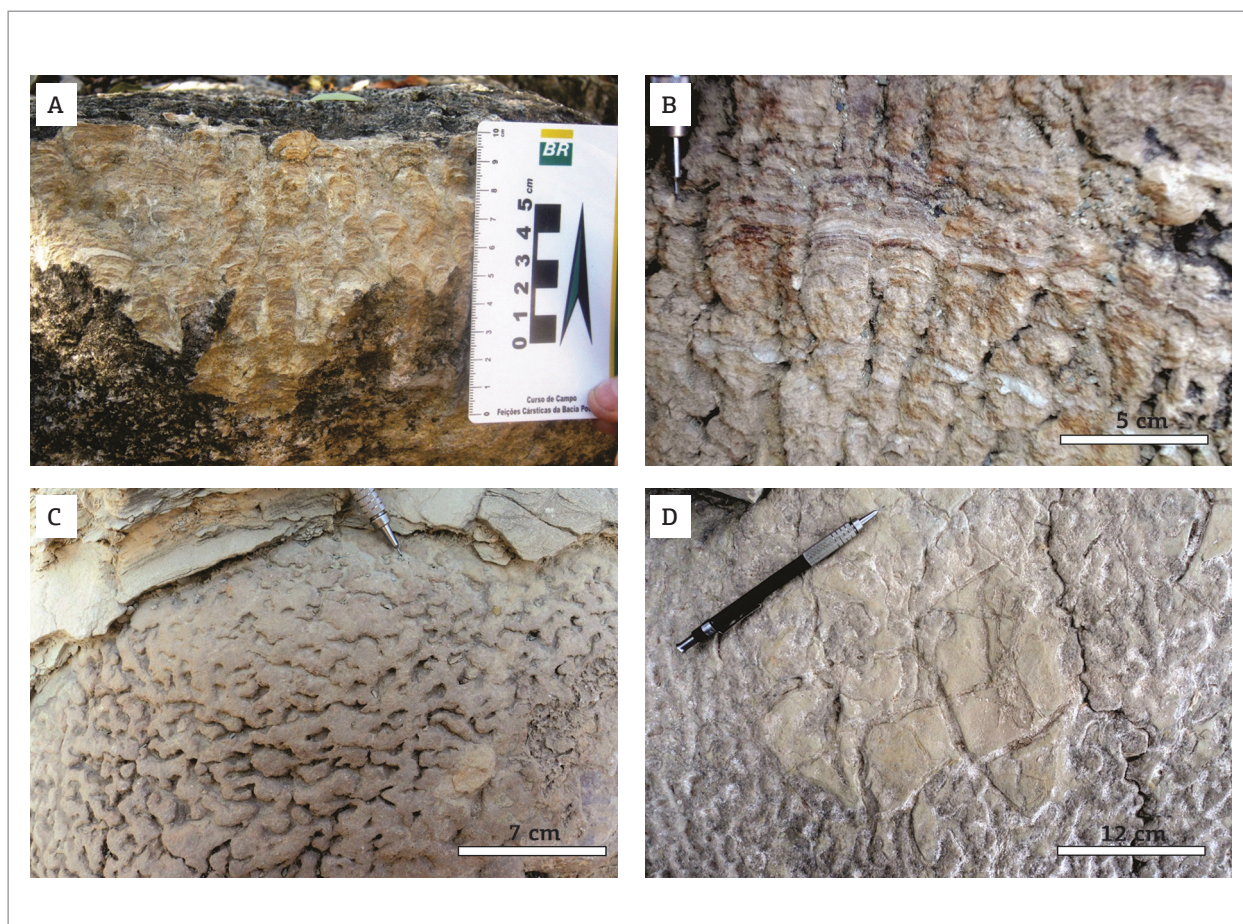


Figure 7. Photographs of stromatolites. (A) Columnar internal structure at Dique Compensador Ravina outcrop (coordinates 20J 0268101/7200325); (B) columnar internal structure at Vapumas outcrop (coordinates 20J 0260670/7202127); (C) cerebroid aspect (plan view) at Assado outcrop (coordinates 20J 0260922/7202232); (D) desiccation cracks (plan view) at the top of the stromatolite build-up at Astronauta outcrop (coordinates 0258453/7204027).

