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The barium biogeochemical cycle in the Mediterranean: bacterial barite precipitation and implication for paleoproductivity reconstructions

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Pedes in terra ad sidera visus
Los pies en la tierra, la mirada en el cielo

A mis padres

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CONTENTS

Relación de acrónimos

AMOC - Atlantic Meridional Overturning Circulation

CFU - Colony-Forming Units

CM - Central Mediterranean

CMBA - Culture Medium enriched in Ba (Ba-enriched Marine Broth)

CTD - Conductivity, Temperature and Depth

DCM - Deep Clorophyll Maximum

DIC - Dissolved Inorganic Carbon

DNA- Deoxyribonucleic acid

DOC - Dissolved Organic Carbon

DOM - Dissolved Organic Matter

DW - Deep Water

EA - East Atlantic pattern

EA/WR - East Atlantic/West Russian pattern

EBSD - Electron Backscatter Diffraction

EDX - Energy-Dispersive X-ray microanalysis

EM - Eastern Mediterranean

EMDW - Eastern Mediterranean Deep Water

EMT - Eastern Mediterranean Transient

EPS - Extracellular Polymeric Substances

FWM - Far Western Mediterranean

HRSEM - High Resolution Scanning Electron Microscopy

ICP-MS - Inductively Coupled Plasma Mass Spectrometry

IW - Intermediate Water

LDPE - Low Density Polyethylene

LIW - Levantine Intermediate Water

LSR - Linear Sedimentation Rates

MARs - Mass Accumulation Rates

MAW - Modified Atlantic Water

MB - Marine Broth

MDW - Mediterranean Deep Waters

MIW - Mediterranean Intermediate Water

MO - Mediterranean Oscillation

MOW - Mediterranean Outflow Water

NAO - North Atlantic Oscillation

ODP - Ocean Drilling Program

OM - Organic Matter

OML - Oxygen Minimum Layer

ORLs - Organic-Rich Layers

POC - Particulate Organic Carbon

PCR - Polymerase Chain Reaction

rep-PCR - Repetitive element sequence based-polymerase chain reaction

rRNA - Ribosomal ribonucleic acid

RT - Residence Time

SGD - Submarine Groundwater Discharge

SW - Surface water

TOC - Total Organic Carbon

UV-Vis - Ultraviolet-Visible Spectroscopy

WM - Western Mediterranean

WMDW - Western Mediterranean Deep Water

XRD - X-ray Diffraction

Resumen

Esta tesis aborda el estudio del ciclo biogeoquímico del Ba en el Mediterráneo a partir del análisis de este elemento en el registro sedimentario de las diferentes cuencas mediterráneas, de su distribución en la columna de agua, así como la investigación de los mecanismos de precipitación de barita ($BaSO_4$) en medios marinos. El ciclo biogeoquímico del Ba está estrechamente relacionado con el ciclo del carbono dado que este elemento es un indicador de la productividad biológica marina y, por tanto, de la respuesta oceánica al cambio climático. Ello se debe a la precipitación abundante de barita en la columna de agua cuando existe una alta productividad biológica, lo que se traduce en una alta concentración de Ba en sedimentos. Aunque han sido muy diversos los indicadores tradicionalmente utilizados para reconstruir la productividad biológica en el pasado, la barita frente a otros paleoindicadores de productividad presenta la ventaja de que en medios lo suficientemente oxigenados y expuesta a procesos de alteración no es eliminada del registro sedimentario, por lo que este mineral se ha convertido en un indicador fiable y ampliamente usado en investigaciones paleoceanográficas. En el caso del Mar Mediterráneo, su carácter de cuenca semicerrada y circulación restringida han hecho que la respuesta a la variabilidad climática se haya visto amplificada, en particular la respuesta de la productividad biológica marina. Así, durante periodos de cambio climático hacia condiciones de mayor humedad y precipitación, el mayor aporte de nutrientes dio lugar a un incremento significativo de la productividad. Además, en muchos casos el mayor aporte de agua dulce también conllevó una estratificación de masas de agua y con ello la menor oxigenación de las aguas profundas, depositándose y preservándose sedimentos ricos en materia orgánica, comúnmente conocidos como sapropeles. En esta tesis se ha hecho un análisis detallado del contenido en Ba en los sapropeles depositados en las distintas cuencas mediterráneas para analizar su variabilidad espacial y temporal, así como un estudio de la barita preservada en dichos sapropeles. En cuanto a las diferencias entre las distintas cuencas mediterráneas, puede establecerse que en zonas más someras, una mayor tasa de sedimentación ha podido dar lugar a una dilución muy significativa del contenido en Ba. También se corrobora el gradiente de productividad en relación a la principal fuente de nutrientes

que fue el Nilo. En el Mediterráneo occidental se han depositado capas ricas en materia orgánica, pero con un contenido en materia orgánica y Ba inferior al de los sapropeles y en el Tirreno, por ejemplo, si bien se han depositado sapropeles, el contenido en Ba es sensiblemente menor que en las cuencas más orientales. La variabilidad temporal es aún más pronunciada. En general, los sapropeles pleistocenos tienen un contenido en Ba mayor que sus equivalentes más modernos, en concreto el sapropel más reciente (S1). El contenido en materia orgánica a lo largo del depósito de los distintos sapropeles también ha variado, siendo mayor en sapropeles más antiguos. Aunque guarda cierta correlación con el contenido en Ba, el contenido en materia orgánica responde también a las condiciones de preservación, siendo particularmente elevada en sapropeles del Plioceno debido al desarrollo de condiciones anóxicas. Por ende, a pesar de que el contenido en materia orgánica es mucho mayor en los sapropeles pliocenos, el contenido en Ba es comparable a los del Pleistoceno, indicando en ambos periodos una mayor productividad, si bien la preservación de materia orgánica durante ciertas épocas ha podido ser sensiblemente mayor. En los distintos sapropeles se ha separado la barita para realizar un estudio comparativo de su naturaleza y preservación, comprobándose en todos los casos, independientemente de la edad y región considerada, que dicha barita presenta la morfología y composición habitual de la barita autigénica formada en la columna de agua, por lo que se corrobora el importante aumento de productividad que tuvo lugar durante el depósito de sapropeles y la óptima preservación del registro del Ba en el Mediterráneo. A pesar de amplio uso del contenido en Ba como indicador de productividad, los mecanismos de precipitación de la barita en la columna de agua están aun pobremente comprendidos. Para avanzar en el conocimiento de la biogeoquímica de este elemento también se ha realizado un estudio del Ba disuelto en la columna de agua en un transecto a lo largo del Mediterráneo y así dar un paso más en el entendimiento de su distribución en las distintas masas de agua de esta cuenca. Los valores obtenidos varían entre 6.5 y 11.1 µg/l, en acuerdo con datos publicados por otros autores. Existen variaciones significativas en profundidad y a lo largo del transecto estudiado, con un promedio de 8.3 µg/l en los primeros 200 m en la estación más próxima a Gibraltar y 7.5 µg/l en las regiones más orientales. A profundidades intermedias (200-600 m) la concentración va de 8.9 µg/l en el Mediterráneo occidental a 9.5 µg/l en el oriental. En aguas profundas la concentración es más uniforme, en torno a 10.3 μg/l. Aquí la concentración es más elevada debido a la disolución parcial de la barita que llega al fondo marino. En general, la distribución de Ba en profundidad refleja la precipitación de barita marina en la zona mesopelágica en relación con la degradación de la materia orgánica. Además, se han evaluado las distintas fuentes de Ba al Mediterráneo incluyendo, entre otras, el aporte del Atlántico y Mar Negro, el aporte fluvial y eólico, hidrotermal y de sedimentos profundos. Con respecto a esto se destaca que en ciertas regiones un elevado contenido en Ba podría relacionarse con el aporte local derivado de la disolución de barita, la influencia de sedimentos de cuencas hipersalinas o la difusión de aguas subterráneas. En cuanto a los mecanismos de precipitación de este Ba disuelto como barita, éstos se han discutido durante décadas. En general las aguas marinas están subsaturadas en Ba, por lo que otros mecanismos, como la actividad biológica, se hacen necesarios para explicar dicha precipitación. Se ha propuesto que la barita precipita en microambientes sobresaturados en Ba generados por la descomposición de la materia orgánica. Por otra parte, también desde hace décadas, se viene proponiendo la posible participación bacteriana en este proceso. Precisamente, en los grupos de investigación en los que se ha desarrollado esta tesis doctoral se viene investigando desde hace tiempo sobre este particular habiendo sido los primeros en haber obtenido la producción bacteriana de barita en condiciones de laboratorio. En primer lugar, en el año 2003, la obtuvieron con bacterias del suelo, concretamente con especies de mixobacterias y, posteriormente, en 2012 con bacterias marinas procedentes de colecciones de cultivo. En este segundo caso con varias cepas de diferentes especies: *Idiomarina loihiensis*, *Marinobacter hydrocarbonoclasticus* y *Planomicrobium okeanokoites*. Teniendo en cuenta la importancia de estos hallazgos, así como de las perspectivas que abrían en este campo de investigación se propuso, como uno de los objetivos de esta tesis doctoral, el ensayar la producción de barita con bacterias marinas aisladas directamente de sus hábitats naturales. La oportunidad para llevar a cabo esta investigación la brindaba el hecho de poder participar en la campaña "Ristretto e Lungo" organizada por la Universidad de Utrecht (Países Bajos), que permitía poder disponer de muestras de agua de mar procedentes de diferentes localizaciones y profundidades. Con esta finalidad se eligieron muestras procedentes de la cuenca Algero-Balear y del Mar Jónico. A partir de ellas, y utilizando un medio de cultivo adecuado para crecimiento general de bacterias marinas (el denominado Marine Broth, de DIFCO laboratories, gelificado con agar-agar), se procedió al aislamiento de numerosas colonias bacterianas. Este aislamiento se llevó a efecto de manera que el número de colonias aisladas supusieran, en conjunto, un número representativo de las que presentaban diferentes morfologías, tamaños, coloraciones, etc., caracteres todos ellos que podían indicar que las respectivas colonias estaban producidas por bacterias que pertenecían a especies y/o cepas diferentes. Siguiendo las técnicas adecuadas se procedió a la identificación molecular de cada uno de los aislados y se realizaron ensayos de producción de barita siguiendo la metodología desarrollada por los autores antes mencionados. Hay que destacar que estos ensayos han puesto de manifiesto que, de entre las bacterias aisladas, hay un número elevado de diversas cepas de especies diferentes pertenecientes, asimismo, a grupos filogenéticos diversos. En concreto se trata de cepas de los géneros *Marinobacter*, *Alteromonas*, *Pseudoalteromonas*, *Idiomarina*, *Bacillus* y *Brevibacillus*. Además de esta diversidad detectada entre las cepas aisladas en este trabajo hay que resaltar que, según se encuentra descrito en la bibliografía consultada, diversas cepas de estas especies se hallan distribuidas de manera abundante en muy diversas localizaciones y/o hábitats marinos y oceánicos. Por otra parte, los ensayos realizados con ellas en relación con su capacidad de producción de barita, en los medios de cultivo diseñados por los autores mencionados, han puesto de manifiesto dos cosas interesantes. En primer lugar, que en los casos en los que el crecimiento bacteriano en estos medios era notable se producían, también de manera abundante, precipitados de mayor o menor cristalinidad, mientras que en los casos en los que el desarrollo bacteriano era escaso la producción de esos precipitados no se daba o, al menos, esa producción era dudosa. Y, en segundo lugar, que de entre las cepas que claramente eran productoras de estos precipitados, esta producción se daba con mayor abundancia por las cepas bacterianas que presentaban una mayor presencia o cantidad de sustancias poliméricas extracelulares (EPS). El análisis de los precipitados producidos se realizó mediante difracción de rayos X y Microscopía electrónica de barrido de alta resolución acoplada con microanálisis de energía dispersiva de rayos X. Los resultados de estos análisis indican que se ha producido barita, pero además, junto con ella, otras fases minerales más o menos cristalinas o amorfas en las que conviven fosfato y sulfato de bario, pudiéndose apreciar fases en que predomina el fosfato de bario, prácticamente amorfas, y fases en las que, a medida que disminuye el contenido en fosfato y aumenta el sulfato de Ba, presentan un mayor grado de cristalinidad. Esto lleva a concluir, como ya apuntaron los autores mencionados más arriba, que el EPS bacteriano jugaría un papel destacado en la producción de barita: fijar el Ba a grupos fosfato presentes en estos polímeros extracelulares bacterianos, produciendo fosfato de Ba que posteriormente iría evolucionando a sulfato de Ba. Este sulfato, en el caso de los ensayos que se han llevado a cabo en el laboratorio, procedería del metabolismo bacteriano. ¿Qué ocurriría, entonces, en los hábitats marinos u oceánicos? ¿Qué papel jugarían allí las bacterias en relación con la producción de barita? En estos hábitats, donde la concentración de Ba es de subsaturación en relación a la que se precisa para precipitar barita, pero hay suficiente concentración de sulfato para que la misma ocurra, no sería necesario el aporte del mismo por el metabolismo bacteriano. Aquí, el papel de las bacterias sería fundamentalmente el aporte de EPS que ofrecería abundancia de grupos fosfato a los que se fijará el Ba, danto lugar a concentraciones locales del mismo suficientes para que, a través de la evolución que se acaba de mencionar, sea posible la producción de barita. En base a esta idea se hicieron los recuentos del número de unidades formadoras de colonias (UFC) totales en las muestras utilizadas, que dieron como resultado (7.19 ± 1.0) x 10^4 UFC/ml en la muestra de la cuenca Algero-Balear y (3.6 ± 1.2) x 10^5 UFC/ml en la muestra del Mar Jónico. Sin embargo, hay que tener en cuenta que el medio de cultivo Marine Broth es un medio de cultivo general y que, por tanto, no todas las bacterias presentes en las muestras pueden desarrollarse en el mismo sino solo aquellas cuyo tipo de metabolismo lo permita, por lo que es lógico deducir que el número de UFC presentes en el agua de mar del que proceden las muestras es muy superior al detectado en los ensayos. Teniendo en cuenta la enorme diversidad metabólica de las bacterias, lo que les permite vivir y desarrollarse en condiciones muy diversas, parece claro que su papel en la precipitación de barita ha sido en el pasado y lo es en la actualidad de gran relevancia. En consecuencia, por todo lo anteriormente mencionado se puede proponer que la producción bacteriana impactaría significativamente en la precipitación de barita, y esto sería un proceso importante a tener en cuenta cuando se utilicen indicadores de Ba en reconstrucciones paleoceanográficas.

