Holocene Vegetation and Climate inferences from Phytoliths and Pollen from *Lagoa do Macuco*, North Coast of Espírito Santo State (Brazil)

Inferências sobre Vegetação e Clima no Holoceno a partir de Fitólitos e Pólen da Lagoa do Macuco, Litoral Norte do Estado do Espírito Santo (Brasil)

[ae]**Marcia Regina Calegari**, [bf]**Marco Madella**, [cg]**Antonio Alvaro Buso Jr**, [dh]**Margaritha L Osterrieth**, [ci]**Flávio Lima Lorente**, [cj]**Luiz Carlos R. Pessenda**

[a]Colegiado de Geografia , Universidade Estadual do Oeste do Paraná - UNIOESTE - *Campus* Marechal Cândido Rondon, [b]ICREA - Complexity and Socio-Ecological Dynamics (CaSEs), Department of Humanities Universitat Pompeu Fabra – IMF Consejo Superior de Investigación Cientifica (CSIC), [c]Laboratório de Carbono-14, CENA/USP, [d]Centro de Geología de Costas y del Cuaternario FCEyN, Universidad Nacional de Mar del Plata

[e]marciareg_calegari@hotmail.com, [f]marco.madella@icrea.cat, [g]aabuso@cena.usp.br, [h]mosterrii@hotmail.com, [i]flimalorente@yahoo.com.br, [j]pessenda@cena.usp.br

Resumo

Uma abordagem *interproxy* focada na análise de fitólitos e de grãos de pólen, também incluindo datação ^{14}C e análise granulométrica de sedimentos, foi aplicada ao estudo da Lagoa do Macuco, em Linhares no Estado do Espírito Santo. Foram identificadas três zonas polínicas definidas com base nas mudanças do registro polínico, a primeira (MAC-C I) indicando a presença de uma vegetação de mangue entre 7700 anos cal AP e 4396 anos cal AP, a segunda (MAC-C II) que representa o deslocamento da vegetação das áreas mais baixas para as partes marginais mais elevadas do vale (4396 anos cal AP - 1287 anos cal AP) e a terceira (MAC-C III) que representa o período da instalação do atual lago, entre 1287 anos cal AP e os dias atuais. O deslocamento do mangue parece estar conectado a migração em direção ao paleo-estuário e o alagamento das partes baixas do vale, refletindo a elevação do nível relativo do mar depois de 7000 cal anos AP. A análise fitolítica indicou quatro zonas principais. A zona fitolítica I (7700-7100 anos cal AP), não apresentou fitólitos. A zona fitolítica II (7100-3400 anos cal AP) indica a presença de uma vegetação predominantemente composta por gramíneas, enquanto a zona fitolítica III (3400-400 anos cal AP) mostrou uma fase com uma vegetação mais florestada. Finalmente, a zona fitolítica IV, (400 anos cal AP até os dias atuais) apresentou decréscimo da cobertura arbórea e uma estrutura de vegetação com forte presença de gramíneas em sua composição. Os índices fitolíticos indicaram tendência de umidade e temperatura em escala local permitindo a compreensão das condições ambientais durante a formação do lago atual

Palavras-chave: opala biogênica; pólen; índices fitolíticos; vegetação; Holoceno

Abstract

An interproxy approach focused on phytoliths and pollen, including radiocarbon dating and sediment particle-size analyses, has been carried out at Lagoa do Macuco, Linhares municipality, Espírito Santo State, Brazil. Three pollen zones were identified: the first one (MAC-C I) indicates the presence of mangrove vegetation between 7700 cal yr BP and 4396 cal yr BP; the second one a displacement of the vegetation from the lower areas to the more elevated margins of the valley (MAC-C II, from between 4396 to 1287 cal. yr BP) and third one (MAC-C III), representing the period of the current lake evolution, between 1287 cal. yr BP and the present day. The displacement of the mangrove seems to be connected to the landward migration of the palaeo-estuary and the flooding of the lower parts of the valley, reflecting the relative sea-level highstand, which occurred after 7000 cal yr BP. The phytolith analysis indicated four main zone. The phytolith zone I (7700-7100 cal yr BP) did not show the presence of phytoliths. The phytoliths zone II (7100-3400 cal yr BP) indicates the presence of an open vegetation predominantly of grasses while phytolith zone III (3400-400 cal yr BP) shows a phase with more forested vegetation. Finally, the phytolith zone IV (400 cal yr BP to modern) has a decrease in tree cover and the setting of vegetation with a strong grasses component. Phytoliths indices indicate trends of humidity and temperature at a more local scale, allowing understanding the environmental conditions during the formation of the current lake.

Key-words: biogenic opal; pollen, phytoliths índices; vegetation; Holocene

1. Introduction

The use of interproxies (biotic and abiotic) strategy strongly enhances the study of past environments (e.g. Curtis *et al.* 1998, Birks & Birks 2006, Buso Jr. *et al.* 2013) since they provide distinct complementary information, reinforcing and extending our interpretative power. The combination of two (strong) vegetation proxies, such as pollen and phytoliths, increases the capability of gathering information about regional and local past vegetation communities, reflecting species migration and ecological successions after disturbances, environmental change, and global climatic changes.

