

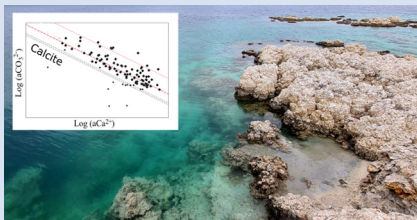
The chemical conditions necessary for the formation of microbialites

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Abstract



Whether biological and/or physico-chemical variations may have changed the abundance of microbialites over geological times remains debated. This calls for a better appraisal of the conditions necessary for the formation of modern microbialites. Here, we compiled datasets from 140 aqueous environments where modern microbialites have been reported, including their respective physico-chemical parameters. By analysing this database, we find that solutions where microbialites form have a relatively high saturation with calcite, the chemical activities of Ca^{2+} and CO_3^{2-} being controlled by the solubility of amorphous calcium carbonate (ACC) or monohydrocalcite (MHC) in most of these localities. Further statistical analyses of two databases listing ~ 8000 continental aqueous systems show that only a few of them display saturation with ACC/MHC, appearing as candidates for the detection of overlooked microbialites.

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Introduction

Microbialites are organosedimentary deposits formed by benthic microbial communities that mediate authigenic mineral precipitation (Burne and Moore, 1987). They are found throughout the geological record up to 3.43 billion years ago and are considered as among the oldest traces of life on Earth (Allwood *et al.*, 2007). It has been argued that the diversity of microbialites has varied over geological time with an overall decline at the end of the Proterozoic (*e.g.*, Awramik, 1971). The causes of these fluctuations have fed debates opposing two major models: (i) one involving biotic causes suggests that grazing by Metazoans induced the decline of microbialites (Walter and Heys, 1985), and (ii) a second “abiotic” model proposes that changes in the chemical composition of the ocean were responsible for microbialite decline (Fischer, 1965; Kempe and Kaźmierczak, 1994; Peters *et al.*, 2017).

While this debate is difficult to directly tackle, it emphasises that we still do not understand the conditions necessary for microbialites to form. Modern microbialites have been described in diverse environments (*e.g.*, marine, freshwater, hypersaline) and in the presence of very diverse microbial communities (*e.g.*, Iniesto *et al.*, 2021). Presently, there are many reports in the literature characterising the aqueous geochemistry of single sites where modern microbialites form. However, only a few meta-analyses gathering some of these data provide a broader, statistical overview (*e.g.*, Zeyen *et al.*, 2021). Here, we achieve an unprecedented compilation of datasets from 140 locations where modern microbialites form, spanning freshwater, brackish, saline and hypersaline environments (Table S-1).

We analyse the variability of the chemical parameters of microbialite-hosting environments and look for possible invariants. Moreover, in order to assess the rareness/commonness of conditions encountered in microbialite-hosting environments, we compare our database with two databases of continental aqueous systems.

Results

Physico-chemical parameters of microbialite-hosting environments. The compilation was achieved by systematically searching the terms “stromatolite”, “thrombolite” or “microbialite” in the literature (see Supplementary Information). The 140 compiled modern microbialite-hosting systems occur on all continents (Fig. S-1) in a diversity of climates and geological contexts. Most of the environments were freshwater, but 34 were marine. The database includes emblematic microbialites, such as those of Shark Bay, Lagoa Vermelha or the Bahamas, that have received much attention as modern analogues of ancient microbialites. Dissolved Ca^{2+} and Mg^{2+} concentrations were measured in a majority ($\sim 90\%$) of the corresponding aqueous solutions and ranged from 0.001 to 643 mM and 0.001 to 1325 mM, respectively. Concentrations of other major chemical species (Na^+ , Cl^- , K^+ , SO_4^{2-}) were also generally well documented ($\sim 88\%$ of systems on average). Measurements of the dissolved inorganic carbon (DIC) concentrations ranged between 0.022 and 6236 mM. These measurements were less documented ($\sim 76\%$ of systems) in the database, despite their importance in carbonate-rich environments. In seven lakes, alkalinity values

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were available and assumed to be equal to DIC (Dickson *et al.*, 1981; Fig. S-2).

Overall, microbialite-forming waters in our database span a high diversity of water chemical types as defined by Boros and Kolpakova (2018), which were: saline (53 % of the occurrences), soda-saline (23 % of the occurrences) and soda (24 % of occurrences) (Fig. S-3). Principal component analyses (PCA) were conducted on 10 chemical variables measured on 545 samples of microbialite-hosting environments in order to find the variables contributing to most of the dataset variability. It showed that most of the variance (~62 %) in the dataset was explained by (i) salinity (logarithms of Na, Cl, K activities) (35.22 %), and (ii) the logarithm of CO_3^{2-} activity anticorrelated with the logarithm of Ca^{2+} activity (26.32 %) (Fig. S-4).

Assessment of the saturation index of waters in which modern microbialites form. The anticorrelation between Ca^{2+} and CO_3^{2-} activities was further analysed by plotting their logarithms against each other. This plot also allows us to assess the saturation index (defined as $\text{SI} = \log(\text{IAP}/K_s)$, where IAP is the ion activity product and K_s is the solubility constant) of solutions with various CaCO_3 phases such as anhydrous crystalline phases and amorphous phases (ACC). Four hundred and sixteen chemical measurements performed on 102 microbialite-forming environments that were available in the database. The mean value was considered for the environments for which several measurements were available (Fig. 1). Conclusions were similar when using the median.

A large majority (89 %) of aqueous environments were highly supersaturated with respect to calcite and aragonite and aligned between the solubility lines of vaterite and an ACC phase (ACC2) as determined by Brečević and Nielsen (1989) (Fig. 1).

Diverse ACC phases exist with $\log K_s$ varying between -7.63 and -6.04 . Here, many points align close to the solubility lines of MHC and ACC1 phase (another ACC phase as determined by Kellermeier *et al.*, 2014).

A few environments ($n = 5$) appeared significantly undersaturated with calcite. However, three of them harboured microbialites that were siliceous (*e.g.*, Great Obsidian Pool, Mound Spring; Table S-1) or formed by pure trapping and binding of clays (*e.g.*, Lake Untersee, Antarctica; Table S-1). In two others (Kauai caves, Hawaii), microbialites formed on cave walls in freshwater seeping out of basalts and may experience significant chemical variations by evaporation and CO_2 degassing (Léveillé *et al.*, 2007).

Finally, some environments hosting carbonate microbialites were on average supersaturated with anhydrous carbonates but undersaturated with ACC and vaterite ($n = 9$; Figs. 1, S-5). Only one single analysis was available for Ciocaia drill (Romania). However, several ones were available for the other locations. For Pavilion Lake, solution geochemistry varied over time, reaching saturation with ACC at certain periods (Fig. S-5). For Lake Kelly, none of the several available measurements were saturated with ACC but it was reported that microbialites may no longer actively form (Lim *et al.*, 2009). Last, at least four other environments showed spatial chemical heterogeneities. They were undersaturated with ACC on average but supersaturated with ACC at certain locations. For example, Lakes Joyce and Hoare (Antarctica) have chemically stratified waters, and microbialites form at depths where DIC water content rises (Mackey *et al.*, 2018). Pastos Grandes (Bolivia) harbours groundwater outflows chemically evolving by evaporation along their travel away from the source. Microbialites form when water reaches saturation with ACC (Muller *et al.*, 2022). Overall, 98 % of aqueous