Abstract

This thesis aims to investigate the Ba biogeochemical cycle in the Mediterranean Sea by analyzing this element in both the sedimentary record in the Mediterranean basins and in the water column, and also from exploring the barite ($BaSO_4$) precipitation mechanisms in marine environments. The biogeochemical cycle of Ba is closely related to the carbon cycle since this element is a proxy for marine biological productivity and, therefore, for the oceanic response to climate change. This is due to the abundant precipitation of barite in the water column in regions of high biological productivity, which results in high Ba concentrations in marine sediments. Although diverse proxies have been traditionally used to reconstruct biological productivity, barite has revealed as a reliable proxy being usually well preserved in the sedimentary record under enough oxygenated conditions, and it has been widely used for paleoceanographic reconstructions. In the case of the Mediterranean Sea, its semi-enclosed character and restricted water circulation have amplified its response to climate variability, in particular regarding marine biological productivity. Thus, during periods of climate change towards conditions of higher humidity and precipitation, the enhanced nutrient input led to a significant increase in productivity. In addition, the high contribution of freshwater also entailed stratification of the water masses leading to decreasing oxygenation in deep waters, and enhancing organic matter preservation and sapropel deposition. In this thesis, a detailed analysis of the Ba content in sapropel layers in the different Mediterranean basins has been made in order to assess their spatial and temporal variability, as well as the Ba record preserved within sapropels. Regarding the differences between the Mediterranean basins, higher sedimentation rates in shallower areas have led to a significant dilution of the Ba content. Moreover, the spatial variability is consistent with the Nile being the main source of nutrients. In the western Mediterranean, Organic Rich Layers have been deposited with lower organic carbon content and Ba than sapropels. For instance, in the Tyrrhenian, although sapropels have been deposited, the Ba content is significantly lower than in the easternmost basins. Temporal variability is even more significant. In general, Pleistocene sapropels have a higher Ba content than their younger equivalents, in particular, the most recent sapropel (S1). The organic carbon content also depends on

preservation, being particularly high in Pliocene sapropels due to the development of anoxic environment. However, the Ba content is comparable to those of the Pleistocene, indicating greater productivity in both periods, although the preservation of organic matter during the Pliocene may have been significantly higher. Barite has been separated from different sapropel layers to compare its nature and preservation. It is demonstrated that regardless of the age and location, barite has the usual morphology and composition of the authigenic barite formed in the water column, which corroborates the significant increase in productivity during sapropel deposition and the optimal preservation of the Ba record in the Mediterranean. However, despite the wide use of the Ba content as a productivity proxy, the mechanisms leading to barite precipitation in seawater are still poorly understood. To advance in the knowledge of the biogeochemistry of this element, a study of dissolved Ba in the water column has also been carried out in a transect along the Mediterranean to further understand its distribution in the different water masses of this basin. The values obtained vary between 6.5 and 11.1 µg/l, in agreement with data published by other authors. There are significant variations with depth and along the studied transect, with an average of 8.3 µg/l in the first 200 m in the closest station to Gibraltar and 7.5 µg/l in the easternmost region. At intermediate depths (200-600 m) the concentration ranges from 8.9 µg/l in the western Mediterranean to 9.5 µg/l in the eastern basins. In deep waters, the concentration is more uniform, around 10.3 µg/l. It is higher due to the partial dissolution of barite that reaches the seafloor. In general, the Ba distribution in deep waters reflects the precipitation of marine barite in the mesopelagic zone. In addition, the different sources of Ba to the Mediterranean have also been evaluated, including, among others, the contribution of the Atlantic and the Black Sea, the fluvial and wind inputs, as well as the hydrothermal and deep sediment contributions. It is also highlighted that in certain regions a high content of Ba could be related to the local contribution derived from the dissolution of barite, the influence of sediments from hypersaline basins, or the diffusion from groundwater. Regarding the Ba precipitation mechanisms, these have been debated for decades. In general, marine waters are subsaturated in Ba, so biogeochemical processes would be required to explain barite precipitation. It has been suggested that barite precipitates in Ba-supersaturated microenvironments where the decomposition of organic matter occurs. The bacterial role in this process has also been suggested. In the research groups in which this doctoral thesis has been carried out, this has been investigated since time ago, and it was demonstrated for the first time that microbial barite can precipitate under laboratory conditions. In 2003 barite was obtained in experiments using soil bacteria, specifically Myxobacteria species, and later on, in 2012 using marine bacteria from culture collections. Several strains of different species: *Idiomarina loihiensis, Marinobacter hydrocarbonoclasticus and Planomicrobium okeanokoites* were used in those experiments. Considering these important findings, as well as the field of research opened, this thesis was conceived to test the precipitation of barite by marine bacteria isolated directly from their natural habitats. The participation in the "Ristretto e Lungo" cruise (University of Utrecht) provided samples of seawater from different locations and depths across a Mediterranean transect, and two stations were selected, one in the Algero-Balearic basin and another one in the Ionian Sea. A suitable culture medium for the usual growth of marine bacteria (the so-called Marine Broth, from DIFCO laboratories, gelled with agar-agar), was used and numerous bacterial colonies were isolated. Bacteria were isolated in a way that the whole isolated colonies would represent different morphologies, sizes, colour, etc. in turn representing bacteria belonging to different species and/or strains. Appropriate techniques were used for molecular identification of each bacterial isolate and barite production assays were carried out following the methodology previously developed by the host group. These assays have shown that among the isolated bacteria there is a high number of strains of different species belonging to diverse phylogenetic groups. Specifically, these are strains of the genera *Marinobacter, Alteromonas, Pseudoalteromonas, Idiomarina, Bacillus* and *Brevibacillus*. In addition to this diversity among the strains isolated, several strains of these species are also abundantly distributed in diverse locations and/or marine and oceanic habitats. The assays carried out with such genera to test their barite production capacity have provided new insights into barite precipitation: i) when the bacterial growth in these media was significant, precipitates with different crystallinity were abundantly produced, whereas in the cases in which the bacterial growth was scarce the production of those precipitates did not occur or, at least, the production was questionable. ii) Among the strains that were clearly producers of these precipitates, this production was abundant in the cultures with bacterial strains showing large quantities of extracellular polymeric substances (EPS). The analysis of the precipitates was carried out using X-ray diffraction and high-resolution scanning electron microscopy coupled with X-ray energy dispersive microanalysis. The results of these analyses have served to demonstrate that barite shows different crystallinity, including mineral phases where barium phosphate predominates which mostly amorphous, and phases in which the phosphate content decreases while the Ba sulfate increase, showing higher degree of crystallinity. Hence, as already suggested in previous studies, it is demonstrated that bacterial EPS play a major role in barite precipitation by fixing Ba ions to phosphate groups present in these bacterial extracellular polymers, and producing barium phosphate that would eventually evolve to Ba sulfate. Under experimental conditions, the sulfate derives from bacterial metabolism. However, what would it happen in marine habitats? What would be the role of bacteria in barite production? In these habitats, where the concentration of Ba is undersaturated in relation to that required for barite precipitation, there is enough sulfate thus the sulfate supply by bacterial metabolism is not required. Here, the role of bacteria would be essentially producing EPS that would provide abundant phosphate groups to which Ba binds, this would increase local Ba concentrations triggering the aforementioned crystallization process. Based on this hypothesis, the total number of colony-forming units (CFU) was counted, which resulted in (7.19 ± 1.0) x 10^4 CFU/ml in the sample from the Algero-Balear basin and (3.6 ± 1.2) x 10^5 CFU/ml in the Ionian Sea sample. However, it must be also taken into account that the Marine Broth culture medium is a general culture medium and not all bacteria present in the samples can properly grow in it but only those whose type of metabolism allows growing. Therefore, it is logical to deduce that the number of CFUs present in the seawater from which the samples come is much higher than the detected in the tests. Taking into account the high metabolic diversity of bacteria, which allows them to live and develop in diverse conditions, it is suggested that their microbial role in barite precipitation has been of major importance over geological time scales. In summary, it is demonstrated that bacterial production has a significant impact on barite production, and this would be a major process to take into account when using Ba proxies for paleoceanographic reconstructions.