In the Neotropic pollen analysis has already being proved to be an important proxy from waterlogged sediments while phytoliths were successfully used in both waterlogged and terrestrial deposits (e.g. Bartlett & Barghoorn 1973, Piperno 1993, Alexandre *et al.* 1997b, Mworia-Maitima 1997, Piperno & Pearsall 1998, Piperno & Jones 2003, Thorn 2004, Penny & Kealhofer 2005, Piperno 2006). In Brazil, there are surprisingly few works focusing on phytolith (Kondo & Iwasa 1981, Alexandre & Meunier 1999, Borba-Roschel *et al.* 2006, Calegari *et al.* 2013a, Coe *et al.* 2013) and none in which pollen and phytoliths have been analysed together.

The objective of the present study was to exploit/use the combined potential of opal phytoliths and pollen from the lagoon sediments of Macuco to understand the Holocene vegetation history and palaeoclimatic conditions in a coastal *Mata Atlântica* forest (Ombrophylous Dense Forest).

2. Regional setting

Lagoa do Macuco (19°02.584'S/39°56.695'W) is a freshwater lake in the lower course of Barra Seca River (figure 1). The lake is set in an incised valley 23 Km from the sea, along the southeastern coast of Brazil. The valley cuts the Barreiras Formation, a sedimentary domain composed by sandstones, conglomerates and mudstones mainly of Neogene fluvial and alluvial fan deposits with possible inclusions from coastal onlaps associated with Neogene marine transgressions (Arai 2006, Dominguez 2009).

The vegetation developed on the Barreiras Formation is largely composed by the *Mata Atlantica* tropical rainforest. The most representative plant families of this biome are Fabaceae, Myrtaceae, Sapotaceae, Bignoniaceae, Lauraceae, Hippocrateaceae, Euphorbiaceae, Annonaceae and Apocynaceae (Peixoto & Gentry 1990). The vegetation of the lake and its surroundings comprises trees such as *Tabebuia cassinoides* (Lam.) DC., *Alchornea triplinervia* (Spreng.) Müll.Arg. and *Cecropia* sp., and herbaceous plants from the Cyperaceae, Poaceae (e.g. *Typha* sp.) and Pteridophytae as well as floating and submerged flora such as *Salvinia* sp., *Cabomba* sp., *Utricularia* sp., *Tonina* sp. A freshwater marsh with herbaceous vegetation develops on the lowlands of the Barra Seca Valley.

At the time of coring, the water column in the deepest part of Lagoa do Macuco was ca. 3 m and the altitude at the water surface was 1 m above sea level (a.s.l.).

3. Material and methods

A piston corer (Livingstone 1955) was used to collect a 204 cm sediment core (MAC-C) at the center of the freshwater lake Lagoa do Macuco. The core tube was opened longitudinally in two halves and samples collected at 2 cm intervals in the ^{14}C Laboratory (CENA/USP).

3.1. Sediments description and granulometry

The sediment core was described based on its colours and macroscopic characteristics. Particle-size determination was carried out in 71 samples, pre-treated with hydrogen peroxide (10%) for organic matter removal and ultrasonically dispersed before laser diffraction analysis in a Laser Particle Size Shimadzu Sald 3101. Results of particle-size distribution are presented according to the Wentworth (1922) scale.

3.2. Radiocarbon age

Sixteen samples were selected for ^{14}C dating. Samples were treated with 2% to 4% hydrochloric acid at 60º C during four hours, washed and dried at 50º C (Pessenda *et al.*, 2009; Buso Jr. *et al.* 2013). After combustion, the CO_2 was used for AMS dating. Ages are expressed in years before present (yr BP) and calibrated ages (cal yr BP, 2σ;). Radiocarbon ages were normalized to a δ^{13}C of -25‰ VPDB and reported as calibrated years (cal yr BP, 2σ) according to Reimer *et al.* (2004). For post-bomb sample the ^{14}C activity is provided as the percentage of modern carbon (pMC) and calibrated age in cal AD, according to the CALIBomb software (McCormac *et al.* 2004). Calibrated ages were used for the construction of an age-model in the Tilia software v 1.7.16 (Grimm 1992), using a B-spline approximation model, which is used for age interpolation of any undated sample.

3.3. Pollen analysis

For pollen analysis, 26 samples of 1 cm^3 were processed according to Colinvaux *et al.* (1999), with the addition of exotic *Lycopodium* spores to determine pollen and spores concentrations (grains/cm^3) and accumulation rates (grains cm^{-2} yr^{-1}) as per Davis & Deevey (1964). Percentages of pollen and spores were calculated based on the total pollen sum, which includes all pollen types and indeterminates.