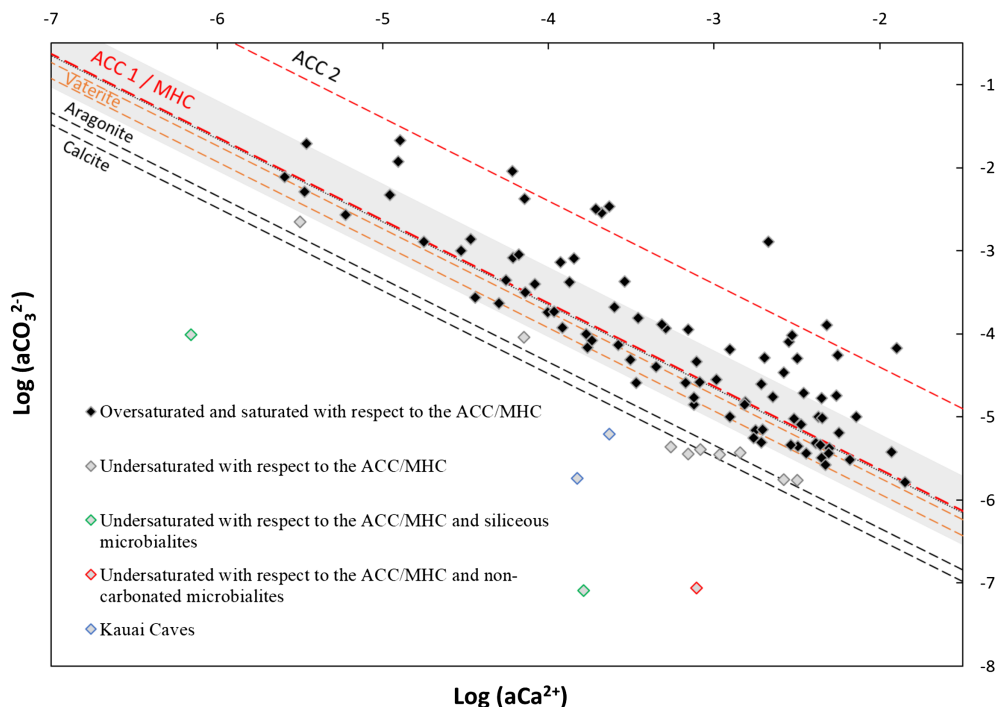


Figure 1 Plot of the log of the activities of CO_3^{2-} vs. Ca^{2+} for 102 microbialite-hosting environments. Solubility lines (dashed lines) are reported for calcite ($\log K_s = -8.48$), aragonite ($\log K_s = -8.34$), monohydrocalcite (MHC; $\log K_s = -7.6$), vaterite (De Visscher and Vanderdeelen, 2003), ACC1 as reported by Kellermeier *et al.* (2014; $\log K_s = -7.63$) and ACC2 as reported by Brečević and Nielsen (1989; $\log K_s = -6.39$). Only mean values are provided for each environment by a diamond. Green diamonds correspond to siliceous microbialites; red diamond corresponds to a clayey microbialite. Black diamonds correspond to environments saturated and oversaturated with ACC1 and/or vaterite; grey diamonds correspond to environments undersaturated with vaterite. The grey shaded zone highlights a 95 % confidence interval on the saturation values of microbialite-hosting environments with respect to ACC1/MHC.

environments hosting modern microbialites were saturated with ACC at least part of the time.

Comparison of the chemistry of modern microbialite-hosting aqueous solutions with two global freshwater databases. The chemical compositions of carbonate microbialite-hosting environments were compared with many aqueous environments listed in two general databases, in order to assess how unique the former might be. The Global Lake Ecological Observatory Network (GLEON) database compiles 105,678 samples from 6422 North European lakes with only temperature, pH, DIC and Ca^{2+} concentrations as physico-chemical parameters (Weyhenmeyer *et al.*, 2019; see *Supplementary Information*). The Environmental Data Initiative (EDI) groups 28,455 samples from 1547 North American lakes with several additional

parameters (*Supplementary Information*). Some of the lakes compiled in the EDI and GLEON databases possibly harbour microbialites but if this is the case, they have not been described in the literature and therefore are not in our microbialites database. The comparison showed that pH, DIC and calcium concentrations in the carbonate microbialite database were significantly higher on average than in lakes from the GLEON and EDI databases (Wilcoxon comparison tests of means; threshold of 0.05 and p values of 0, 2.1×10^{-210} and 3.8×10^{-175} , respectively) (Figs. 2a–c, 3 and S-6). A principal component analyses on the physico-chemical parameters from EDI and microbialite-hosting environments databases outlined that ~88 % of the variance of all these aqueous environments is explained by the activity of Cl^- or Ca^{2+} (70.2 % of the variability) and DIC (17.9 %) (Fig. 2d). The EDI lakes spread as