Chapter I. Introduction

At present, gaining further knowledge on climate change impacts and climate responses is essential to assess future climate change scenarios. In this regard, oceans play a major role as essential components of our climate system. In recent decades they have undergone major physical and biogeochemical modifications in response to natural and human-induced global change. Warming, acidification, deoxygenation and changes in primary productivity are considered as four major stressors of ocean ecosystems (e.g., Bopp et al., 2013). In the coming decades the effect of these stressors will be amplified. The Mediterranean Sea in particular is considered an important hot spot for climate change and one of the most vulnerable regions with regard to the aforementioned ocean stressors. Hence, understanding temporal and spatial variations in ocean productivity is fundamental for assessing global changes. Most productivity reconstructions to date are based on Ba proxies, whereas the nature of marine barite and mechanisms for Barium precipitation in seawater are largely unknown. The biogeochemistry of barium has intrigued geochemists for decades because of its nutrient-like distribution in seawater and its precipitation as barite in the water column, despite widespread undersaturation conditions (Monnin et al., 1999). To explain this apparent contradiction, it has been suggested that barite precipitates in barite-supersaturated microenvironments generated by the biological degradation of sinking organic matter (Dehairs et al., 1980; Bishop et al., 1988). This is of great relevance, since barium fluxes to the deep ocean and barite accumulation in sediments is widely used as a proxy to reconstruct past ocean export productivity (e.g., Paytan and Griffith, 2007); this in turn has implications for reconstructions of the marine carbon cycle and global climate. It has recently hypothesized that barite formation in the ocean may be linked to microbial activity (González-Muñoz et al., 2003; 2012; Martínez-Ruiz et al., 2018, 2019, 2020), a possibility supported by the correlation between the abundance of bacterial activity and barite in the ocean water column (Dehairs et al., 2008; Jacquet et al., 2011; Planchon et al., 2013). The capability of certain marine bacteria to mediate barite precipitation under experimental conditions has also served to demonstrate that bacteria likely mediate barite precipitation in natural environments. Within this context, exploring barite precipitation and the biogeochemistry of Ba in the Mediterranean are the main aims of this thesis.

I.1. The global carbon cycle and productivity oscillations

Carbon is the fourth most abundant element on Earth and it is the fundamental basis of life, its biogeochemical cycle describing the carbon fluxes between atmosphere, biosphere, hydrosphere and geosphere. A continuous CO_2 exchange between the atmosphere and the oceans takes place, oceans being major carbon reservoirs, occupying 70% of the Earth´s surface. The distribution of sources and sinks depends on oceanic circulation and biological productivity, since the efficiency of the ocean as a CO_2 buffer relies on temperature and density. CO_2 is more soluble in cold and saline waters, but also depends on atmospheric CO_2 concentrations regulated by the biological pump (Passow and Carlson, 2012). The emission of greenhouse gases to the atmosphere raises temperatures, decreasing carbon sequestration efficiency in oceans due to lower CO_2 dissolution rates at high temperatures. The biological pump plays an important role in ocean chemistry and the global carbon cycle, through carbon sequestration mediated by biological productivity (Figure 1), which transforms inorganic carbon from atmospheric CO_2 (dissolved inorganic carbon, DIC) into organic carbon (or dissolved organic matter, DOM) that is vertically transported to the deep ocean (Chisholm, 2000; Meyer et al., 2016). Organic carbon is commonly divided into particulate organic carbon (POC), composed of phytoplankton, zooplankton, bacteria and detritus; and dissolved organic carbon (DOC), mainly composed of molecules and mostly produced in the surface layers by phytoplankton (Eppley and Peterson, 1979; Volk and Hoffert, 1985; Cavan et al., 2017; Boyd et al., 2019; Kujawinski, 2011; Hansell, 2013). Dissolved organic carbon is mostly used as nutrient by microrganisms, while larger organisms consume particulate organic carbon (e.g., Middleburg, 2019). DOC is remineralized by heterotrophic prokaryotes through the water column, then exported to the deep ocean, where it forms the largest reduced carbon pool in the biosphere. In the euphotic zone (0-200 m), the DOC concentrations ($10 - 10^2$ µmol/l) are usually one order of magnitude higher than those of POC (1 – 10 µmol/l), while in sediments the trend is the opposite, being of the order of 10^6 µmol/l and DOC in pore waters of $10^2 - 10^3$ µmol/l (e.g., Wilson and Arndt, 2017). DOC concentrations decrease downward in the water column, for instance in the central Red Sea, where the DOC values (from 41.4 to 95.4 µmol/l) are roughly 70.0 µmol/l in the epipelagic zone, then decrease to 50.7 µmol/l in the mesopelagic zone (Calleja et al., 2019). In the Weddell Sea the average water column DOC is 52 ± 11 µmol/l, in surface waters up to 69 µmol/l, and within denser water layers, in the lower pelagic zone, up to roughly 61 µmol/l (Geuer, 2015). In the Equatorial Pacific, the DOC ranges are 60 – 70 µmol/l and the deep water values range 35 – 40 µmol/l (Sharp et al., 1995). After atmospheric CO_2 sequestration and its transformation into organic carbon by the biological pump, it is again transformed into DIC and eventually upwells to the surface (Álvarez-Salgado et al., 2001; Mills et al., 2004; Rykaczewski and Dunne, 2010). Roughly 25% of the carbon fixed in the upper ocean sinks to the depths (Falkowski et al., 2000), and only a small fraction of the POC can elude the mineralization processes and contribute to accumulation and burial in sediments, some 0.5% of the gross production (Hedges and Keil, 1995; Passow and Carlson, 2012; Boyd et al., 2019). The current carbon oceanic pool is of 624 Gt C, comparable to the atmospheric CO_2 reservoir of roughly 750Gt C, which highlights oceans as an efficient atmospheric CO_2 removal system (Hansell, 2013; Hansell and Carlson, 2015; Calleja et al., 2019). Thus, an understanding of past variations related to biological pump efficiency, through paleoproductivity reconstructions, can be considered a key factor for assessing current climate change (e.g., Paytan et al., 1996; Capotondi et al., 2016; Diester-Haass et al., 2018). Paleoclimate and paleoproductivity reconstructions are based on a wide range of proxies. Among others, Ba excess is held to be a reliable proxy for productivity reconstruction when deriving from biogenic barite, as barite is a refractory well preserved mineral in the sedimentary record under oxygenated conditions (Gingele et al., 1999; Paytan and Griffith, 2007; Liguori et al., 2016). The mineral barite (barium sulphate, $BaSO_4$) forms and accumulates below areas of high productivity (Pfeifer et al., 2001; Griffith and Paytan, 2012; Costa et al., 2017; Diester-Haass and Faul, 2019). A close link between Ba excess and organic carbon has been demonstrated both in sediment traps and in sediments (Dymond et al., 1992; Dymond and Collier, 1996; Schoepfer et al., 2015). Marine barite crystals, ranging in size from 0.5 to 5 µm, precipitate in the water column within Ba-saturated decaying organic matter microenvironments, even though the ocean is subsaturated in this element (Monnin et al., 1999; Rushdi et al., 2000; Horner et al., 2017). The link between Ba excess in marine sediments and higher biological productivity was already suggested by Goldberg and Arrhenius in 1958 in the Pacific, then by Turekian and Tausch in 1964 in the Atlantic. Since then, many studies have demonstrated the association between barium distribution and carbon fluxes (e.g., Dymond and Collier, 1996; Jacquet et al., 2004, 2005; Guieu et al., 2005; Paytan and Griffith, 2007; Jacquet et al., 2016; Pyle et al., 2018; Carter et al., 2020; Martínez-Ruiz et al., 2020). Dissolved barium has a nutrient-like behavioral profile in the water column, with low concentrations in surface waters and increased content in deep waters (Jeandel et al., 1996; Paytan and Griffith, 2007; Roeske et al., 2012). Particulate barium has a maximum 100-600 m water depth within the mesopelagic zone (Dehairs et al., 1997; Jacquet et al., 2005). The mechanisms controlling Ba precipitation in the water column are not fully understood, despite the widespread use of Ba proxies to reconstruct productivity and the clear link between the Barium cycle and biological processes in the ocean (Lemaitre et al., 2018; Figure 2). Although the formation of microenvironments where marine barite precipitates requires barium and sulphate saturation (e.g., Horner et al., 2017), the processes leading to such saturation are not well known. It has been shown that diatoms, acantharia and fecal pellets can provide Ba ions, but their abundance is insufficient to explain Ba concentrations in water and sediments (Fresnel et al., 1979; Gooday and Nott, 1982; Bernstein et al., 1992; Bertram and Cowen, 1997). The precipitation of barite by bacteria (González-Muñoz et al., 2003, 2012) was a major finding in this respect, advancing our understanding of the organisms involved in marine barite precipitation mechanisms.