For each sample, pollen and spores were counted until reaching a minimum of 300 arboreal pollen grains (mangrove included). The identification was based on published pollen morphology and on the pollen reference collection of the ^{14}C Laboratory (CENA/USP). Identified types were grouped into

mangrove trees, other trees/shrubs, aquatics, herbs, and terrestrial ferns. Indeterminate pollen includes unidentified and reworked grains. CONISS (Grimm 1987) was used to run a cluster analysis based on pollen and spore counts from all taxa.
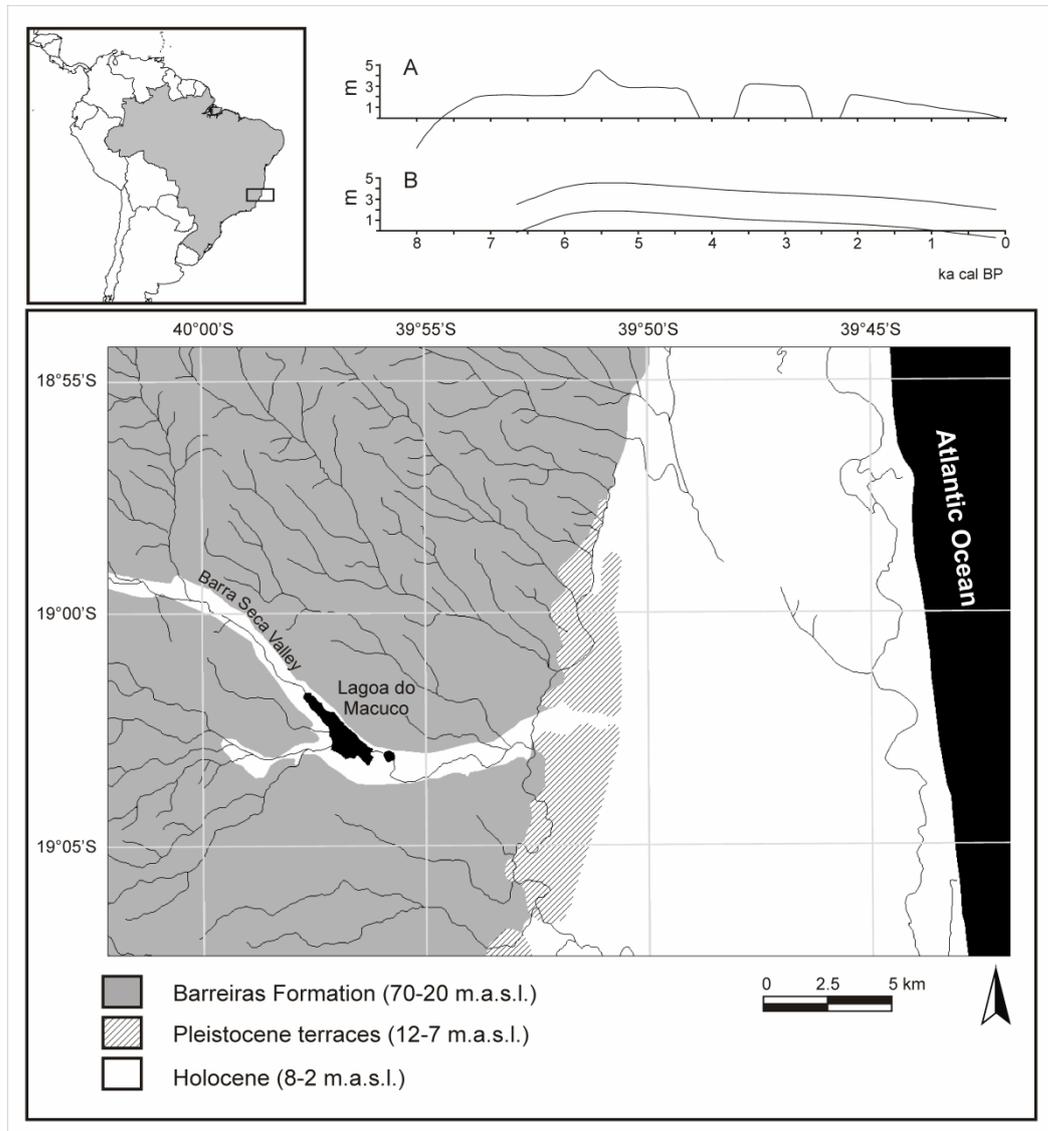

Figure 1: Study site. Map showing the location of the study site at the Brazilian coast (upper left panel), and in the geologic and elevation contexts (lower panel); relative sea-level curves for the southeastern Brazilian coast (upper right panel), in meter above present sea-level (m.a.s.l.), versus the time in thousands of calibrated years (ka cal BP); (A) subpanel from Martin *et al.* (2003); (B) subpanel from Angulo *et al.* (2006).

3.4. Phytolith sampling and extraction

The phytolith samples were taken in parallel to the pollen samples down to a depth of 194 cm. Phytoliths were extracted from twenty-three samples of 1 cm^3, but only fifteen of these samples contained phytholits. The average weight of dry sediment was 0,50 grams with 0,15 standard deviation. Extraction followed Madella *et al.* (1998) and Calegari et al (2013b), consisting in the removal of carbonates and iron oxide and/or aluminum by hydrolysis with strong acid (HCl - 5N) followed by oxidation with H_2O_2 (30%) for the removal of the SOM. The concentration and separation of phytoliths was done gravimetrically, using a solution of sodium polytungstate $(Na_6(H_2W_{12}O_{40})H_2O)$ with density of 2.35 g cm^{-3} (Madella *et al.* 1998, Osterrieth *et al.* 2009).