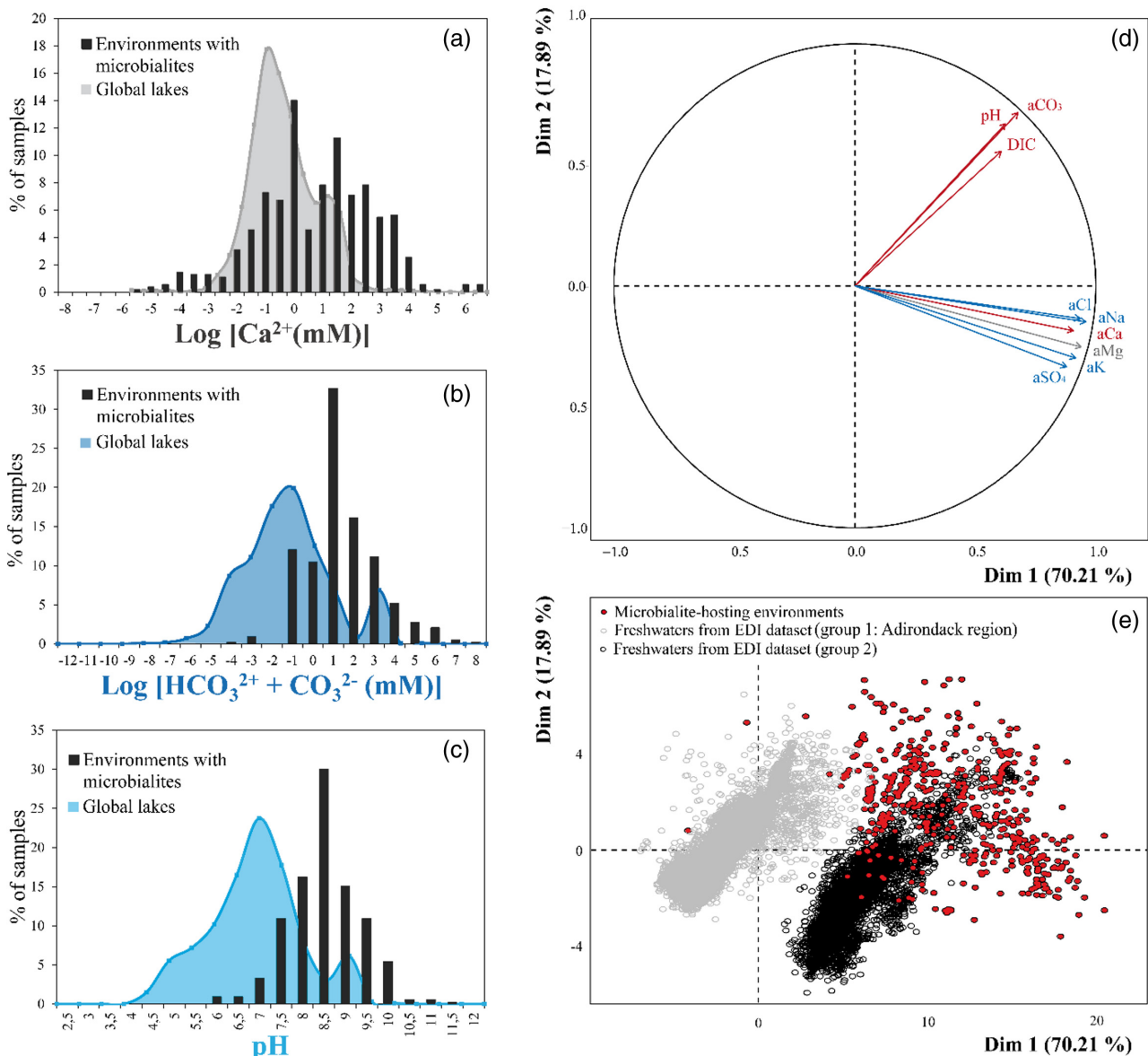


Figure 2 Comparison of the microbialite-hosting environments and lakes in the EDI and GLEON databases. Compared distributions are (a) the log $[\text{Ca}^{2+}(\text{mM})]$, (b) the log $[\text{HCO}_3^- + \text{CO}_3^{2-}(\text{mM})]$ and (c) the pH of the microbialite-hosting environments (black bars) and the EDI and GLEON databases (grey or blue surface). Ca^{2+} activities were calculated using all major ion concentrations in the EDI database, whereas they were approximated to concentrations in the GLEON database. Errors due to this approximation are estimated to be minor (Fig. S-7). (d) Correlation circle from the global PCA of the physico-chemical parameters of EDI lakes and microbialite solutions. The logarithms of the activities and DIC were used. The colours correspond to those used to differentiate the principal components on the microbialite-hosting environments PCA (Fig. S-4). (e) Plot of all aqueous environments hosting microbialites (red dots) and from the EDI database (black and grey dots) along the two main dimensions of the ACP.

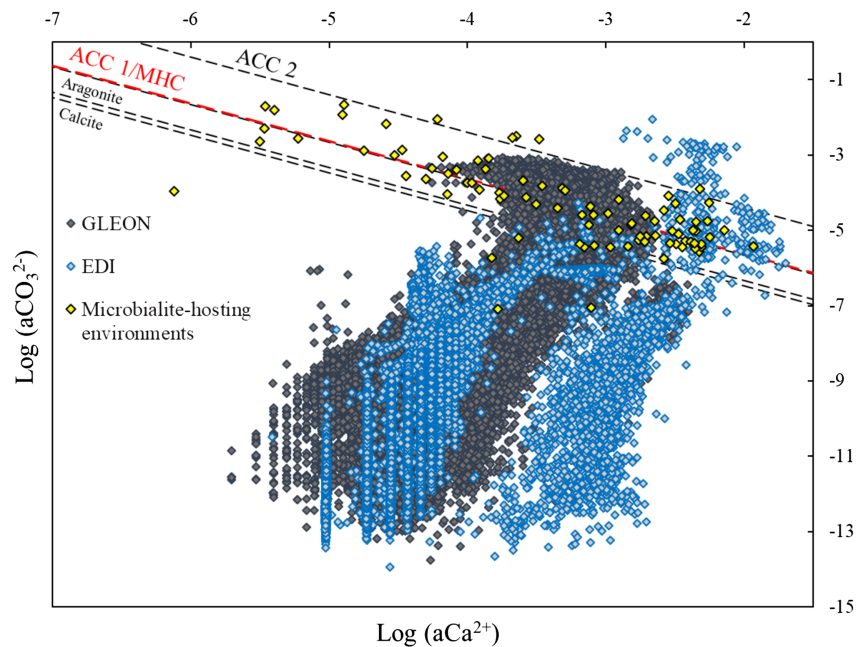


Figure 3 Plot of the activity of CO_3^{2-} vs. Ca^{2+} for all lakes from the EDI (blue diamonds), GLEON (grey diamonds) and microbialite-hosting environments (yellow diamonds) datasets. Solubility lines of aragonite, calcite, monohydrocalcite and ACC are the same as in Figure 1.

two groups in the plot of the two PCA dimensions. One group contains Adirondack region lakes (USA), including Mirror Lake, New York, whereas the second group is composed of all other more saline lakes. In this plot, microbialite-forming solutions mostly plot separately and stretch orthogonally to the freshwaters from the EDI database (Fig. 2e). GLEON data were not included in this PCA analysis, because too many parameters were missing.

Finally, by plotting the EDI and GLEON environments in the CaCO_3 solubility diagram, less than 5 % of them show saturation with ACC/MHC (Fig. 3).

Discussion

Saturation with vaterite/ACC/MHC as a necessary condition for the formation of microbialites. The need for a relatively high apparent critical saturation of the solutions so that microbialites form is suggested by the analysis of the compiled database. Arp *et al.* (2001) argued that a SI of ~ 1 relative to calcite was required for biofilm calcification to occur, based on the study of modern non-marine calcifying cyanobacterial biofilms. Such a value approximately corresponds to the solubilities of vaterite, ACC1 or MHC. Moreover, Fukushi *et al.* (2020) observed that several alkaline lakes were saturated with MHC and suggested this phase as a precursor of anhydrous carbonates. Accordingly, many microbialite-hosting environments in our database are alkaline. More recently, the same observation was done by Zeyen *et al.* (2021) in microbialite-hosting lakes in Mexico, who suggested ACC as an alternative precursor. In order to explain these observations, they proposed that ACC may precipitate first, before transforming to the less soluble MHC, then anhydrous carbonate phases, including vaterite. Here, based on a new database of 140 microbialite-bearing systems, encompassing freshwater, saline and hypersaline conditions, we generalise this observation to all environments where microbialites form, suggesting that saturation with ACC1/MHC or with vaterite appears as a necessary condition for the formation of modern microbialites. This model may be further adjusted in the future.

The ACC1 solubility line reported in Figure 1 is a lower bound for ACC. Indeed, the solubility of ACC increases with the Mg content (Mergelsberg *et al.*, 2020). According to Blue and Dove (2015), the partition coefficient K_d of Mg vs. Ca in ACC is constant (0.047) for pH below 9.5. Based on this, the Mg/Ca ratio in ACC possibly formed in microbialite-bearing systems ranges between 0.012 and 225, which may correspond to a variation in solubility by ~ 1 log unit.

Several conditions may allow the achievement of relatively high SI values with CaCO_3 as observed in microbialite-hosting environments. Aqueous alkaline environments are fed by fluxes of Ca^{2+} and HCO_3^- ions, *e.g.*, from water-rock interactions, and/or atmospheric inputs (Pulido-Villena *et al.*, 2006). This generally does not directly create high saturation conditions, except when locally, *e.g.*, Ca^{2+} -rich groundwater outflows in high DIC lakes such as in Lake Van (Shapley *et al.*, 2005). In many cases, evaporation is a major driver for concentrating solutions and achieving high saturation with CaCO_3 (Pecoraino *et al.*, 2015). This is consistent with the abundance of microbialites in saline environments. The SI values of aqueous solutions are not controlled by the precipitation of crystalline anhydrous CaCO_3 , which are the least soluble CaCO_3 phases. Instead, SI values are higher. Recently, Pietzsch *et al.* (2022), suggested that in some alkaline lakes, orthophosphate concentrations are high and inhibit crystalline anhydrous CaCO_3 precipitation, allowing the onset of saturation levels as high as the solubility of ACC. Unfortunately, this cannot be further tested here since dissolved PO_4^{2-} concentrations are only reported in eleven microbialite-forming environments of our database. Similarly, the role of exopolymeric substances (EPS) in locally controlling SI values could be questioned. Whether this inhibition may be a major process in all reported microbialite-hosting environments will require further data acquisition. Once Ca^{2+} and CO_3^{2-} activities reach values high enough so that MHC/ACC solubility is attained, we hypothesise that any further input of Ca^{2+} and/or DIC to the system can contribute to the accumulation of carbonates, partly as microbialites, helping to control their overall abundance in the aqueous system.