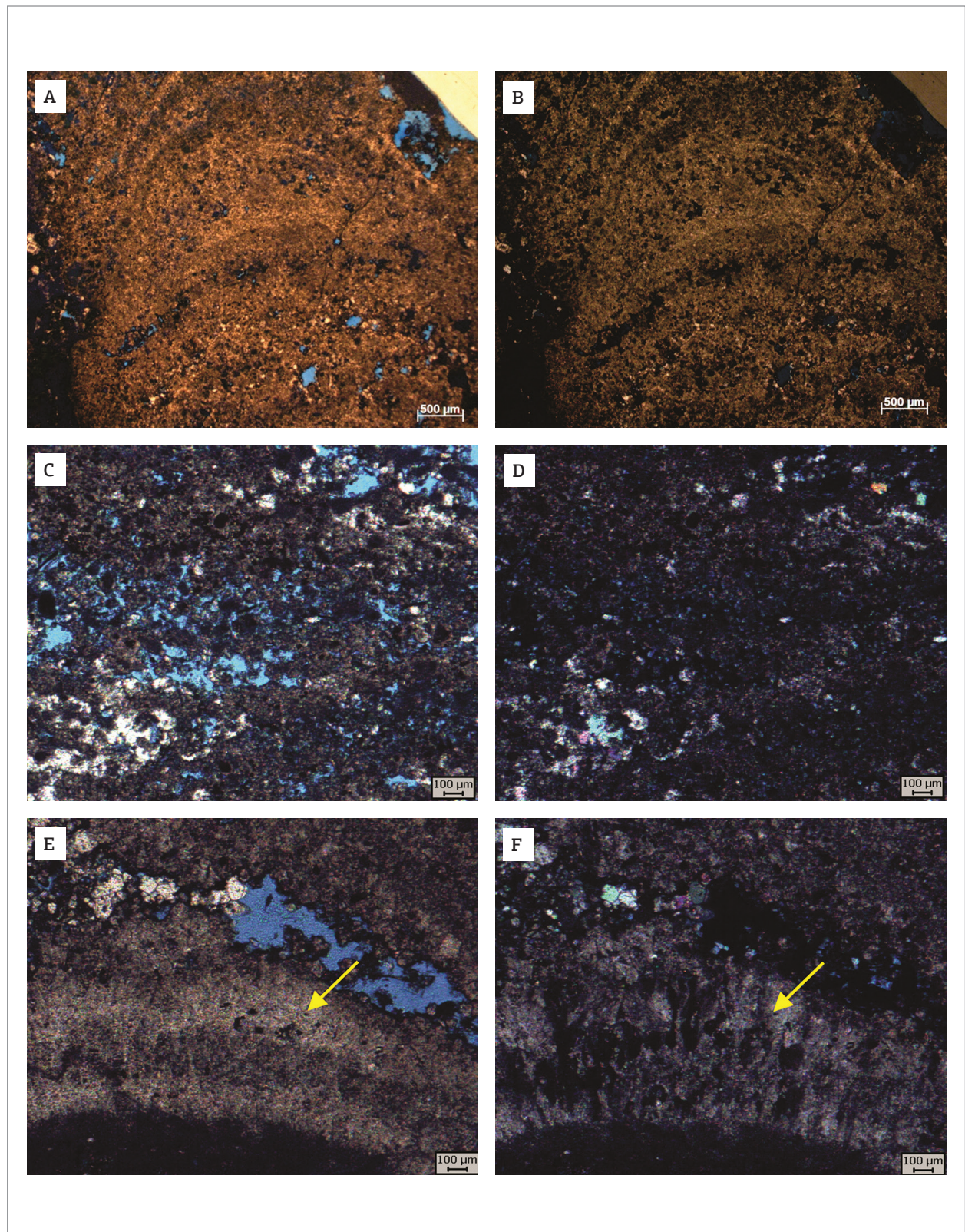


Figure 8. Photomicrographs of columnar stromatolites. (A and B): Columnar laminated structure. Sample from Tartaruga Outcrop (coordinates 20J 0256158/7195458); (C and D): peloidal portion with high porosity. Sample from Enseada outcrop (coordinates 20J 0261590/7202266); (E and F): calcite fascicular crystals level (yellow arrows) with growth-framework porosity type. Sample from Enseada outcrop. (A, C and E): parallel polarizers; (B), (D) and (F): crossed polarizers.