3.5. Classification and counting of phytoliths

Each microscopy slide was prepared with immersion oil and phytoliths were observed across three horizontal transects (Carnelli 2002): top, middle and bottom. Observations were carried out with a Condo microscope at 400x magnification. The identified morphotypes were named according to the *International Code for Phytolith Nomenclature* (Madella *et al.* 2005). Phytoliths were combined into five major groups according to their taxonomic origin: Poaceae (grasses) (Twiss *et al.* 1969, Tieszen *et al.* 1979, Twiss 1992), Arecaceae (palms) (Kondo & Iwasa 1981, Piperno 2006, Alexander *et al.* 1997b, 1999, Barboni *et al.* 1999), Cyperaceae (sedges), Maranthaceae (Piperno, 2006) and *Eudicotiledoneae* (mostly woody dicots) (Runge 1999).

From the assemblages, three palaeoenvironmental phytolith indices were calculated:

- The Humidity-Aridity Index (Iph) (Diester-Haas *et al.* 1973, Alexandre *et al.* 1997b), which is based on the ratio of chloridoid versus chloridoid and panicoid phytoliths. High Iph values suggest open woodlands and/or grasslands dominated by xerophytic Chloridoideae, indicating dry edaphic and/or climatic conditions. Low Iph values indicate the predominance of mesophytic Panicoideae, suggesting more humid conditions.

- The Climatic Index (Ic) (Twiss 1987, 1992) is the ratio of pooid versus the sum of pooid, chloridoid and panicoid morphotypes. High values indicate the predominance of C3 Pooideae grasses, suggesting cold climatic conditions.

- The Tree Cover Density Index (D/P) (Alexandre *et al.* 1997b, 1999, Barboni *et al.* 1999, 2007) consists of the D/P ratio, where D is the number of dicotyledon phytoliths (globular morphotypes) and P is the number of Poaceae phytoliths (pooids, chloridoids, panicoids, trichomes and bulliforms). High values indicate open vegetation, adapted to warm and dry climates, as in the African tropical and intertropical zones; lower values indicate forest vegetation with warm and wet climates

4. Results and discussion

4.1. Sediments description and granulometry

The MAC-C core consists of 5 sedimentary intervals. From 204 to 200 cm the sediment is dark green silt, abruptly changing to dark greenish-gray laminations of sand and silt, from 200 to 144 cm. From 144 to 78 cm the sediment gradually changes to a massive light gray mud, with predominance of silt and clay. In the zone between 78 to 70 cm the sediment grades to dark gray silt with transitional contacts. The uppermost 70 cm is a black gyttja with sand and silt.

4.2. Radiocarbon age estimation

The AMS ^{14}C dates range from 7667–7430 cal yr BP near the base of the core to cal AD 1958–1996 at the top (table 1). These dates can be considered reliable because:

- The date obtained at 198–200 cm is from terrestrial plant fragments;
- All samples were pre-treated for carbonate removal;
- Lake reservoir effect, hard water effect, or the assimilation of some other form of old carbon are probably negligible, considering the modern age obtained from sample 4–6 cm (which is the expected age for this depth);
- The dates are all in stratigraphic order and no inversion is evident.

4.3. Pollen record

Pollen and spores diagrams of percentages and accumulation rates for MAC-C core are shown in figure 2. Pollen and spores concentrations ranged from ca. 11,000 grains per cm^3 (76 cm) to more than 200,000 grains per cm^3 (68 cm). Three pollen zones have been defined from the cluster analysis.

Pollen Zone MAC-I from 7623 to 4396 cal yr BP (204 to 96 cm). Pollen accumulation rates are relatively low in this pollen zone (475-2316 grains. cm^{-2} /yr^{-1}). The interval is characterized by high percentages and accumulation rates of pollen grains from mangroves (23-53%) and other trees and shrubs (20-46%). In the first part of this pollen zone (from 204 to 150 cm) the input of herbaceous (6-9%) and aquatic (0-1%) pollen grains and spores (3-7%) is very low. From 150 to 96 cm herbaceous (11-25%) and aquatic (0-2%) pollen grains and spores (12-23%) present higher percentages (figure 2).

Higher percentages of Cyatheaceae spores are found from 144 to 96 cm, which justifies the delimitation of the sub-zone MAC-Ib dated at 7085-4396 cal yr BP (see Buso Jr. *et al.* 2013b). In Brazil, Cyatheaceae is represented by tree ferns that usually colonize humid forests (Lorscheitter *et al.* 1999) and according to Marchant *et al.* (2002) these spores should be indicative of humid environmental conditions. This higher humidity could be a consequence of a higher water table connected to the estuarine movements. Although the vegetation dynamics inside Barra Seca Valley may have been primarily a response to relative sea-level changes, further investigations are necessary to clarify these dynamics connected to possible climatic fluctuations during the period from 7000 to 4300 cal yr BP (Buso Jr. *et al.* 2013b).

Pollen Zone MAC-II from 4396 to 1287 cal yr BP (96 to 72 cm). Pollen accumulation rates (41-1247 grains cm^{-2} yr^{-1}) are lower than pollen zone MAC-I. During this interval mangrove pollen grains are rare or absent (0-8%). The majority of the retrieved pollen is represented by herbaceous (25-50%) and trees/shrubs pollen (30-70%), nonetheless aquatics (4-11%) and spores (4-21%) have significant frequencies.