Another outcome of the present analyses is that they may help in finding localities where modern microbialites have been potentially overlooked so far, by screening aqueous environments based on their SI with CaCO_3 (Fig. 3). Some environments listed in the GLEON and EDI databases are saturated with ACC/MHC. However, we presently do not know if they host microbialites. Two options are possible: (i) all these environments host microbialites, which would imply that saturation with ACC/MHC is a necessary and sufficient condition for microbialite formation; and (ii) some of these environments do not contain microbialites, which means that some other conditions may be necessary in addition to saturation with ACC/MHC to form microbialites.

Presently, it may be speculated that changes in seawater chemistry, possibly at the end of the Proterozoic, from ACC-saturated to ACC-undersaturated seawater, may have caused the decline of microbialite abundances (Fig. S-8). However, to be validated, this speculation will need further constraints on the value of SI_{ACC} in past oceans.

Finally, a control of water chemistry by MHC/ACC precipitation as a necessary condition for the growth of microbialites does not mean that microbial communities or any biological parameter have no impact at all in the formation of microbialites. Instead, it suggests that in order for them to participate in authigenic microbialite formation, at least some specific physico-chemical environmental conditions, *i.e.* saturation with ACC/MHC, must be met.

Acknowledgements

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Additional Information

Supplementary Information accompanies this letter at <https://www.geochemicalperspectivesletters.org/article2311>.



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Supplementary Information

The Supplementary Information includes:

- Materials and Methods
- Tables S-1 and S-2
- Figures S-1 to S-8
- Supplementary Information References

Materials and Methods

Setup of a new modern microbialite-hosting environment database and description of the database. Modern microbialite localities were identified by searching the literature published between 1962 and 2022. For this purpose, the terms “microbialite”, “microbialite-forming mat”, “stromatolite” and “thrombolite” were used in search engines such as Web of Science or Google Scholar as well as the ScienceDirect, SpringerLink and ResearchGate platforms. Papers describing microbialites and containing chemical data on the aqueous environments where these microbialites form, were retrieved from papers listed in the database. Sedimentary structures referred to as “tufas” or “travertines” were not compiled in the database because of the significant differences sometimes attached to these terms (Pentecost and Viles, 1994). While there might be some subjectivity in choosing the term microbialites instead of another one, we did not add further subjective bias by reappraising ourselves the validity of the terms used in published studies.

The physicochemical parameters of the aqueous environments in which modern microbialites form were further searched when needed in additional publications not focusing on microbialites. Overall, we managed to compile 140 localities with 30 major variables into this database. The database is reported in Table S-1. Temperature, pH, conductivity, major ion and trace element concentrations, and some isotopic data (e.g., $\delta^{18}\text{O}$, $\delta^{34}\text{S}$, δD , $\delta^{13}\text{C}_{\text{org}}$, $\delta^{13}\text{C}_{\text{DIC}}$) were compiled when available. The type of environment (lacustrine, marine) as well as some details on the data collected from the literature and references were added. Moreover, the database contains the sampling dates and depth of microbialites, the major mineral phases composing them and the geological context. For aqueous environments of the GLEON database where both DIC and alkalinity were available, we checked that there was a 1:1 correlation between these parameters (Fig. S-2; Dickson, 1981). Then, for seven microbialite-hosting environments for which DIC was not available, DIC was equated to the alkalinity. Concentrations provided in mg L^{-1} or ppm were converted into mM in the database. For 81 localities, several water sample analyses were available and all of these data are reported, resulting in a total of 610 aqueous geochemistry analyses in the database.

Figure S-1 shows that modern microbialites can be found all over the world. The chemical types of all aqueous environments were determined according to the approach by Boros and Kolpakova (2018) by plotting them in Ca^{2+} – Na^+ + K^+ – Mg^{2+} and SO_4^{2-} – Cl^- – CO_3^{2-} + HCO_3^- ternary diagrams (Fig. S-3a). This shows that environments hosting modern microbialites are hydrochemically diverse and can be 'saline' for lakes with calcium and carbonate concentrations below 25 %, 'soda-saline' for lakes with Ca^{2+} and DIC contents >25 % and Na^{2+} as the dominant cation and carbonates not as the dominant anions, and 'soda' for lakes with Ca^{2+} and DIC contents >25 % and Na^{2+} and carbonates as the dominant cations and anions. In this classification, 53 % of microbialite-hosting environments are saline, 23 % soda-saline, and 24 % soda on a total of 95 environments. Microbialite-hosting aqueous solutions were also plotted in a ternary diagram defined as Mg^{2+} – SO_4^{2-} – HCO_3^- + CO_3^{2-} (Deocampo and Jones, 2014) (Fig. S-3b). Red arrows indicate chemical evolutions upon evaporation and precipitation of calcite and dolomite. This shows that many saline microbialite-hosting environments may result from advanced evolution of an initially high $\text{Ca}^{2+}/\text{CO}_3^{2-}$ solution. However, several saline environments may have had a low initial $\text{Ca}^{2+}/\text{HCO}_3^-$ and became enriched in sulfates. By contrast, for soda-saline and soda microbialite-hosting environments, most may result from the evolution of an initially low $\text{Ca}^{2+}/\text{CO}_3^{2-}$ solution and kept relatively sulfate poor.

Environmental Data Initiative (EDI) and Global Lake Ecological Observatory Network (GLEON) datasets.

The dataset of freshwater environments from the Global Ecological Observatory Network (GLEON, <https://gleon.org/>) was previously analysed by Weyhenmeyer *et al.* (2019). It groups 160,660 analyses of lake samples and reports six parameters: water depth, pH, temperature, Ca^{2+} , and dissolved CO_2 and DIC contents. Only data of lakes with available data for Ca^{2+} and DIC concentrations were extracted and used here, which represent 105,678 samples from 6,422 North European environments. Of those, at least 78 % had pH and/or temperature parameters.

The dataset from the Environmental Data Initiative (EDI, <https://portal.edirepository.org/nis/browseServlet?searchValue=limnology>) in the section “limnology” contains analyses of 29,176 samples from ~1600 environments, with 18 main variables: lake elevation, size, water depth, pH, temperature, conductivity, DIC, dissolved organic matter, dissolved oxygen, Ca^{2+} , Mg^{2+} , Si, Na^+ , Cl^- , K^+ , SO_4^{2-} , NO_3^- and NH_4^+ contents. Only 28,455 analyses from over 1,547 lakes from North America were selected as they included both Ca^{2+} and DIC contents. Interestingly, this database contained more parameters than the GLEON one, including the concentrations of several other major ions for over 40 % of the data. This allowed us to compare this database with the modern microbialite database on more parameters using PCA (Fig. 2 in the main text).

Speciation calculations. The activities of major ions were calculated with the PHREEQC software (Parkhurst and Appelo, 2013) based on the Hückel-Debye method, and taking into account the temperature in the activity calculation for the solubility products of CaCO_3 . The thermodynamic database “thermoddem.dat” (Blanc *et al.*, 2012) was used, with the solubility constants of calcite and aragonite from Plummer and Busenberg (1982), monohydrocalcite (MHC) from Hull and Turnbull (1973), and those of amorphous calcium carbonate (ACC) measured by Brečević and Nielsen (1989) at $K_s = 10^{-6.4}$ (*i.e.* ACC2) and Kellermeier *et al.* (2014) at $K_s = 10^{-7.63}$ (*i.e.* ACC1).

For the calculation of activities in the microbialite-hosting aqueous solutions, all available chemical species concentrations and temperature were considered. In the GLEON dataset only Ca^{2+} content, DIC content, temperature and pH were available to calculate the ion activities. The water chemistry analyses from all the databases were validated by verifying that the electric charge imbalance was between –10 % and +10 %.