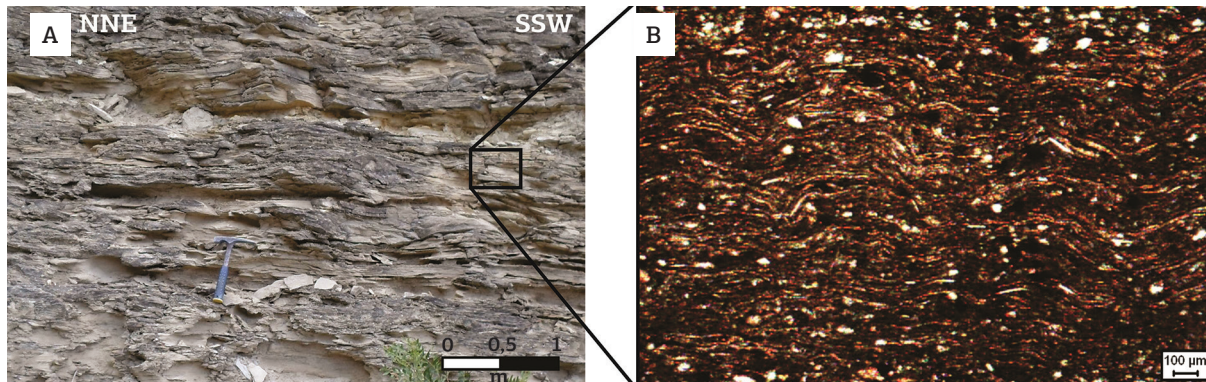


Figure 9. Crenulated laminite at Finca el Retiro outcrop (coordinates 20J 0260135/7202159). (A) Occurrence of crenulated laminite in the outcrop; (B): crenulated laminite photomicrograph. Sample is stained by alizarin and shown with parallel polarizers.