I.2. Barium biogeochemistry

Barium is a relatively abundant trace element in the oceans, its concentrations varying from 30 to 140 nM, and having a residence time (RT) of roughly 10,000 years (Chan et al., 1976; Rubin et al., 2003; Hsieh and Henderson, 2017). Ba usually shows a spatial correlation with Si distribution even though their cycles are decoupled, with a nutrient-type vertical profile increasing from 30-60 nM in surface waters —except in upwelling or river areas— to 140 nM in deep waters (Chan et al., 1976; Jeandel et al., 1996; Jacquet et al., 2007, 2016). Dissolved Ba concentrations in the Atlantic (Chan et al., 1976), the Indian and the Southern Ocean (Jeandel et al., 1996; Jacquet et al., 2004, 2007, 2008; Hoppema et al., 2010), the North Pacific (Dehairs et al., 2008) and the Mediterranean Sea are of the same order of magnitude, although the Mediterranean vertical gradient is smaller compared to the open oceans. This suggests that the Mediterranean Sea is even more undersaturated in Ba than other ocean waters; its dissolved Ba distribution appears to be mainly affected by water circulation in the epipelagic and mesopelagic zones (Pyle et al., 2018). Meanwhile, the high Ba concentrations in deep waters are linked to particulate Ba dissolution and could be further enhanced by low ventilation and residence time (Chan et al., 1976; Jacquet et al., 2016). The nutrient-type behavior of Ba is reflected in its isotopic variations in the water column, affected not only by hydrodynamic processes but also by biogeochemical processes (Horner et al., 2015; Cao et al., 2016; Bates et al., 2017). Surface waters are relatively depleted in the lighter Ba isotope due to organic matter production; in contrast, deep waters are enriched in the light isotope. This isotopic fractionation points out that Ba is incorporated into organic matter during marine barite precipitation, the main Ba carrier mineral, which latterly sinks to the deep ocean and may partially dissolve. Seawater $^{\delta138/134}$Ba values range from 0.24 to 0.65 ‰ (Hsieh and Henderson, 2017), yet in open ocean sediments the value decreases to 0.09 ± 0.01 ‰, correlated with particulate Ba sinking to the deep ocean (Horner et al., 2015; Bridgestock et al., 2018). The biogenic fractionation of Ba isotopes supports its use as a proxy for land–sea interactions and ocean mixing processes (Horner et al., 2015; Cao et al., 2016; Bates et al., 2017; Hsieh and Henderson, 2017). Regarding barium distribution in the ocean water column, it has been shown that higher mesopelagic particulate Ba (marine barite) is tied to higher bacterial production (Dehairs et al., 2008; Jacquet et al., 2011; Planchon et al., 2013). The precipitation of barite under laboratory conditions (González-Muñoz et al., 2003, 2012; Martínez-Ruiz et al., 2018) further supports this link. Under experimental conditions, extracellular polymeric substances (EPS) serve as nucleation sites to concentrate Ba ions (Martínez-Ruiz et al., 2018). These findings point to the same role in the seawater column, given the presence of abundant bacteria in natural environments. It has moreover been demonstrated that microorganisms can generate a saturation of sulphates through sulphur compound oxidation as well as enhance Ba accumulation linked to biofilms (Glamoclija et al., 2004; Sanchez-Moral et al., 2004; Senko et al., 2004; Bonny and

Jones, 2008; Stevens et al., 2015; Singer et al., 2016). Although the exact mechanisms for barite are still poorly understood, it has been demonstrated that bacterial cells and EPS phosphate groups act as precursors of barite (Martínez-Ruiz et al., 2019).

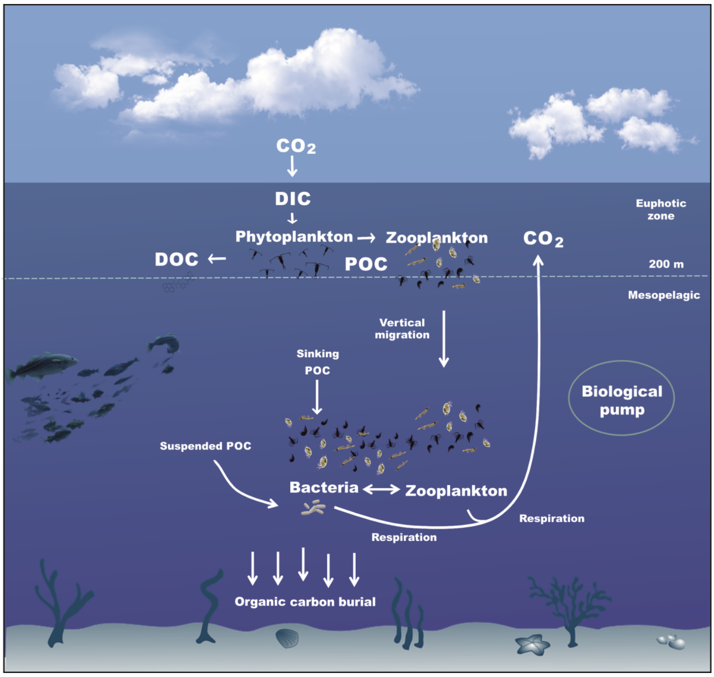

Fig.1. Simplified scheme of the biological pump in oceans.

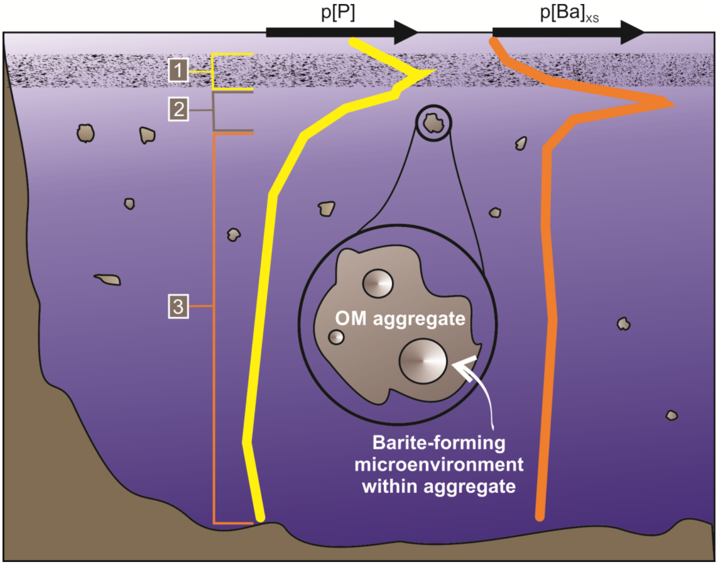

Fig.2. Conceptual model of barium cycling in low-sulphate water columns. Depth profiles of p[P] and p[Ba]XS in Lake Superior (North America), an extreme Ba undersaturated environment alike to those occurring in marine basins. (1) Autotrophic production leads to a peak in OM (organic matter) near the surface, indicated by the maximum in p[P]. (2) Microbial respiration within aggregates of decaying OM leads to development of barite-supersaturated microenvironments and precipitation of barite. (3) Continued respiration diminishes OM concentrations and destroys protected microenvironments, preventing further build-up of Ba^{2+} and sulphate ions and thus any additional barite formation; settling barites are exposed to undersaturated waters and may start to dissolve. This scheme highlights the necessity of a constrained framework for barite precipitation and the importance of the microenvironment (From Horner et al., 2017).

The role of EPS is particularly important in this regard, living cells are not a requirement for cation bioadsorption in carbonate precipitation, since it can also occur in EPS without living cells (Bontognali et al., 2014). In general, bacterial biomasses favoring accumulation mechanisms, and not only of Ba but also of diverse metals (Kikuchi and Tanaka, 2012; Merroun and Selenska-Pobell, 2008; Sheshadri et al., 2012). A soil bacterium (*Myxococcus xanthus*) was the first used to test prokaryotic barite precipitation under laboratory conditions showing Ba bioadsoption (González-Muñoz et al., 1997, 2003). These landmark studies using bacteria, were preceded by others reporting intracellular barite crystals in some algae and protozoa, though their abundance cannot account for that of marine barite (Fresnel et al., 1979; Bernstein et al., 1987, 1992; Levin, 1994; Bertram and Cowen, 1997). In sum, it has been broadly demonstrated that organic microenvironments linked to decaying organic matter are appropriate sites for Ba saturation; therefore, the Ba cycle is closely related to the carbon cycle.