Changes in the accumulation rates of mangrove pollen and associated proxies (C and N isotopes, chemical analyses, siliceous sponge spicules) in Macuco sequence have been related to fluctuations of Holocene relative sea-level of the Brazilian coast (Buso Jr. *et al.* 2013b). At the beginning of Pollen Zone MAC-I (ca. 7700 cal yr BP) arboreal and dense mangrove vegetation would have occupied the lower reaches of Barra Seca Valley. From ca. 7000 to 3200 cal yr BP (144 to 80 cm) the reduction of pollen accumulation rates (figure 2) should represent the displacement of the vegetation to the higher reaches of the valley due to the landward migration of the palaeo-estuary and the sea flooding of the lower parts of the valley (Martin *et al.* 2003, Angulo *et al.* 2006, Murray-Wallace 2007). This process is not reflected in the pollen diagram (figure 2A), however it is evident in the

carbon and nitrogen stable isotopes signatures as well

as the sponge spicules record (Buso Jr. *et al.* 2013a).

Table 1: ^{14}C results of the core MAC-C from Lagoa do Macuco

Laboratory #	Dated material	Depth (cm)	Age (BP)	Age (cal yr BP; 2σ)
UGAMS3405	Bulk sediment	4-6	101.39±0.33*	1,958-1,996**
UGAMS3406	Bulk sediment	20-22	100±30	253-0
UGAMS3407	Bulk sediment	48-50	110±30	253-0
UGAMS4265	Bulk sediment	64-66	290±25	440-154
TO13702	Bulk sediment	68-70	1,090±90	1,171-767
UGAMS3408	Bulk sediment	72-74	1,430±30	1,348-1,190
TO13483	Bulk sediment	76-78	2,440±70	2,717-2,208
UGAMS4266	Bulk sediment	80-82	3,260±25	3,479-3,361
UGAMS4267	Bulk sediment	92-94	3,820±25	4,238-3,992
TO13699	Bulk sediment	108-110	4,560±60	5,431-4,888
TO13700	Bulk sediment	122-124	4,580±120	5,571-4,858
UGAMS4268	Bulk sediment	126-128	5,460±30	6,293-6,024
UGAMS4269	Bulk sediment	134-136	5,780±30	6,634-6,414
TO13484	Bulk sediment	138-140	6,100±80	7,159-6,694
TO13701	Bulk sediment	170-172	6,470±70	7,458-7,172
TO13485	Plant fragments	198-200	6,730±80	7,667-7,430

UGAMS, AMS, University of Georgia, USA; TO, AMS, Isotrace Laboratory, Toronto, Canada
*^{14}C activity in pMC
** calibrated age in cal AD

From 3190 to 1287 cal yr BP (80 to 72 cm) the increase of the accumulation rates of pollen and spores and the high percentage of herbaceous pollen grains (figure 2) are interpreted as the colonization of the area by pioneer vegetation due to marine regression (Buso Jr. *et al.* 2013a, b). Although during this interval the pollen record of the core shows increasing concentration of Poaceae (23%), carbon isotopes analysis indicates C3 plants as the most important origin of the organic matter (Buso Jr. *et al.* 2013b).

Pollen Zone MAC-III, 1287 cal yr BP to the present (70 cm to the top of the core). This interval has the highest pollen accumulation rates of the entire deposit (978-37,064 grains cm^{-2} yr^{-1}). Mangrove pollen is not present in this pollen zone and herbs (31-69%) and trees/shrubs (25-43%) dominated the interval. Herbaceous pollen grains are mainly Poaceae (16-29%) at the beginning and then Cyperaceae (25-62%) in the uppermost samples. At the same time, pollen from aquatic plants increases reaching the value of 27% at the top of the core. The modern lake environment was established during this interval (Buso Jr. *et al.* 2013b).

4.4. Phytolith assemblage record

There is a great variability for phytoliths from the Lagoa do Macuco core. Many samples had very few or no phytoliths (e.g. samples between 194-144 cm, and 46 and 58 cm) while others show good concentrations (e.g. sample 72-74 cm or 76-78 cm), as illustrated in table 2.

Table 2: Absolut phytolith number counted in three lines to each sample and phytolith indices.