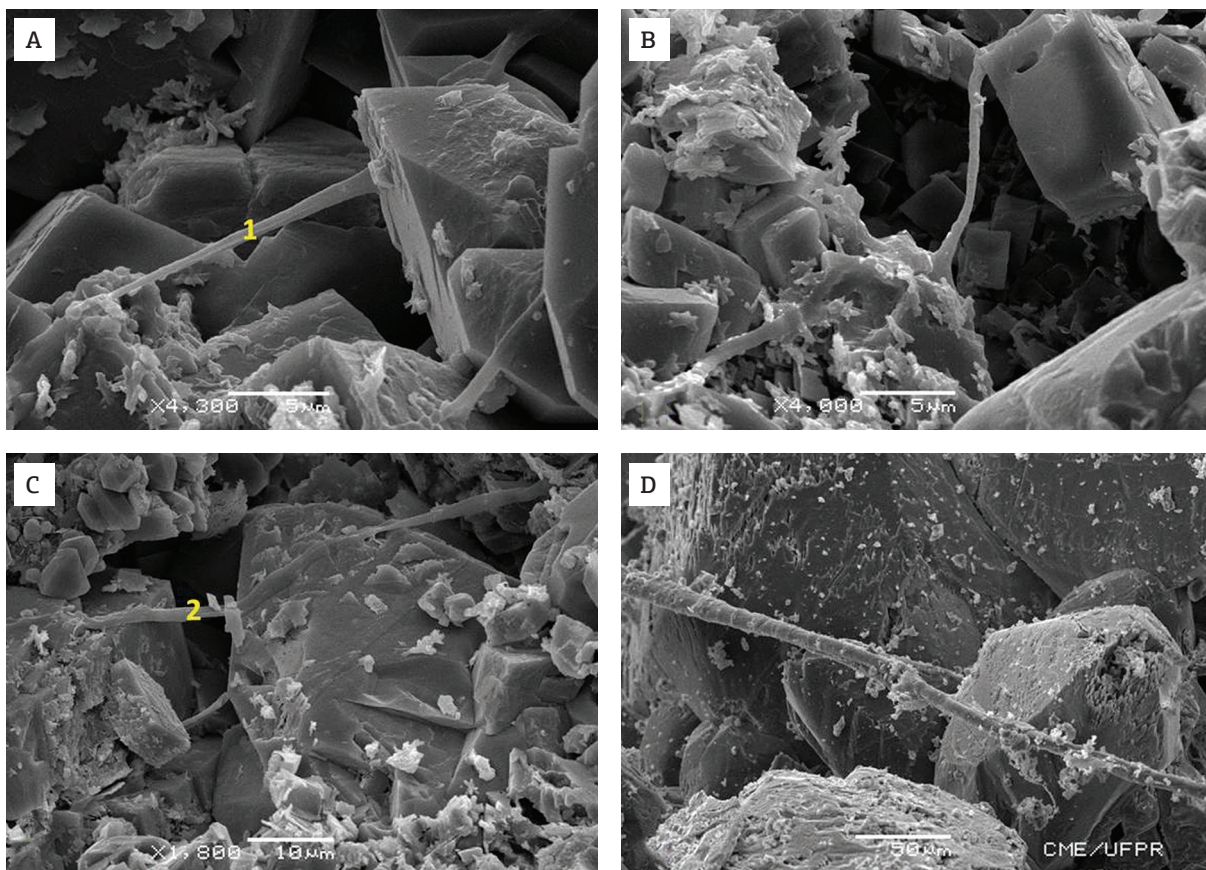


Figure 10. Microbial filaments within the columnar stromatolites. Scanning electron microscope images of fresh surfaces from sample of Tartaruga outcrop (coordinates 20J 0256158/7195458). The numbers 1 (in A) and 2 (in C) corresponding to energy dispersive spectrometry spots.