I.3. The microbial role in mineral precipitation

Mineral precipitation derives from processes that separate solids from a solution through changes in physical and chemical conditions: pressure, temperature, saturation and biological activity. Even though the body of current knowledge about microorganisms is broad, only about 1% of them are known to play an important role in mineral precipitation. Diverse prokaryotic precipitation mechanisms have been reported. For instance, certain bacteria use metal ions as an energy source, reducing them and releasing energy used for bacterial processes; they are thereby able to bioremediate contaminated soils and waters by mine waste (e.g., Merroun et al., 2006; Newsome et al., 2014; Chung et al., 2014; Romero-González et al., 2016). Chromium reduction by microbes can transform a highly bioavailable and toxic species into a comparatively immobile Cr species, helping to mitigate its toxicity. While the copper present on electronic circuit boards can be extracted using acids or high temperatures, its recovery through biolixiviation is a cheaper and more eco-friendly process (Chen et al., 2015; Yang et al., 2014; Awasthi et al., 2016). Certain iron, copper, uranium and gold ore deposits have been traditionally attributed to inorganic precipitation, but this paradigm changed when it was shown that they can be mediated by microorganisms (Sillitoe et al., 2005; Sanyal et al., 2019; Tornos et al., 2019). Microorganisms also affect aquifers by modifying the environmental conditions controlling dissolution processes, increasing dissolution rates of the host rock; and the mineral background of sediments or rocks can also be affected by microbial communities (e.g., Rodríguez-Navarro et al., 2012). Overall, microbes play a major role in many biogeochemical cycles through mineral and metal speciation, mineral precipitation and mineral dissolution. Distinguishing between biotically-mediated minerals and abiotically-mediated precipitates is no straightforward matter, however. Crystal shape is usually indicative of microbial precipitation. Spheres, ovoids, dumbells and rounded shapes may be linked to such precipitation, with amorphous phases as mineral precursors. Still, morphology cannot be systematically applied, as inorganic precipitation also results in spherical morphologies (González-Muñoz et al., 2003, 2008, 2010; Bonny and Jones, 2008; Morcillo, 2010; Rodríguez-Navarro et al., 2007; 2012; Widanagamage et al., 2018). In the case of barite, morphology is certainly characteristic: authigenic barite, diagenetic barite and hydrothermal barite are clearly differentiated. In addition, Sr and S isotopes with crystal morphology characterize barite origin (Paytan et al., 2002). Because organic interfaces enhance strontium content in marine barite (Wallace, 2019), this would lead to an enriched lighter Sr isotope. Sr isotopic fractionation also sheds light on authigenic precipitation and recrystallization (Griffith et al., 2018). Moreover, sulfur and oxygen isotopes can serve to distinguish between sulphate sources and identify biochemical processes such as sulphate reduction.

Bacterial precipitation mechanisms can either be controlled by microorganisms (Mann et al., 1983) or be the indirect consequence of other biological processes, known as biologically induced mineralization (Lowenstam, 1981). In the case of bacteria, except in very specific cases (e.g., magnetosomes), the precipitation processes are considered as biologically induced mineralization. Bacteria promote this kind of mineralization by providing surfaces to bind ions, and/or through their own metabolism. In the former case, the cellular walls and the bacterial secreted polymers contain a large quantity of ligands that participate in cation fixation processes. The cations associate to become a crystallization nucleus, and the type and space of ligands on a surface will condition the cation type to be fixed, as well as the orientation and structure of a future nucleus (Fowle and Fein, 2001; Bazylinski et al., 2006). In the latter case, involving the bacterial metabolism, activity can affect metabolite concentration in the solution and the surrounding parameters determine the precipitation (or not) of some compounds. All these processes are known in general as biosorption, which includes adsorption, absorption, ionic exchange, binding to a surface and precipitation (Gadd, 2009). They require the saturation of a cation or compound in the solution along with adequate thermodynamic and kinetic conditions. Any structure containing proteins, nucleic acids, and polysaccharides can provide sites to fix metallic ions and other elements, their composition affording different functional groups for binding. The cellular wall, the extracellular polymers and the S layer are the most important sites, being external structures in direct contact with the area surrounding the cell. The anionic functional groups present in the cell walls of Gram-positive and Gram-negative bacteria are responsible for the global negative charge;

and the abundant phosphate groups allow for binding with diverse elements (Van der Wal et al., 1997; Zhou et al., 2007). Furthermore, as most microorganisms secrete EPS on their surface —generally composed by polysaccharides and less frequently by proteins— the polymers of this material are also negatively charged due to the presence of uronic acids, pyruvate, phosphate or sulphate. In general, the polymers forming the EPS are produced and secreted by bacteria and other microorganisms to promote cell adaptability. Merroun et al. (1998, 2001, 2003) demonstrated the implication of EPS in silver, plumb and lantene sorption by *M. xanthus*, while González-Muñoz et al. (1997) described uranium sorption by the same bacterium. Other authors have studied the sorption capacity of EPS isolated from bacteria (Guibaud et al., 2008; Morillo-Pérez et al., 2008; Sun et al., 2009). Mata-Gómez (2006) analyzed halophylic microorganisms of the genera *Halomonas, Alteromonas* and *Idiomarina,* among others, finding that all the studied EPS were heteropolysaccharides mainly made up of carbohydrates and organic components, but also containing an inorganic fraction as sulphates and phosphates. In general, sulphates and phosphates are usual ligands of the EPS; their presence lends the exopolysaccharides significant binding properties. The structure of the most abundant insoluble phosphate, apatite [$(Ca_5(PO_4)_3(F,Cl,OH)$], is very tolerant to substitutions and could incorporate most of the elements in its crystalline structure (McConnell, 1973). González-Muñoz et al. (2010) showed that soil bacteria of the genus *Myxococcus* induced precipitation of a number of phosphates, carbonates, sulphates, chlorides, oxalates and silicates. Thus, the ionic exchange on the bacteria cell surface and EPS could have a role in the creation of nucleation sites for a wide variety of minerals, including marine barite. It is also known that EPS play a major role in barite precipitation in the ocean, supporting the link of bacterial production and Ba distribution (Martínez-Ruiz et al., 2018, 2019).

I.4. Barium cycling in the Mediterranean

The Mediterranean Sea constitutes a natural laboratory of great relevance, being particularly sensitive to climate variability owing to its semi-enclosed basin character and restricted water circulation. The Mediterranean Sea sediment records contain the so-called "sapropels" (Kidd et al., 1978): organic-matter-rich sediments with dark layers of centimetric thickness having over 2% of Total Organic Carbon (TOC) that are highly enriched in Ba. These deposits, related to variations in environmental conditions, hold keys to understand climate variability and paleoceanographic evolution of the Mediterranean basins. The deposition of sapropels results from a combination of decreased oxygen concentrations at depth and enhanced biological productivity in surface waters (Rohling and Hilgen, 1991; Rohling et al., 2015). The precession minima in the North Hemisphere led to a maximum summer insolation (Rohling and Hilgen, 1991) with a subsequent maximum monsoon activity (Rossignol-Strick and Paterne, 1999). The thermal differences between the ocean and the continent, marked by seasons, provoked an increase in North Africa precipitation, thus a maximum Monsoon. In the Mediterranean, the increase in humidity led to higher river runoff (Rossignol-Strick and Paterne, 1999). The North African Monsoon affected the eastern Mediterranean Sea by providing a high input of nutrients from rivers, which enhanced primary productivity. The river runoff moreover led to density changes in the water column, promoting lower oxygenation in deep waters and poor ventilation. The combination of both factors produced appropriate conditions for organic matter accumulation and for preservation. Spatial productivity variations are reflected by Ba content in Mediterranean sapropels (e.g., Martínez-Ruiz et al., 2000, 2003; Weldeab et al., 2003; Moller et al., 2012; Zwiep et al., 2018). At present, productivity is low and Ba profiles suggest that internal Ba cycling in the Mediterranean is of minor importance compared to fluxes through the system (Dehairs, 1987), a hypothesis supported by Jacquet et al. (2016) with their complete dataset of dissolved Ba for the Mediterranean Sea, showing Ba distribution is mainly driven by hydrodynamics. Here, the Geotraces project has provided trace elements and isotope data that serve to characterize diverse processes and quantify Ba fluxes. The precise Ba distribution in seawater in the Mediterranean Sea analyzed during the GEOTRACES GA-04S MedSeA cruise (Roy-Barman et al., 2019) pointed to more intense mean particulate Ba cycling than previously estimated, though inputs from Messinian evaporites call for further study.

Chapter II. Objectives

The objectives of this thesis rely on significant previous achievements regarding the bacterial precipitation of barite. In particular, the precipitation of barite by bacteria (González-Muñoz et al., 2003, 2012) opened an interesting research field surrounding the Ba biogeochemical cycle and productivity reconstructions. This thesis is contributing to further understanding of the marine barite precipitation mechanisms, with implications for the Ba cycle, but also for paleoproductivity reconstructions and understanding the link between productivity and climate change. Within this context, the thesis mainly addresses the particulate and dissolved Ba distribution in the water column and in the Mediterranean sedimentary record as well as the microbial barite precipitation mechanisms. The Mediterranean basins are of particular interest in this regard since, at the geological scale, they record an important biogenic Ba accumulation linked to cyclic productivity increases deriving from higher humidity and increasing precipitation. Hence, the main objective of this work is to contribute new knowledge regarding the still poorly understood Ba biogeochemical cycle, strongly correlated to the carbon cycle, which is of capital importance in the context of global change. This main objective includes diverse specific objectives:

1. Establish the interrelations between primary productivity variations and the Ba distribution in the sedimentary record, linking paleoproductivity oscillations to climate variability in the Mediterraneans basins over geological time scales. To do so, the Ba distribution in Mediterranean Sea sediments will be reviewed, and mineralogical and crystallographic studies will be performed on barite separated from sapropels from the eastern Mediterranean basin.

2. Determine the distribution of dissolved Ba in the Mediterranean. A high-resolution barium profiles from the water column through a Mediterranean Sea transect will be provided and compared with those from literature to better assess Ba budgets in the Mediterranean Sea. Ba sources will be also discussed.

3. Further investigate the bacterial barite precipitation mechanisms and establish the role that bacteria play in the Ba biogeochemical cycle. Mineral precipitation experiments using microbiological cultures will be carried out, with the purpose to demonstrate if marine bacteria isolated from natural seawater samples can promote barite precipitation.