| Depth (cm)	Total phytolith counted in three lines	Phytolith indices		
		Iph	Ic*100	D/P
22-24	84	0	17	1
24-26	24	0	17	2
26-28	35	0	0	1
44-46	52	0	8	0
46-48	0	0	0	0
54-56	0	0	0	0
56-58	22	0	0	1
58-60	208	0	7	1
72-74	254	0	0	2
76-78	230	0	11	8
78-80	206	0	0	3
84-86	151	0	33	3
90-92	194	0	8	1
100-102	188	15	18	0
110-112	175	20	7	1
118-120	76	0	44	1
130-132	0	0	0	0
142-144	57	50	30	1
152-154	0	0	0	0
162-164	0	0	0	0
176-178	0	0	0	0
184-186	0	0	0	0
192-194	0	0	0	0
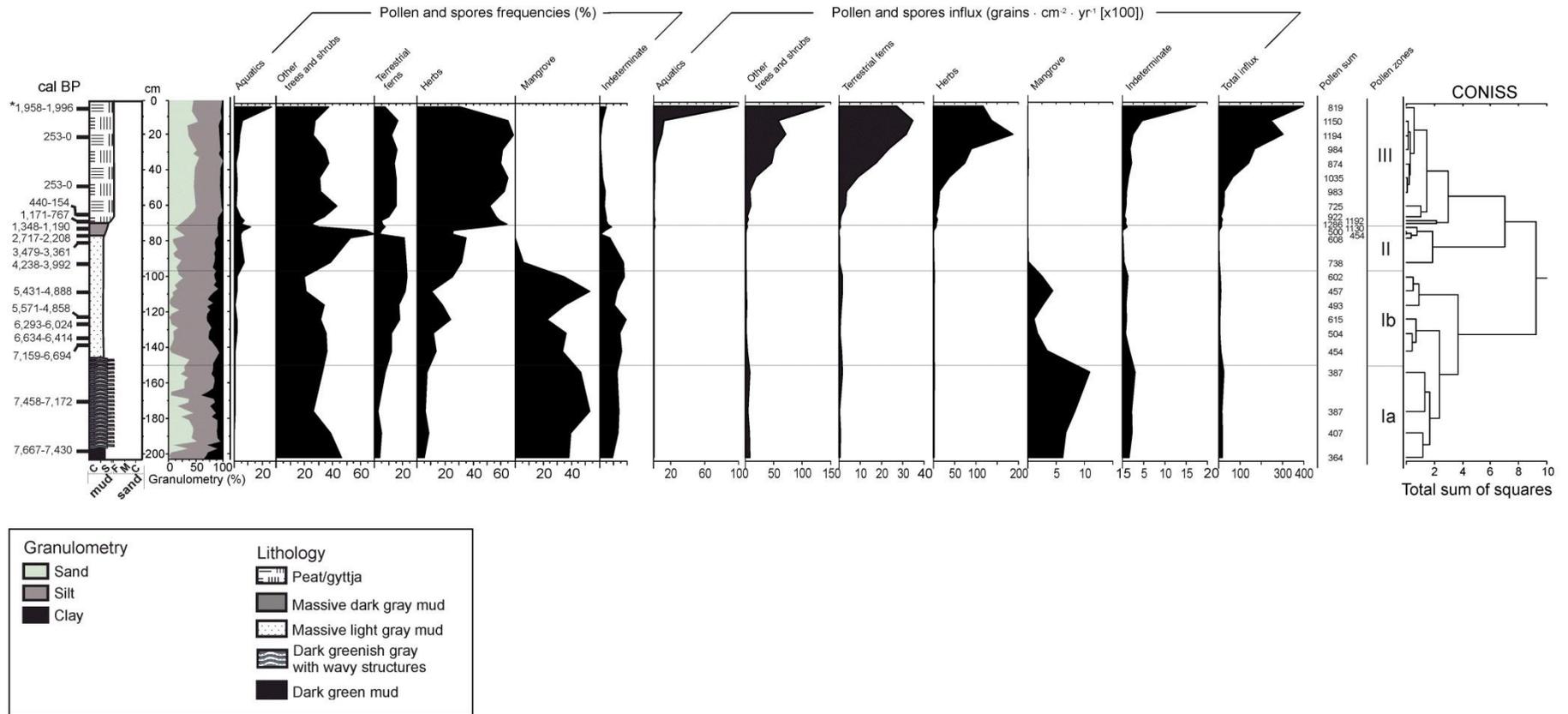
Figure 2: Calibrated ages, lithology; Pollen and spore percentage diagram, pollen and spore influx, cluster analysis, and pollen zones;
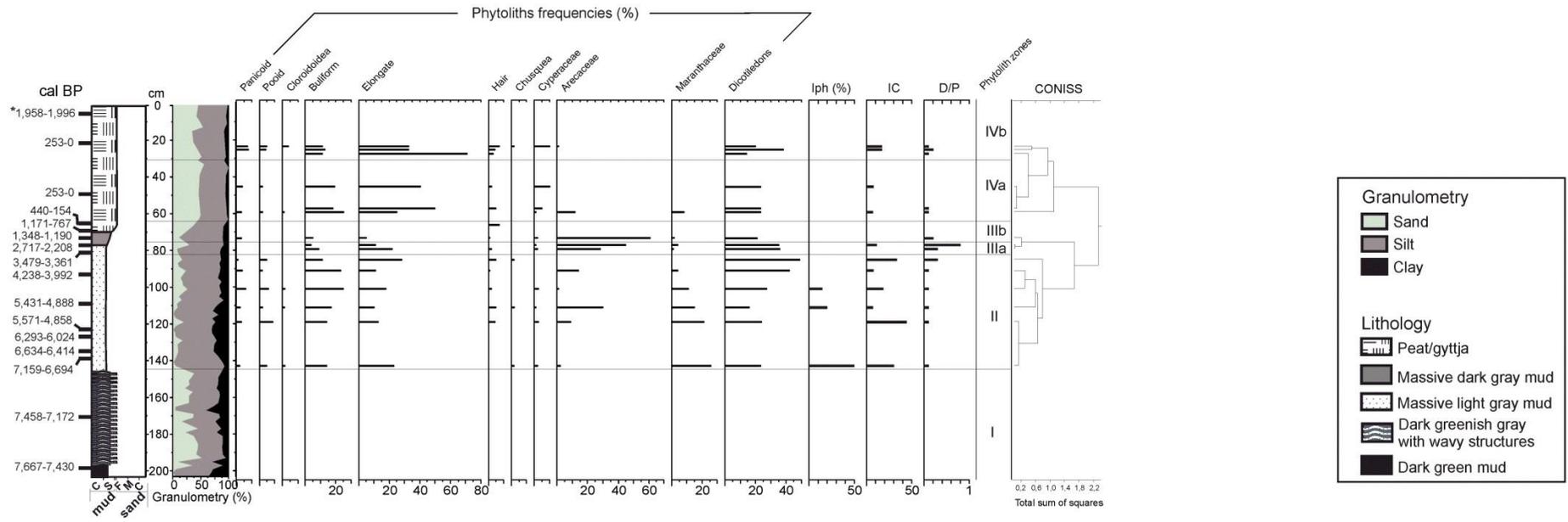
Figure 3: Calibrated ages, lithology; Phytolith percentage diagram, cluster analysis, phytolith indices and phytolith zones for the MAC-C core.