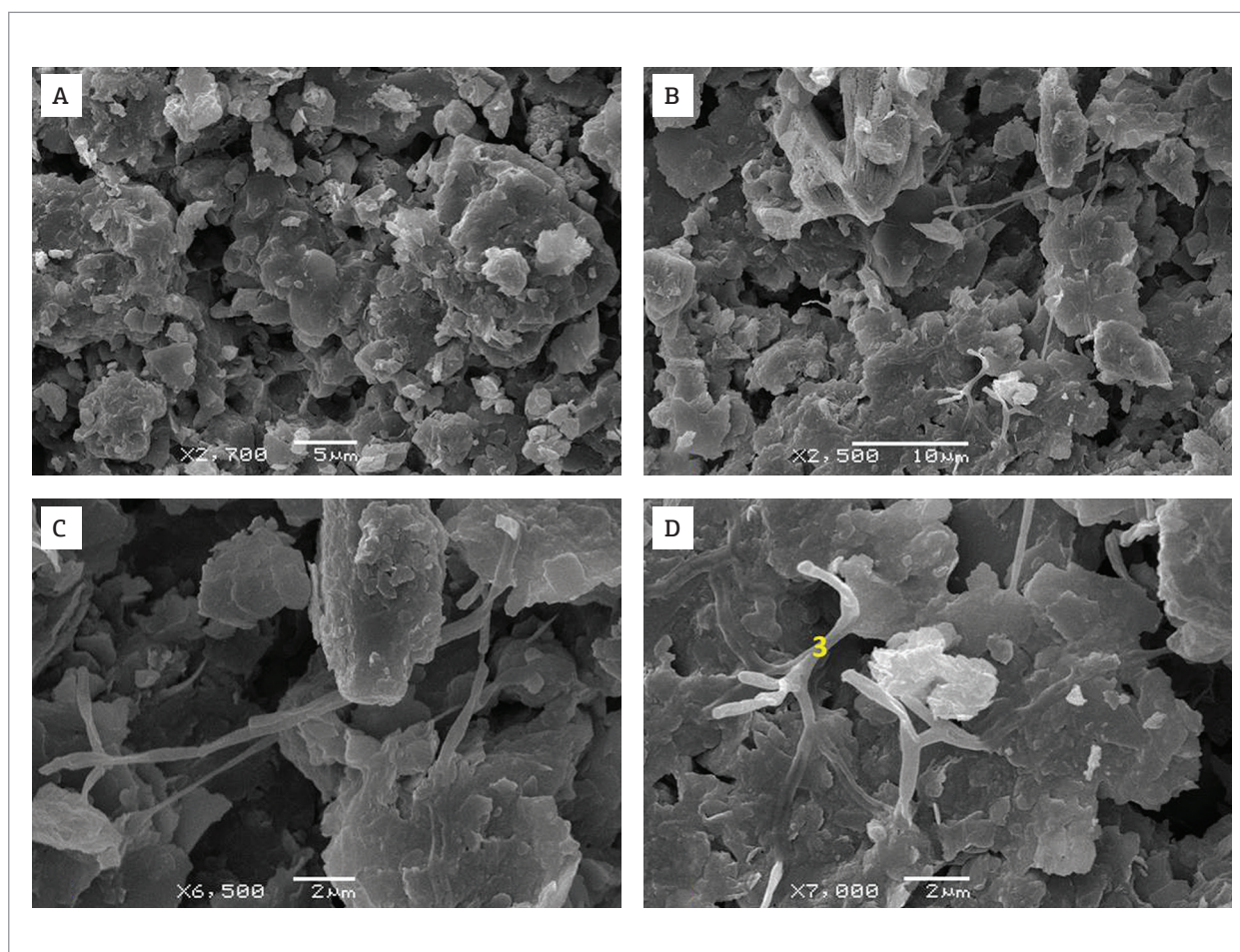


Figure 11. Scanning electron microscope images in crenulated laminite of fresh surfaces. Observe occurrence of microbial filaments in B, C and D. Sample from Vapumas outcrop (coordinates 20J 0260670/7202127). Number 3 (in D) is an energy dispersive spectrometry spot.

Table 1. Energy dispersive spectrometry semiquantitative results for carbon, oxygen, magnesium, aluminum, silicon, potassium, calcium and iron (in weight %) from stromatolite and laminite samples of Balbuena III Sequence

EDS spot	Lithotype	wt%							
		C	O	Mg	Al	Si	K	Ca	Fe
1	Stromatolite	36.13	25.00	1.73	-	10.06	-	20.43	6.65
2	Stromatolite	39.25	21.21	2.55	-	4.10	0.77	26.73	5.38
3	Laminite	50.6	11.13	0.57	3.79	21.9	2.56	2.47	6.97

CONCLUSIONS

The occurrence of microbial filaments in stromatolites and laminites indicates the influence of organic activity in their sedimentation. Both, columnar stromatolites and crenulated laminites from Balbuena III Sequence, may be interpreted as microbially (probably by cyanobacteria) induced sedimentary deposits.

The interaction of biological activity in the formation of stromatolites and laminites denotes their occurrences are restricted to the photic zone, since cyanobacteria require light to perform photosynthesis. Therefore, stromatolites and laminites are lithotypes that will occur in coastal areas in lacustrine environment under shallow waters. Stromatolites will form in a slightly deeper environment where there is enough space available to the generation of domes. Laminites will