Statistical analyses. The distributions of the major ion concentrations in the GLEON and EDI datasets *vs.* in the microbialite dataset are shown in Figure S-6. The differences between these distributions in the GLEON and EDI datasets *vs.* in the microbialite dataset were statistically tested. As these values are not normally distributed, the non-parametric Wilcoxon-Mann-Whitney test was chosen with a *p*-value threshold of 5 %.



Principal component analyses (PCA). Similar to what was done by Zeyen *et al.* (2021), we conducted PCA to assess which parameters explain most of the variability in the datasets. In these analyses, each axis (or principal component) is a linear combination of variables defined so that they maximise the spatial discrimination of samples; coordinates of vectors on each axis indicate the coefficient of the variables in the linear combination. The longer the orthogonal projection of an arrow on a given axis, the stronger the contribution of the variable to this axis. The smaller the angle between arrows, the stronger the positive linear correlation between corresponding variables. Orthogonal arrows indicate no correlation, while arrows pointing in opposite directions indicate a negative correlation. The samples are plotted in two dimensions using their projections onto the first two principal components. The part of the total sample variability explained by each axis is indicated in the axis label (Zeyen *et al.*, 2021). We compared EDI and microbialite-hosting environments. One PCA was performed on the microbialite-hosting environments dataset (Fig. S-4). A second PCA was performed on the combined EDI and microbialite-hosting environments datasets (Fig. 2d, e). For each PCA dataset, the parameters missing for more than half of the environments were removed. The environments where more than half of the parameters were missing were then removed. The missing values were further imputed based on the nearest neighboring environments. Imputation of missing values and PCA were performed using the R packages missMDA, FactoMineR and Factoshiny (Lê, Josse and Husson, 2008; Josse and Husson, 2016). The GLEON dataset was not used in this analysis because most of the water chemistry parameters describing the microbialite-hosting environments are missing.

Prediction of overlooked microbialite-hosting environments. The present analysis may also help finding localities where modern microbialites have been potentially overlooked so far, by screening aqueous environments based on their SI with CaCO_3 . Among the 7969 lakes in the GLEON and EDI databases, around 318 were on average saturated with ACC/MHC (Fig. 3; Table S-2) and therefore appear as potentially favorable for the development of microbialites. Looking back at the literature on some of these lakes, we found reports of (i) biogenic carbonate precipitation in Locknesjön lake (Sweden; Labuhn *et al.*, 2022), (ii) tufa precipitation in Sinijärv lake (Estonia; Laumets *et al.*, 2014), (iii) calcifying microbial mats in Deer lake (Canada; Zorz *et al.*, 2019), (iv) cyanobacterial calcareous mud in Storfjärden, Hällsjön and Käringsjön lakes (Sweden; Brunberg and Blomqvist, 1999), and (v) seasonal CaCO_3 production in Ripley lake (US; Garrison and Pillsbury, 2009). Only future systematic and careful campaigns will be able to determine whether modern microbialites may form in these localities or not. If all of these lakes eventually prove to host microbialites, this will further support the conclusions of the present study. If some do not host microbialites, it will be important to understand what other parameters (including biological parameters) may prevent microbialite formation in an otherwise chemically favorable environment.



Supplementary Tables

Table S-1 Modern microbialite-hosting environments database.

Table S-2 List of the names of the lakes in GLEON and EDI datasets that are saturated with ACC as shown in Figure 3. Only six cases of microbially induced carbonates have been found in the literature in some of these lakes. Microbialites may have been overlooked in these environments. Some lakes may become artificially ACC-saturated, and possibly not prone to microbialite formation, following anthropic alkalinisation using liming to fight acidification. This is particularly the case in more than 20,000 rivers and lakes in Sweden (Svenson *et al.*, 1995; dataset used by Weyhenmeyer *et al.*, 2019).

Tables S-1 and S-2 (.xlsx) are available for download from the online version of this article at <https://doi.org/10.7185/geochemlet.2311>.

Supplementary Figures

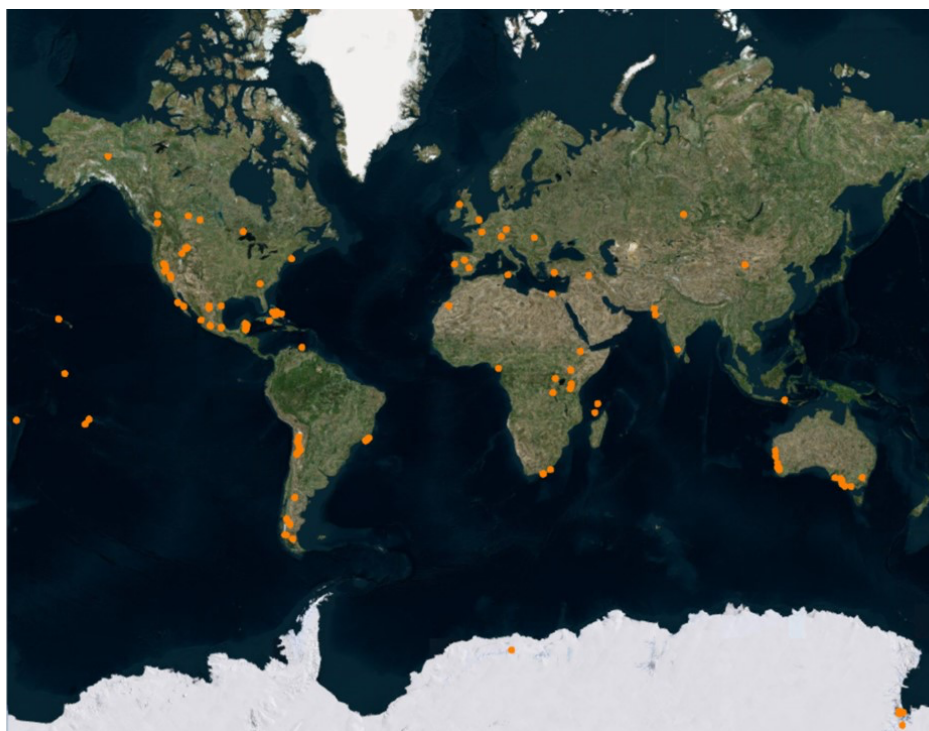


Figure S-1 All modern microbialite-hosting environments listed in the database represented on a world map (provided by Microsoft Excel).

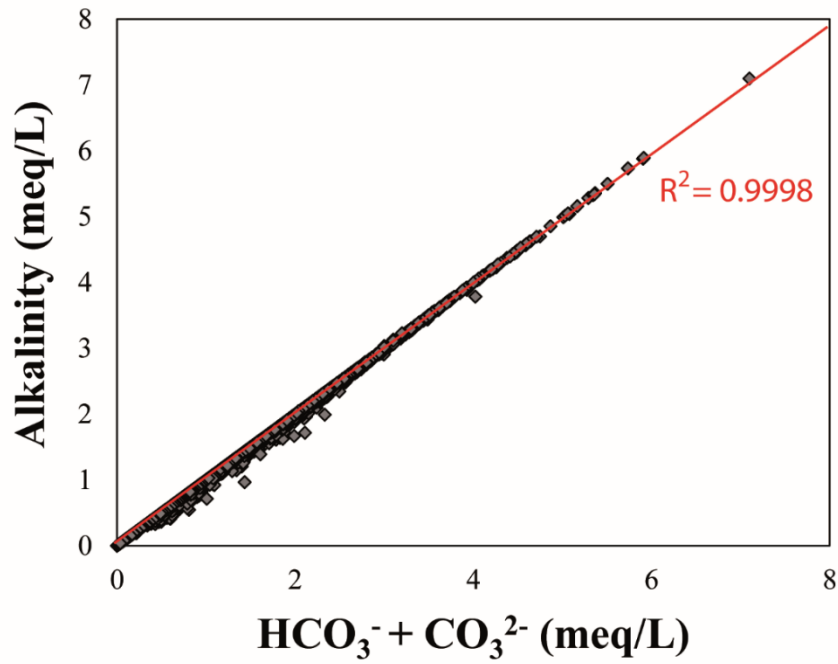


Figure S-2 Linear correlation between the alkalinity and the DIC in freshwater environments from the GLEON dataset. We note that the pH range encompassed by these lakes is such that H_2CO_3 and CO_2 are not important contributors to DIC.