The phytolith morphotypes identified are in figure 4 and the relative frequencies (%) are given in figure 3. The analysis of phytoliths shows a predominance of Monocotyledons plants (between 51% and 86%), including Poaceae, Arecaceae and Cyperaceae. The bulliform (4% and 25%) and elongate (5% and 71%) forms from grasses are the most common morphotypes in all assemblages (figure 3)
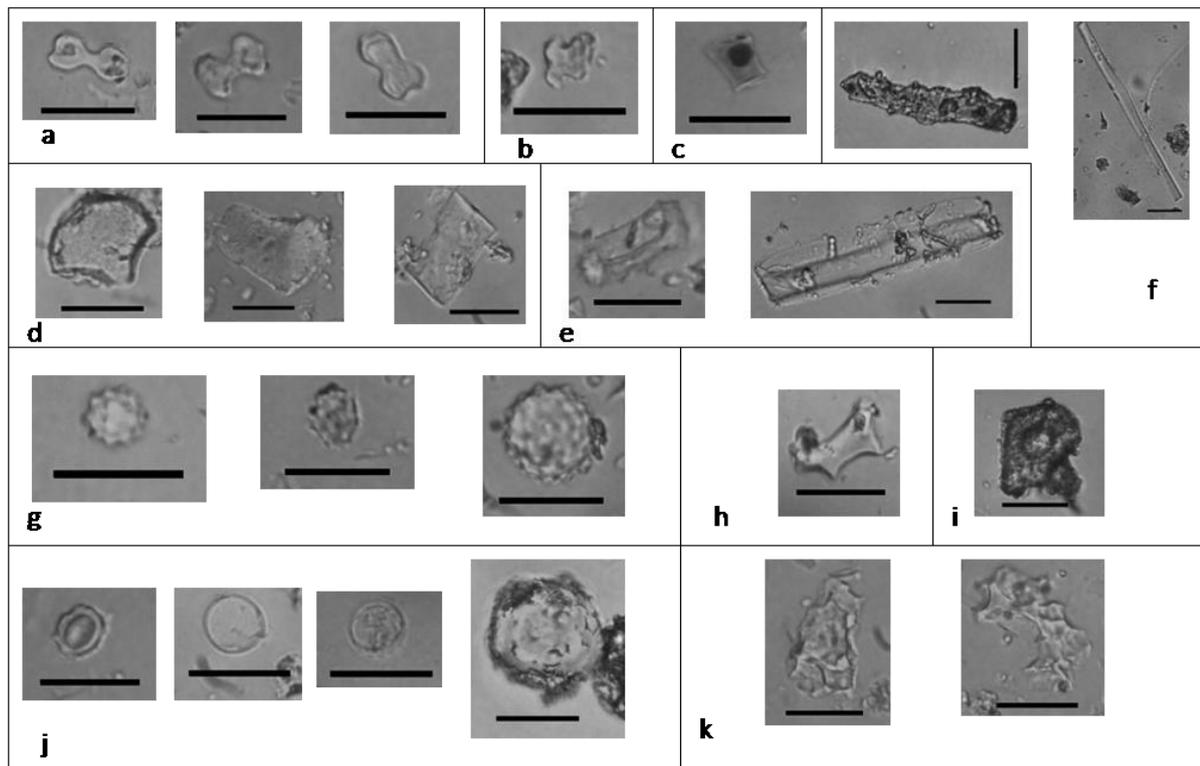

Figure 4: Microphotograph Phytoliths morphotypes: Poaceae: a) bilobates; b)cross; c) rondel; d) bulliform; e) trapeziform; f) elongate; Arecaceae: g) globular echinate; Chusquea: h) chusquea morphotype; Ciperaceae: i) ciperous morphotype; Eudicotiledoneae: j) globular ; Marantae: k)irregular faceted (magnification 40X). Bar=10µm

Panicoid short cells (C4) range between 3% and 8%, and while the highest frequencies of Pooid (C3) phytoliths are from the base of the core, between 100 and 120 cm of depth. Chloridoids (C4 and C3) short cells were observed only at the top of the sequence (22-24 cm), representing less than 4% of the assemblage (figure 3). Frequencies of *Chusquea* (Poaceae, Bambuseae) phytoliths are always below 2% and the morphotype is found only in samples at 22-24 cm, 84-86 cm and 110-112cm of dept. Cyperaceae phytoliths were between 1% and 10% of the total assemblages but their presence is observed only between 144-142, 120-58 and 46-22 cm (figure 3).