The proposed objectives are intended to improve our knowledge of the Ba biogeochemical cycle and the role of bacteria in barite precipitation as well as the implication of microbial processes for productivity reconstructions based on Ba proxies.

Chapter III. Methods

The data and materials used for this study were collected during diverse oceanographic cruises and also derived from further investigations related to such campaigns. The materials used, as well as the experiments and analyses performed, are summarized below.

III.1. Sampling

III.1.1. Water sampling for bacteria isolation

For bacterial isolation and culture experiments, samples were obtained during the oceanographic campaign "Ristretto e Lungo M83/3" on board research vessel "R/V Meteor". During this cruise, water samples were collected at different depths (between 50 and 1200 m depth) with a Niskin bottle rosette, which incorporates a recording system that measures conductivity, temperature and depth, as well as other measurements such as fluorescence, salinity and oxygen. The depths for collecting the studied water samples (Table 1) were selected according to the CTD (Conductivity, Temperature and Depth) profiles obtained at each station, taking into account the maximum productivity inflections observed in the fluorescence, oxygen, temperature and salinity profiles, as well as the matching the maximums of productivity (e.g., Church and Wolgemuth, 1972; Van Beek et al., 2009). The samples were stored at 4° and preserved for bacterial isolation.

Table 1. Water depth, coordinates, temperature and salinity at the stations where selected samples were selected for culture experiments.

Sample	Depth	Latitude	Longitude	T (°C)	S (g/l)
RL 33 CT_10	250 m	37° 44.05' N	2° 12.43' E	13.5	38.5
RL 81 CT_7	200 m	34° 43.00' N	22° 12.90' E	15.8	39

III.1.2. Sampling to study Ba distribution in the water column

During the past decade, high-resolution data of the water column have been acquired on scientific cruises conducted around the Mediterranean Sea (1999 "Smilable" with RV Logachev; 2010-2011 "Ristretto e Lungo" with RV Meteor; 2000 "Pasap", 2008 "Doppio", 2009 "Macchiato" and 2012 Cortado with RV Pelagia. Respective codes: SL, RL, PS, DP, MP, CP). For this work, seawater samples were recovered from different stations and some of them were selected to study a west to east Mediterranean Sea transect (Table 2, Figure 1). The samples were taken at several depths in each station (about 24 depths per station) with a rosette of Niskin bottles, 12 litres volume, featuring a CTD system that analyzes several physical parameters at the very time of sampling. Turbidity, salinity, temperature, fluorescence and depth were measured with this system. Furthermore, on board measurements were performed for oxygen, nutrients (nitrate, phosphate and silicate), DIC and total alkalinity, described in detail below.

Table 2. Stations, water depth and sampling date of the stations along the studied Mediterranean Sea transect.

Station		Water depth (m)	Latitude	Longitude	Date
1	RL09CT	831	36° 06.11' N	4° 53.03' W	29/12/2010
2	CP04CT	2686	37° 24.48' N	0° 27.50' E	22/11/2011
3	RL33CT	2701	37° 44.05' N	2° 12.43' E	01/01/2011
4	CP05CT	2704	38° 10.14' N	4° 29.44' E	23/11/2011
5	MP01CT	2780	39° 49.54' N	5° 55.94' E	26/11/2009
6	DP08CT	2482	38° 20.80' N	8° 19.00' E	22/10/2008
7	RL39CT	2908	34° 40.78' N	17° 24.56' E	05/01/2011
8	DP17CT	2306	38° 28.36' N	18° 03.50' E	25/10/2008
9	PS67CT	3311	36° 52.49' N	19° 41.12' E	25/05/2000
10	CP21CT	3065	34° 23.61' N	20° 02.10' E	29/11/2011
11	CP47CT	3488	35° 15.28' N	21° 27.74' E	06/12/2011
12	SL76CTD	655	40° 18.90' N	25° 10.48' E	02/09/1999
13	SL56CTD	1513	35° 97.70' N	26° 34.94' E	30/08/1999
14	SL26CTD	2001	33° 39.98' N	33° 00.99' E	24/08/1999

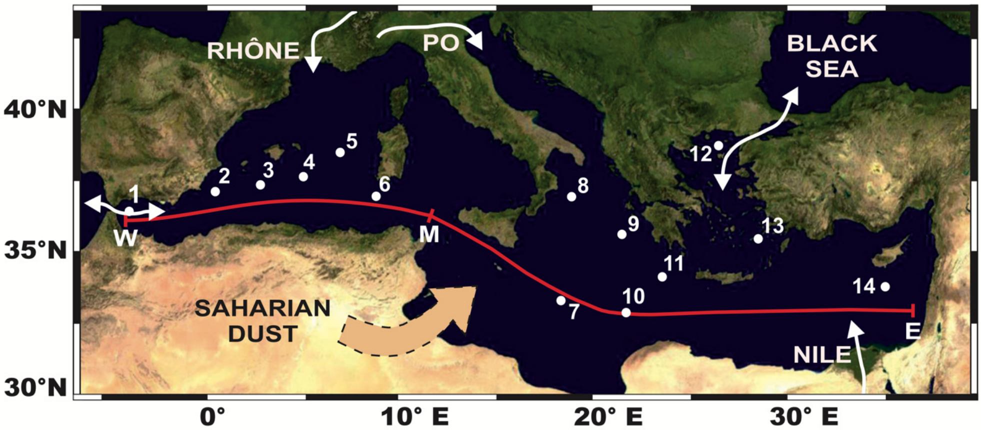

Fig. 1. Studied Mediterranean Sea transect. Numbers indicate the location of the stations where seawater samples were collected during the scientific cruises across the Mediterranean Sea basins. Numbers indicate the location of the stations where seawater samples were acquired on scientific cruises conducted around the Mediterranean Sea (1999 “Smilable” with RV Logachev; 2010-2011 “Ristretto e Lungo” with RV Meteor; 2000 “Pasap”, 2008 “Doppio”, 2009 “Macchiato” and 2012 “Cortado” with RV Pelagia. Respective codes: SL, RL, PS, DP, MP, CP). Map source: Visible Earth-Blue Marble, NASA. Map source: Visible Earth-Blue Marble, NASA.

III.1.3. Sampling to study Ba distribution in sediments

Ba distribution profiles were investigated in diverse sediments records from the Eastern Mediterranean, especially in selected cores from the Ocean Drilling Program (ODP) Leg 160 (Emeis et al., 1996). We selected four sites from different paleoceanographic environments (Figure 2, Table 3); and cores from the Ionian basin (Site 964), the Mediterranean Ridge (site 969) and the Erathostenes Seamount (sites 966 and 967) were sampled to separate marine barite. The methodology and sedimentary descriptions are reported in Gallego-Torres et al. (2010) and the enriched Ba intervals were selected accordingly.

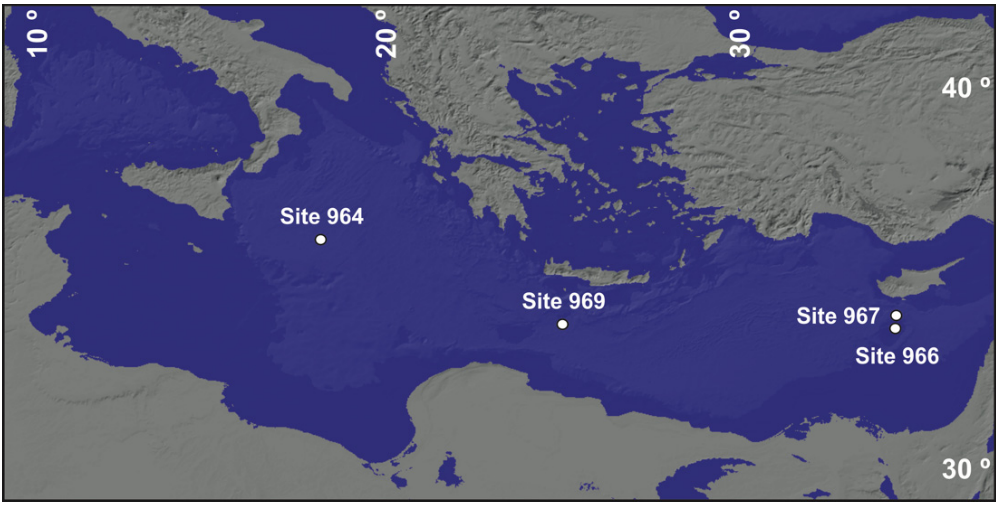

Fig. 2. Location of the ODP Leg 160 sites from which selected cores have been analysed to separate marine barite, map plotted with Global Mapper software.

Table 3. Selected cores/intervals from ODP leg 161 sites studied in this thesis.