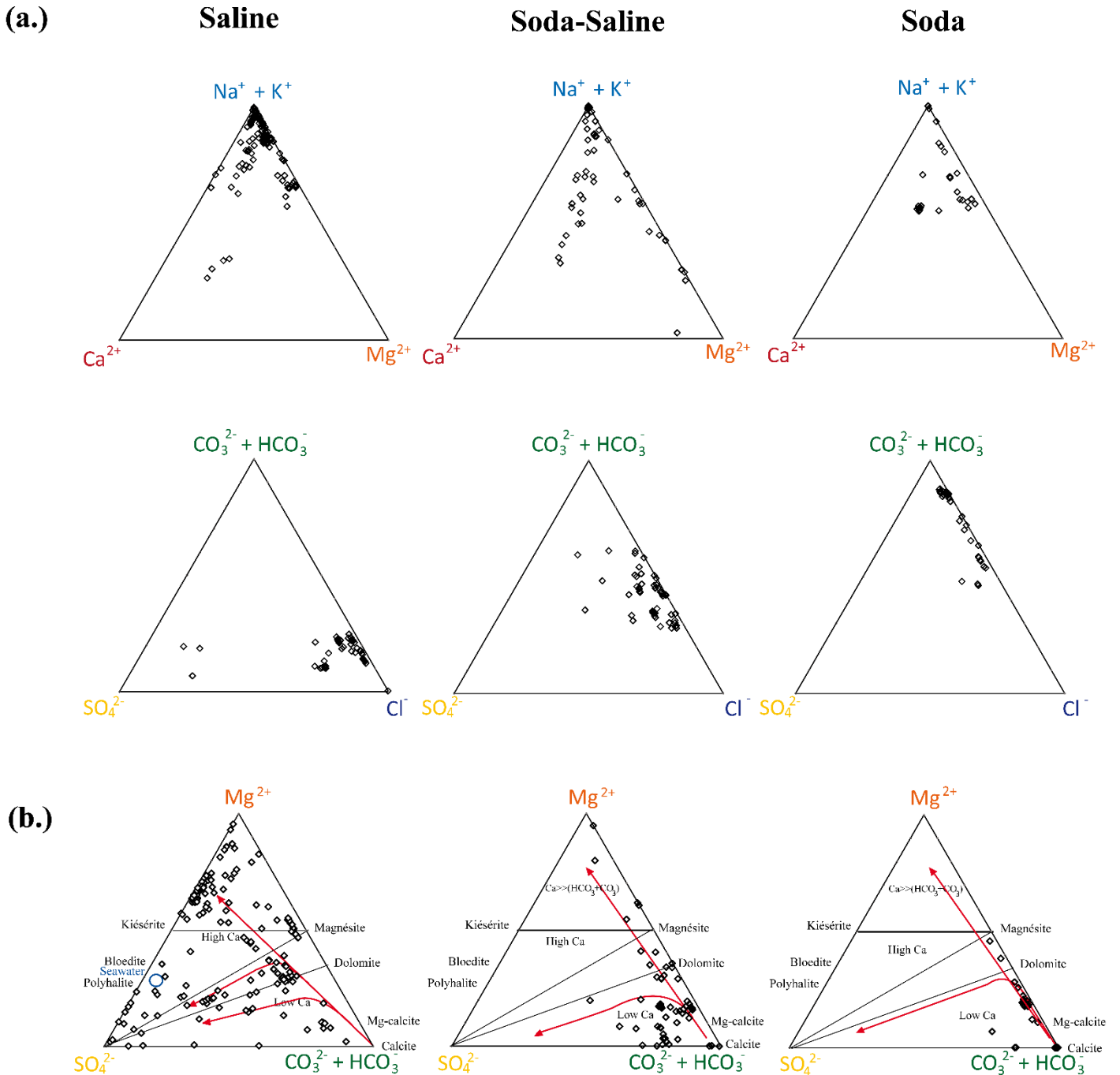


Figure S-3 Ternary diagrams of microbialite environments. **(a)** Chemical types are assessed based on the approach of Boros and Kolpakova (2018). **(b)** Deocampo and Jones' (2014) triangles that show chemical evolution of aqueous environments by evaporation and precipitation depending on their initial $\text{Ca}^{2+}/\text{HCO}_3^-$ ratio (red arrows) between modern microbialite-hosting environments.