Tree phytoliths (Eudicotiledoneae) frequencies varies along the deposit, they increase between 103 and 63 cm to then diminish at 30 cm and increase again at 23 cm (table 1). The frequency of the Arecaceae phytoliths (globular echinate) is higher (28%-61%) between 58-120 cm of depth. Between 72 and 78 cm of depth, it was also observed an abrupt increase in the Arecaceae phytoliths.

The cluster analysis of the phytolith assemblages shows four phytolith zones (figure 3):

Phytolith Zone I, 7700-7100 cal yr BP (204-142 cm): No phytoliths were observed in these samples. The pollen analysis indicates a mangrove environment and because these species do not seem to have silica deposition, it could explain the absence of phytoliths.

Phytolith Zone II, 7100-3400 cal yr BP (142-82 cm): The phytolith assemblages are dominated by monocotiledoneae phytoliths (53-71%) with Maranthaceae phytoliths increasing with depth. According to phytolith indices this zone is a temperate interval, colder (IC 7% - 30%) but moister (Iph 50% - 15%) than Zones III and IV. These results corroborate the hypothesis of a more humid interval between 7000-4000 cal BP (Buso Jr. *et al.* 2013a) , in which the influence of both polar air masses and the summer monsoon system contributed to high humidity levels at Linhares during the whole year. Nonetheless, IC and Iph indices indicate that this humid interval may have extended only up to about 3400 cal yr BP.

The D/P index (between 0.1 and 0.3) shows a reduced tree cover during the interval between 7000 and 3400 cal. BP together with relatively higher values of Cyperaceae and Pooid (C3 - Poideae) that should represent more hydromorphic vegetation. The phytolith evidence seems to show a displacement of the arboreal vegetation to the more elevated margins of Barra Seca Valley due to the higher relative sea level, as evidenced by the pollen dataset (Buso Jr. *et al.* 2013b).

Phytolith Zone III, 3400-400 cal yr BP (82-64 cm) is subdivided into two:

Sub-zone IIIa: The samples between 82 and 76 cm have phytolith assemblages dominated by trees (Eudicotiledoneae) and tree palms (Arecaceae). The

bulliform and elongate morphotypes have low frequencies; however, the constant presence of bulliform can be interpreted as the presence of moisture without water deficit.

Sub-zone IIIb: The samples obtained between 76 cm and 60 cm have phytolith assemblages dominated by trees (Eudicotiledoneae) and palms (Arecaceae), with few other Monocotiledoneae. The phytolith indices highlight a period with the highest presence of arboreal vegetation probably adapted to little temperature variability and with moister conditions Palms constitute part of the arboreal vegetation, similarly to what observed today in the tropical rainforest. The massive dark gray mud sediment and the higher sedimentation rate (see Buso Jr. 2010) also suggest that the vegetation was denser than in the other phytolith zones.

Phytolith Zone IV, 400 cal yr BP to modern (64-00 cm) is subdivided into two:

Sub-zone IVa (from 440 Cal yrs B.P to ca. 253 cal yr BP): There is a decrease in the Eudicotiledoneae (tree) phytoliths frequencies (5-15%) while the Arecaceae morphotypes are absent. This data confirms the palynological evidence of stabilization of the forest in the surrounding of the lake and the formation of an herbaceous layer composed by Panicoid high grasses and Cyperaceae (sedges).

Sub-zone IVb (from ca. 253 cal yr BP to modern): The overall tree cover seems to be slightly reducing (D/P value close to 0.2) but there is an increase in specific taxa such as *Chusquea* (2%) and Cyperaceae (10%), in comparison with the subjacent sub-zone. In this zone Poaceae are more frequent, with an increase in C4 grasses short cells (Panicoid and Chloridoid) and a slight decrease in bulliforms. These data seems to disagree with the $\delta^{13}C$ values, which indicate that the organic matter was derived mainly from C3 plants (Buso Jr. *et al.* 2013a) but it might be due to the diverse input basin for the two different proxies (more local for the phytoliths and more regional for the pollen). The phytolith indexes indicate drier and warmer local environmental conditions (Iph) and arboreal vegetation (D/P).

5. Conclusions

The analysis of phytoliths was a good tool to complement the pollen data in order to provide additional information, specifically on issues of moisture and temperature. The two plant proxies tend to be in good agreement, showing the dynamics of the arboreal and non-arboreal vegetation in respect to the Mid-Holocene humid period and the changes in the sea level.

The phytolith dataset contributed with details the changes in the local vegetation structure, also highlighting the pulses in the C3 and C4 grass composition.

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