Sample	Location	Water depth (m)	Interval (cm)	Age (ky)	Sapropel	I-cycle
969A 1H 03	Mediterranean Ridge	2200	78-80 96-98	120.696 124.944	Pleistocene	12
967D 2H 3A	Erathostenes seamount	2700	80.5-82.5	170.502	Pleistocene	16
966B 1H 03	Erathostenes seamount	926	132.5-134.5 136-138 141-143	170 172 174.28	Pleistocene	16
969D 9H 01	Mediterranean Ridge	2200	30-32	2965	Pliocene	284
964 9H 05	Ionian basin	3660	89-91	2965	Pliocene	284
967A 9H 06	Levantine basin	2700	120-122	2965	Pliocene	284
966D 5H 06	Erathostenes seamount	926	38-40	2965	Pliocene	284

III.2. Bacteria isolation and bacterial culture experiments

III.2.1. Culture media and solutions

The culture medium constitutes an essential nutrient source for microorganism growth under laboratory conditions. An accurate composition depends on the bacterial strain intended to grow, as nutritional needs vary considerably. Culture media for bacteria may be liquid or solid. These nutritive solutions may be prepared with distilled water or tap water, according to the requirements of the bacteria to be cultivated. As solid media, the nutritive solutions are prepared in water and gellified (with a few exceptions) with agar-agar. In preparing the culture media, the first step is to dissolve the medium components in water —which in many cases starts from a commercial preparation with all the dehydrated components— following the manufacturer's instructions. If the media contain a gellified agent (agar-agar), it must to be heated until boiling, stirring occasionally to ensure correct agar fusion and distribution. For liquid media is not necessary to heat, only to stir the mix of solid components within the water. To avoid the growth of contaminants, the next step is to sterilize the culture medium, once its components have dissolved. Depending on whether Petri dishes (for solid media) or liquid media are used, the steps would be as follows:

a) Solid media for use in Petri dishes: the correspondent nutritive solution, with the necessary agar-agar amount added, is prepared in 250 ml Erlenmeyer flasks, 100 ml per flask. In each flask, a magnetic stirrer is used to homogenize the corresponding culture medium once sterilized. Sterilization is carried out in an autoclave at 120 °C for 20 min. Once sterile, the Erlenmeyer flasks are stirred until reaching 50 °C. Then, the gellified culture medium is poured into sterile Petri dishes (20 ml per plate) which stand at room temperature in a laminar flow hood until solidified. The Petri dishes are then stored in a cold chamber at roughly 4 °C until their use.

b) Liquid media: once the components have dissolved, 2-4 ml of the liquid medium is distributed in each test tube, covered with stoppers to sterilize in autoclave. The test tubes containing the sterile culture medium are stored at 4 °C until their use.

The composition of the different culture media and solutions used in this work are detailed below:

- Media for bacterial strain isolation, growth and conservation as work cultures: For marine bacteria isolation and cultivation, “Marine Broth” (MB, DIFCO Laboratories, USA) was used for both liquid and solid culture media. Its composition is described below:

Marine Broth (MB)

Peptone	5 g/l	KBr	0.08 g/l
Yeast extract	1 g/l	$SrCl_2$	34 mg/l
$C_6H_5O_7Fe$	0.1 g/l	H_3BO_3	22 mg/l
NaCl	19.45 g/l	Na_2SiO_3	4 mg/l
$MgCl_2$	5.9 g/l	NaF	2.4 mg/l
Na_2SO_4	3.24 g/l	NH_4NO_3	1.6 mg/l
$CaCl_2$	1.8 g/l	Na_2HPO_4	8 mg/l
KCl	0.55 g/l		
$NaHCO_3$	0.16 g/l		

To prepare liquid MB, suspend 37.4 g of Marine Broth powder in 1 litre of distilled water. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder. Autoclave at 121 °C for 15 minutes. For solid MB add 20 g of Agar (DIFCO) before autoclave. In both cases, pH 6.5 – 7.

- Media for barite precipitation experiments: Barite production assays were conducted in solid culture media enriched with Ba (CMBa) (González-Muñoz et al., 2012) containing no free sulphate to avoid inorganic barite precipitation. We used two CMBa types that differ in the NaCl concentration in order to test the effect of salinity; their composition is detailed below:

Ba-enriched Marine Broth (CMBa1)		Ba-enriched Marine Broth (CMBa2)	
Yeast extract	0.4%	Yeast extract	0.4%
$BaCl_2 \cdot 2H_2O$	2mM	$BaCl_2 \cdot 2H_2O$	2mM
NaCl	1.9%	NaCl	3.5%
Purified Agar	2%	Purified Agar	2%
PH	7	PH	7

The CMBa1 and CMBa2 media preparation is carried out in two steps. First, the $BaCl_2$ solution is prepared and then the yeast extract, NaCl and purified agar are added. Both culture media (CMBa1 and CMBa2) are sterilized and once reached 50 °C they are poured into Petri dishes, 20 ml per plate, and stored at 4 °C until their use.

- Glycerol solutions for bacterial strain conservation during long periods: For conservation of the bacterial strains during long time periods, in conditions ensuring their viability, the procedure was as follows:

a) Cryopreservation tubes are prepared with 0.5 ml of glycerol (99%)/tube and sterilized in autoclave, then stored at 4 °C until their use.

b) Pure cultures of the bacteria to be conserved are prepared by growing the strains in liquid MB medium until roughly half of their exponential growth phase.

c) 0.5 ml from the bacterial culture prepared as described in b) is added in aseptic conditions to a cryoconservation tube prepared as described in a). Then the tubes are stored at -80 °C.

The conservation under these conditions assures the viability of bacteria during long time periods (several years).

- NaCl solutions: Saline solutions were necessary for different purposes, and prepared at two different concentrations of NaCl: 1.9%, the same concentration as in the Marine Broth culture medium, and 3.5%, like seawater. Both solutions, once prepared, were poured in test tubes, 5 ml per tube, which were sterilized in autoclave and stored at 4 °C until their use.

- Solution for marine bacteria fixation: To determine if the filters used for bacterial isolation from seawater samples contained bacterial cells after washing, they were submerged in a special fixative solution for marine bacteria (Morcillo, 2010), with the following composition:

Solution for marine bacteria fixation	
Glutaraldehyde	4 %
Cacodylate buffer	0.2 M
CaCl	0.1 %
Sacarose	0.4 M

This fixative solution is characterized by an osmolarity of 1205 mOsm/l, very similar to seawater, avoiding marine bacteria collapse.

- Cacodylate buffer 0.1 M, to wash filters after bacterial fixation, prepared for High Resolution Scanning Electron Microscopy (HRSEM) observation:

$Na(CH_3)_2\ AsO_3 \cdot 3H_2O$	21.40 g
Distilled water	1000 ml

pH = 7.2 - 7.4

- Osmium tetroxide 1%, for bacteria post-fixation, prepared for HRSEM observation:

Osmium tetroxide	1 g
Cacodylate buffer	0.1 M
Distilled water	100 ml

pH = 7.2 - 7.4

III.2.2. Marine bacteria isolation

Two samples were selected for bacteria isolation, the first from the Balear basin (200 m depth) in the western Mediterranean, the second from the Ionian Sea (250 m depth), both locations being plotted in Figure 1, Chapter VI. From each water sample, 100 ml were aseptically filtered using a 0.45 µm pore size filter (Millipore) in a Kitasato system with a vacuum pump. The filter was recovered with tweezers and gently shaken in 10 ml of NaCl sterile solution (3.5%); it was then washed with the same NaCl solution three times. Afterward, appropriate serial dilutions (until a 10^{-3} dilution) were performed for both samples and several 100 µl aliquots of each dilution were inoculated and spread on MB solid medium and incubated at 28 °C. Bacterial colonies were counted at two time intervals (6 and 20 days). Aiming to (i) isolate the different bacterial strains present in the samples and (ii) to essay their potential capability of enhancing barite precipitation in solid CMBa culture media, was proceed as follows: a selection of colonies —made in view of characteristics such as colour, shape and morphology, considered a priori as belonging to diferent bacteria strains — was isolated in order to be identified. The purity of the selected colonies was confirmed by repeated streaking on solid MB medium and they were afterward regrowth in liquid MB during 48 h at 28 °C for their conservation. These pure bacterial suspensions were stored in glycerol at -80 °C.

III.2.3. Barite production assays

The assays entailed two phases: presumptive tests to detect barite precipitates, and confirmative tests to verify and study the bacterial barite producers and their precipitates. Both phases are described below:

a) *Presumptive test*: *precipitate detection.* The selected bacterial strains were plated by streaking on solid MB and incubated at 28 °C for 48 h. After growth, the colonies were picked up with a sterile toothpick and cultivated on solid CMBa1 and CMBa2 media with the same toothpick drawing several crosses (hence, “the crosses method”). Afterward they were incubated at 28 °C and observed every day with an optical microscope (objectives 4x and 10x) to detect the presence of precipitates.

b) Confirmative test: bacterial barite producers. All the colonies producing precipitates in the presumptive test were isolated in liquid MB and incubated at 28 °C during 24 h. The cultures were centrifuged (15,871 xg for 5 min), washed three times with a NaCl solution (1.9%, the same concentration as for MB) and finally re-suspended in a 5 ml volume of the same NaCl solution. Drops of 20 µl were inoculated on CMBa1 and CMBa2 solid media and incubated at 28 °C. These Petri dishes were observed daily to detect the presence of precipitates until 15 days´ time. Confirmation of barite precipitates was performed by X-ray diffraction (XRD) and HRSEM.

III.2.4. Identification of the isolated bacteria

Genomic DNA was extracted from different bacterial cells in order to construct fingerprint patterns and cluster them in separate groups. The representative strains of each cluster were identified on the basis of their 16S rRNA genes and the sequences compared with those from the GenBank database. Genomic DNA was extracted from bacterial cells using the RealPure Genomic DNA Extraction kit (Durviz, Spain), following the manufacturer's instructions. The quantity of DNA was determined by a Nanodrop spectrophotometer (NanoDrop ND1000, Thermo Fisher Scientific). Additional quality analyses were performed by running an electrophoresis of the extracted DNA (5 µl) on 0.7% agarose gel at 100 V for 40 min, staining with a Gel Red solution (70 µl/l; stock: 10,000 x) for 20 min and visualizing with an UVP documentation system (BioRad Transilluminator, Universal Hood; Mitsubishi P93D-printer).

III.2.4.1. REP-PCR genomic fingerprinting

It consists in amplification of repetitive extragenic palindromic elements (multiple copies of short repetitive sequences) by polymerase chain reaction (PCR) and the use of primers designed to hybridize with repetitive sequences interspersed in the genome. Grouping using the genomic Fingerprint REP-PCR technique was done to choose a representative strain from each group and identify all of them. In this way, a comparison could be made between the barite-producing strains present in the eastern Mediterranean basin and those present in the western Mediterranean basin, and thus determine whether strains of the same genus are present in both basins. To obtain fingerprint patterns of all isolates and cluster them in different groups, rep-PCRs were performed using primers REPIR-I and REP2-I (de Bruijn, 1992), as described in detail in Jroundi et al. (2010). PCR products of all isolates were electrophoresed on 1.5% agarose gel, and the DNA fingerprints were compared manually by visual inspection of the banding patterns.