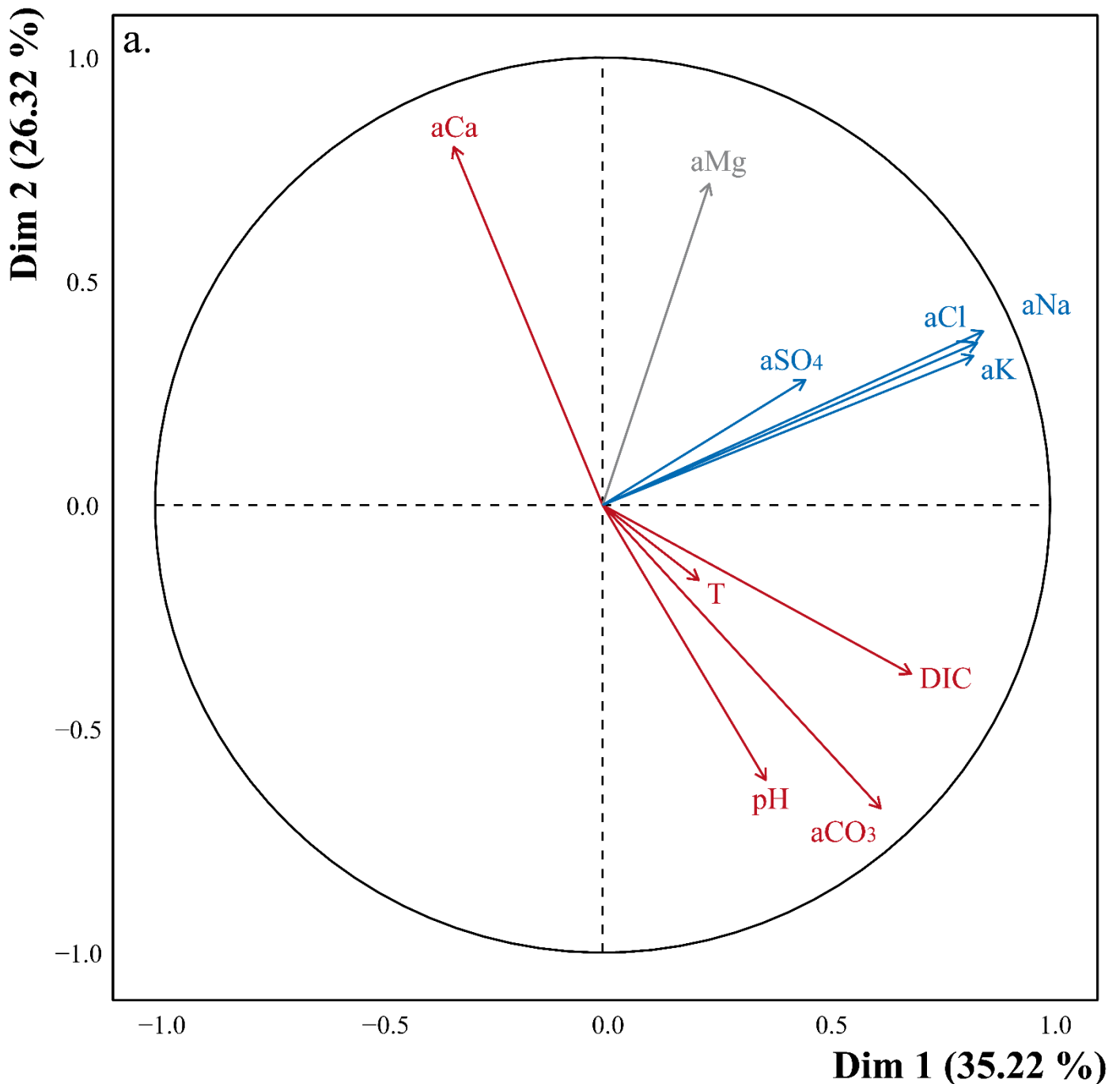


Figure S-4 Correlation circle from the PCA on 10 physicochemical parameters from 545 aqueous samples collected from microbialite-forming environments. Ten variables were available, which are: $\log(a\text{CO}_3)$, $\log(\text{DIC})$, pH, T ($^{\circ}\text{C}$), $\log(a\text{Ca})$, $\log(a\text{Mg})$, $\log(a\text{Na})$, $\log(a\text{Cl})$, $\log(a\text{K})$ and $\log(a\text{SO}_4)$. The variance of these environments is explained for almost 62 % by a dimension (blue arrows, explains ~35.22 % of variability) grouping the chemical species responsible for salinity as well as sulfates and by another dimension (red arrows, explains ~26.32 % of variability) characterising the carbonates contents, pH and temperature anticorrelated to the calcium and to a lesser extent magnesium contents.

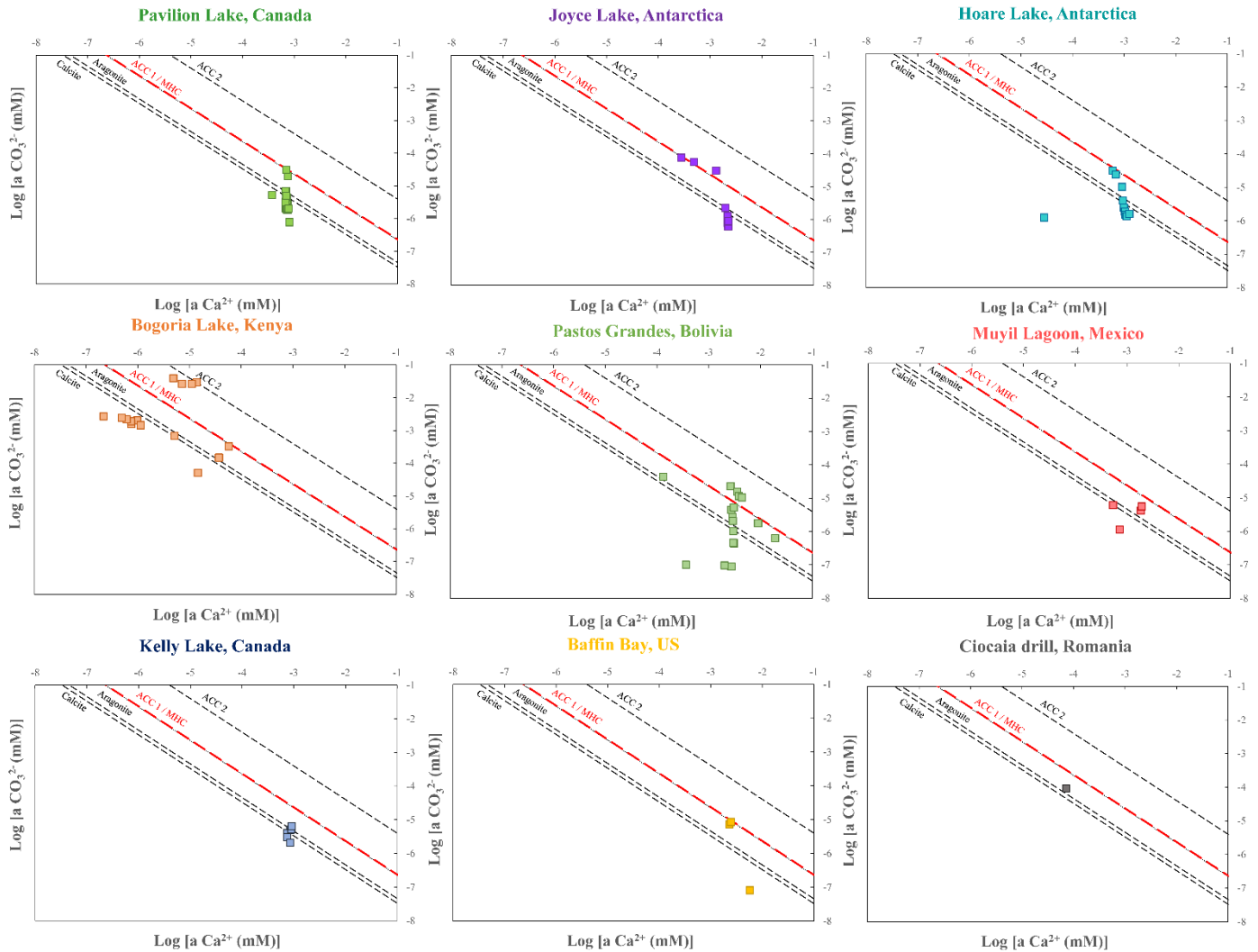


Figure S-5 Plots of CO_3^{2-} vs. Ca^{2+} activities for aqueous environments undersaturated with ACC on average. Different squares represent analyses at different times or different locations along the aqueous system. Table S-2 lists these environments and summarises some of their characteristics. It appears that 73 % of these environments reach saturation with ACC at least once in the time and/or place of sampling; it is assumed the variability in their chemistry makes them outliers.