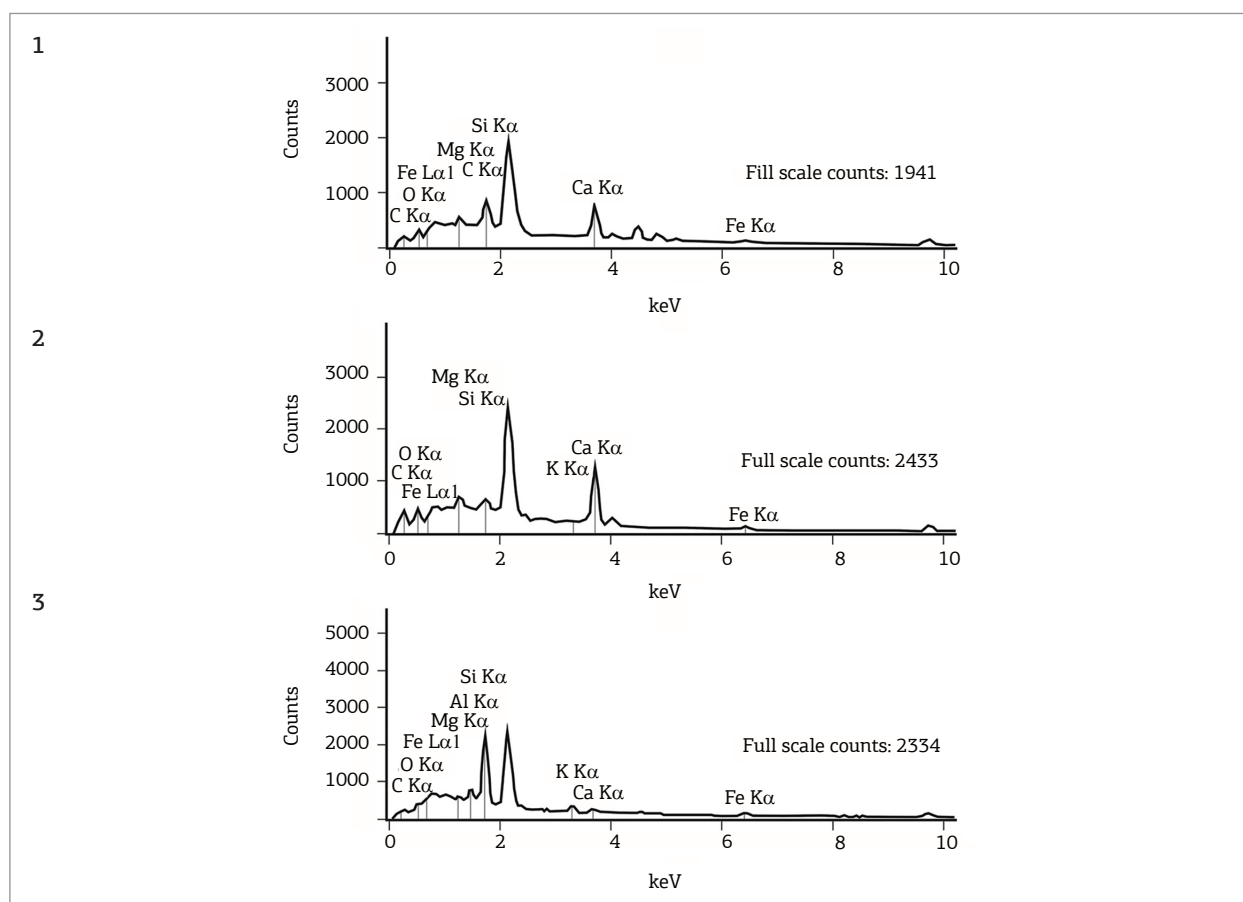


Figure 12. Spectral profiles of the elements identified by energy dispersive spectrometry semiquantitative chemical analysis of microbial filaments in stromatolite (1 and 2) and laminite (3). keV = kiloelectronvolt.

form in mudflat supralittoral zone with few space available, only allowing the formation of crenulated surfaces.

The occurrence of bioinduced deposits in intervals of Balbuena III Sequence interpreted as being a closed lake is consistent with the development of microbial activity. In a closed lake, the water balance is negative (evaporation > precipitation), and the input of extrabacinal sediments is limited, favoring carbonate deposition (the basin is supersaturated of calcium carbonate) and proliferation of microbial activity, since the environment becomes more stressful.

Occurrence of ostracods confirms the increased environmental stress caused by increase of evaporation.

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