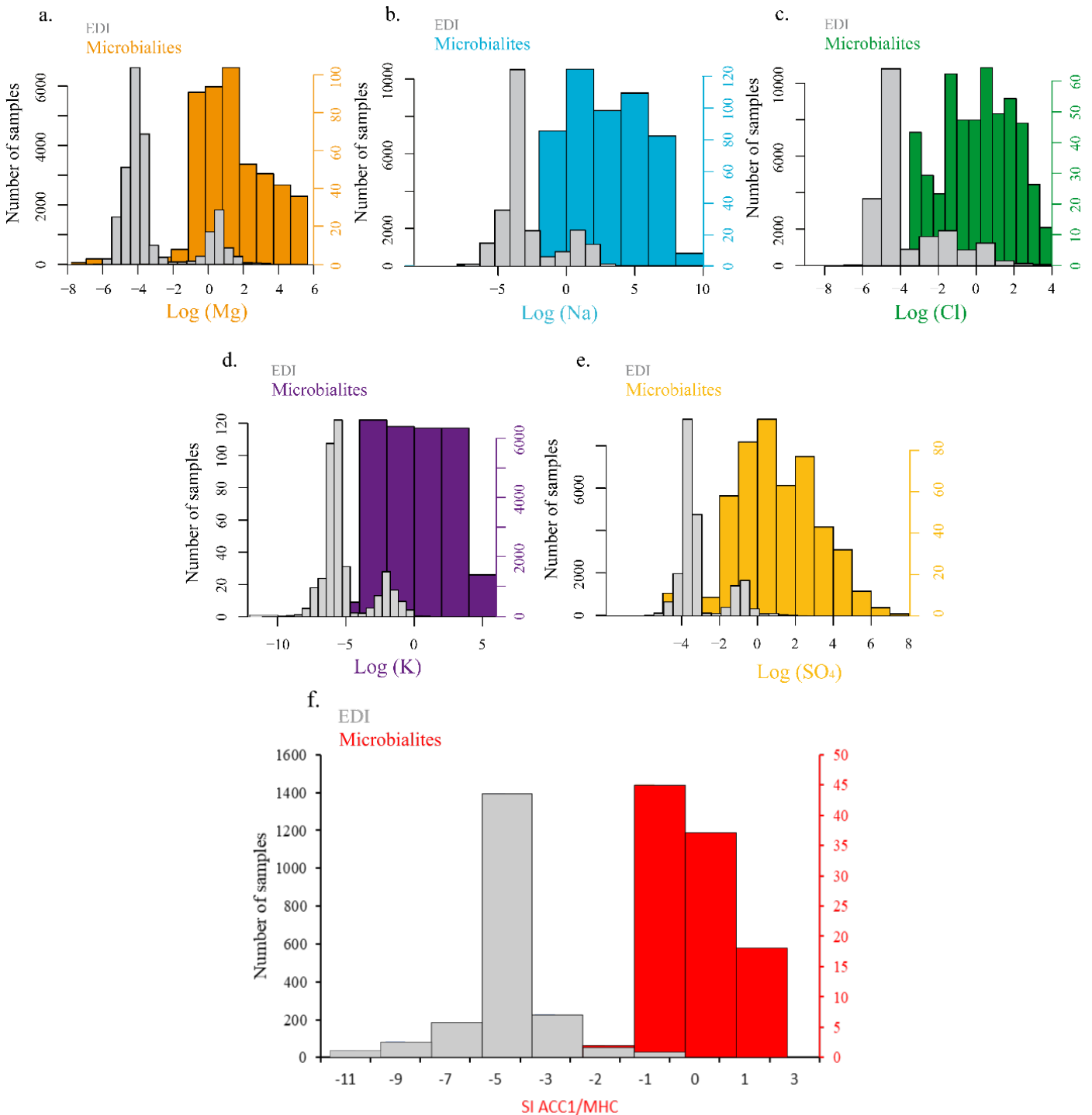


Figure S-6 Compared distributions of the logarithms of the main dissolved chemical species (and saturation index (SI) of ACC1/MHC) in North American (grey) vs. microbialite-hosting (coloured) lakes. **(a)** Magnesium logarithm, **(b)** sodium logarithm, **(c)** chloride logarithm, **(d)** potassium logarithm, **(e)** sulfates logarithm, **(f)** SI of ACC1/MHC.

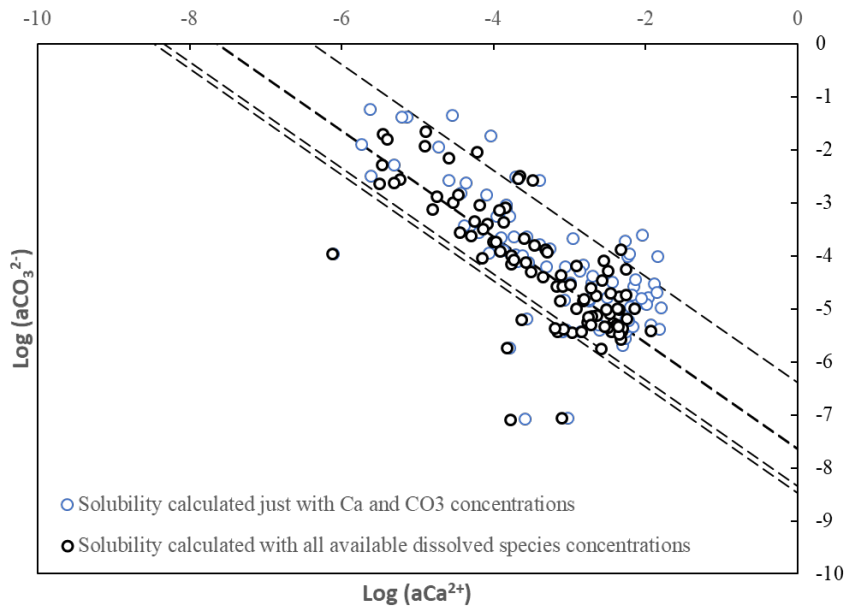


Figure S-7 Comparison between activities in the microbialite-hosting environments (i) calculated based on the knowledge of all the dissolved species concentrations available (open black points; “standard” approach) vs. (ii) approximated based on the knowledge of pH, Ca and DIC concentrations only (open blue circles; “approximated” approach). The differences between the approximated and standard calculated activities appear minor. The mean difference based on Euclidean distances between the values of the $\text{log}(\text{activities})$ calculated by the two approaches was 0.2.

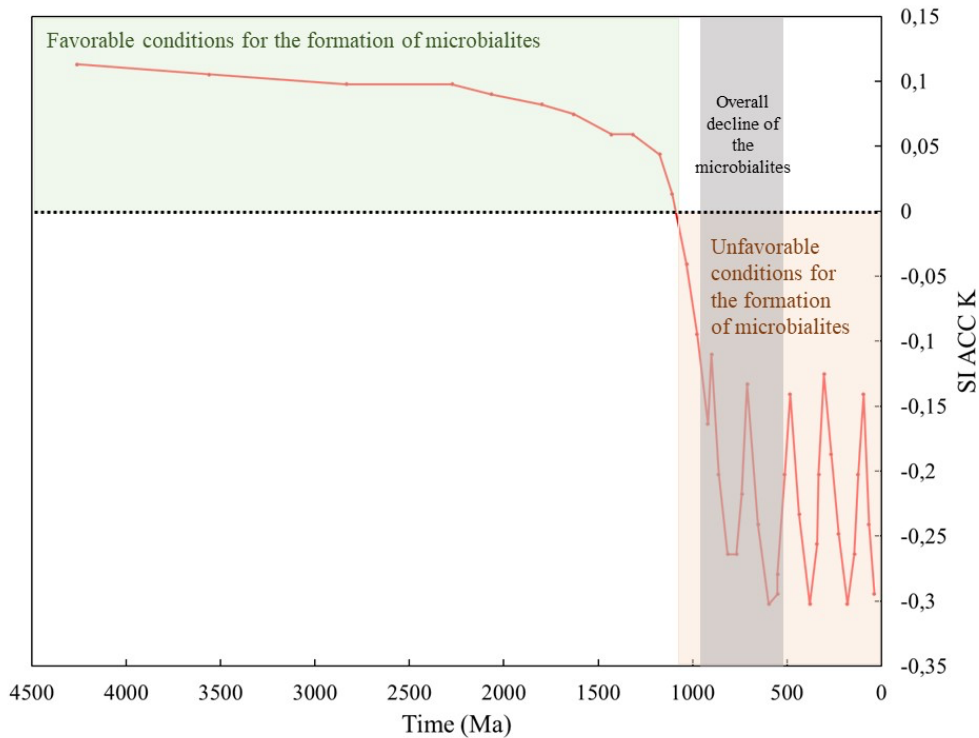


Figure S-8 Geological evolution of the saturation index (SI) of seawater with respect to MHC/ACC1 adapted from the model by Kempe and Kazmierczak (1994). The K_{s-ACC} was derived from Kellermeier *et al.* (2014; ACC1). The black dashed horizontal line indicates saturation with MHC/ACC1.



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