III.2.4.2. Bacterial identification through 16S rRNA gene

Bacterial 16S rRNA gene sequences were amplified through PCR. This allowed the correlation and establishment of phylogenetic relationships between the isolates. Representative strains of each cluster were identified on the basis of their 16S rDNA amplification using the two opposing primers fD1 and rD1 (Jroundi et al., 2010). PCR products were purified using the Qiagen PCR product purification system, and directly sequenced using the same fD1/rD1 primers on ABI PRISM 3130xI Genetic Analyser. The sequences were compared with those from the GenBank using the BLASTN tool (Altschul et al., 1997) available at EMBL-EBI database (http://www.ebi.ac.uk) and the EzTaxon-e tool that is an extension of the original EzTaxon database (Chun et al., 2007) available at EzBioCloud (http://eztaxon-e.ezbiocloud.net). Phylogenetic analyses were performed utilizing the BioEdit computer program (package version 7.1.11; Hall, 2007), which uses the neighbour-joining algorithm, and visualized with MEGA 4.0.2 (Tamura et al., 2007). The 16S rDNA sequences obtained in this study were deposited in the GenBank database.

III.3. Chemical composition of the water column

III.3.1. On-board analyses

III.3.1.1. Oxygen

The water samples for oxygen analyses were taken from the Niskin bottles using rubber tubes and put into 100 ml volume flasks for “iodine titration”. The flasks were filled until their overflow in order to avoid bubbles, and were immediately capped with rubber stoppers and lids. Afterwards, 1 ml of Mn (II) reagent was added and well mixed in basic conditions to fix the dissolved oxygen present in the sample, resulting in a brown precipitate of manganic hydroxide ($MnO(OH)_2$). The samples were shaken and stored in the dark until their analysis, which was done within hours after its recovery. Prior to analysis, the samples were acidified with sulphuric acid (H_2SO_4) and well mixed, to cause dissolution of the precipitated hydroxides, releasing Mn (III) ions. Later, 1 ml of the reagent sodium iodide/sodium hydroxide (NaI/NaOH) was added and the Mn ions oxidized the added iodine ions (I^-) to iodine (I_2), forming a complex (I_3) that is in equilibrium with iodine. The iodine was titrated with a sodium thiosulfate solution ($Na_2S_2O_3$, 1M) and iodine was reduced to its ion (I-), while the thiosulfate oxidized to tetrathionate ($S_4O_6^{2-}$). As the thiosulfate solution is not stable, a standard solution of potassium iodate (KIO_3, 0.01M) had to be added. The procedure and stoichiometric equations for the reactions are well described by Strickland and Parson (1968), Grasshoff (1983) and Dickson (1994).

III.3.1.2. Nutrients (nitrate, phosphate and silicate)

All samples were collected in high-density 125 ml polyethylene sample bottles after rinsing three times with sample water. The measurements were done by ultraviolet spectroscopy (UV-Vis) in the Oldenburg University facilities (Germany). To measure nitrate, it was first reduced separately in a copperized Cd-coil using imidazole as a buffer, to be measured as nitrite. Diazotization of nitrite with sulphanilamide and N-(1-naphtyl)-ethylene diammonium dichloride to form a reddish-purple dye was measured at a wavelength of 540 nm (Grasshoff, 1983). Phosphate was measured as ortho-phosphate formation of a blue reduced molybdenum phosphate-complex at pH 0.9-1.1. Potassiumantimonyltartrate served as a catalyst and ascorbic acid as a reducing reagent. The colour formed is measured at 880 nm (Murphy and Riley, 1962). Then, dissolved silica was measured as a blue reduced silicate molybdenum-complex at 810 nm. Ascorbic acid served as the reducing reagent, and oxalic acid was used to prevent interference of phosphate (Strickland and Parson, 1968).

III.3.1.3. Dissolved Inorganic Carbon

After acidification to a pH <1, inorganic carbon forms CO_2 in its gaseous phase. This gas was dialyzed over a membrane and the colour of an alkaline phenolphthalein solution was reduced, measured at a wavelength of 550 nm (Stoll et al., 2001) in an UV-Vis spectrophotometer (Oldenburg University, Germany).

III.3.2. Ba profiles

Previous to seawater sampling from the Niskin bottles, a cleaning procedure was followed for Low Density Polyethylene (LDPE) and High Density bottles (dissolved and dissolvable trace elements) as recommended in the GEOTRACES protocols book (Cutter et al., 2017). Accordingly, the bottles were rinsed with methanol or acetone to release oils from their manufacturing. Afterwards, they were soaked at 60 °C for one day in an alkaline detergent and rinsed four times with deionized water. They were then rinsed three times with Milli-Q water under a clean atmosphere and filled with 6M HCl (reagent grade), submerged in 2M HCl (reagent grade) at 60 °C for 2 days, rinsed again four times with Milli-Q water, and finally filled with 1M HCl for at least 2 days. The last step was to rinse with Milli-Q water and ship the bottles empty and double bagged. A 50 µl volume of HNO_3 was added to the samples in order to avoid precipitation and to keep trace elements in solution, as well as for avoiding bacterial growth. The samples were stored at 4 °C until their study. For Ba analyses, we used subsamples of unfiltered oxic water stored in 60 ml ultraclean nalgene bottles and filtered analogues for stations 2, 4, 5, 10 and 11. The use of filtered or unfiltered water samples was chosen in function of the availability and conservation state of the sample at the moment of the analysis. Nevertheless, there are no significant differences because of the modest barite formation during the sampling periods. The samples were analyzed with an ICP-MS photospectrometer (Neptune Plus) at the Oldenburg University facilities, in Germany.

III.4. Study of precipitates and bacterial cells

To study the precipitates, we used two techniques: XRD for crystallographic identification, and electronic microscopy to observe the morphologies.

III.4.1. X-ray diffraction

For XRD analyses, the bacterial cultures from the confirmative tests were collected by cutting the agar portions (2 x 2 mm) containing the bacterial colonies, which were killed by exposition to a germicide UV lamp for 2 h. The portions were cleaned twice with Milli-Q water for 24 h in Falcon tubes to eliminate NaCl and bacterial cells. Then the portions were picked up with sterile tweezers, placed on glass sample holders and dried for 24-48 h at 37 °C. The precipitates were recovered by a spatula and stored in Eppendorf vials.

The obtained precipitates were analyzed by XRD using an X'Pert Pro diffractometer with an X'Celerator detector, from the *Instituto Andaluz de Ciencias de la Tierra* XR Diffraction service. The zero background holder was used, introducing the samples in the diffractometer, where $K\alpha_1$=1.540598 Å and $K\alpha_2$=1.544426 Å, with a Cu anode. The voltage used was 45 kV and a 40 mA current, with a counting time of 10 minutes and 16 seconds, scanning 4 to 69.9°, and a 0.01° step size. For precise mineral identification, the diffractograms were interpreted using X-powder software (http://www.xpowder.com; Martín-Ramos, 2004), a method using the nonlinear minimum square procedures to calculate the unit cell axis of the crystalline phases to determine the exact terms of any isomorphic series.

III.4.2. High Resolution Scanning Electron Microscopy

HRESEM was used for observation of barite precipitates from bacterial cultures, marine barite separated from sediments and bacterial cells retained in filters. Morphological and chemical analyses of the precipitates were performed with an AURIGA model microscope (Carl Zeiss SMT) coupled with an energy-dispersive X-ray microanalysis (EDX) and Electron Backscatter Diffraction (EBSD) of Oxford Instruments. The preparation of each kind of sample is detailed below.

III.4.2.1. Precipitates from bacterial cultures

The samples were prepared by mixing the precipitates from several colonies of each strain, in order to have enough quantity, by (i) cutting the agar containing the dead colonies (after exposition to a germicide UV lamp for 2 h), (ii) immersing the portions of the colonies in Milli-Q water (200 ml), (iii) melting these portions in a microwave oven (600 W for 2-3 minutes), and (iv) centrifugating them (30 seconds at 6,261 xg) to retire the supernatant liquid. The step (iv) of this procedure was repeated from 3 to 4 times to eliminate the remaining culture medium, salts and cellular debris. Precipitates were then transferred to a clean glass plate, dried for 24 h at 37 °C, and then prepared and sputtered with a Hitachi carbon evaporator (CIC, Universidad de Granada) for HRSEM observation.

III.4.2.2. Barite extraction from sediments

Following the leaching procedure of Eagle et al. (2003), barite was separated from diverse intervals representative of sapropels spanning different time periods (Pleistocene and Pliocene). This sequential leaching procedure includes reaction with 6N hydrochloric to remove carbonates and sulfides, sodium hypochlorite to remove organic matter, hydroxylamine to remove Fe-Mn oxyhydroxides and hydroflouric acids to remove silica. The barite crystals recovered following this protocol were deposited in a sample holder and sputtered with carbon for their analysis with HRSEM.

III.4.2.3. Marine bacteria retained in filters

To determine if the filters used for bacterial isolation from seawater samples contained bacterial cells after washing, they were submerged in the fixative solution for marine bacteria previously described. Once fixed (4 h at 4^{o}C), the samples were washed with a 0.1 M cacodylate buffer (x3, 20 min, at 4oC). They were then post-fixed with osmium tetroxide solution (1%) during 1 hour (at 4^{o}C, in darkness). After that they were washed with distilled water (x3, 10 min) and dehydrated in an ethanol gradient (50, 70, 90 and 100%, 10 min in each one, x3). To finish this procedure, each sample was dried by means of the critical point method (Anderson, 1951) with carbon dioxyde in a Polaron CPD 7501 dryer (Quorum Technologies Ltd, UK) and small portions were randomly cut and carbon-coated in the sample holder with a Hitachi evaporator (CIC, Universidad de Granada) prior to analysis with HRSEM.

Chapter IV. The Barium record in Mediterranean deep-sea sediments: role of productivity fluctuations and preservation

IV.1. Introduction

The use of barite ($BaSO_4$) and other Ba proxies for reconstructing paleoproductivity has been broadly demonstrated (Dehairs et al., 1980; Bishop, 1988; Paytan et al., 1996; Paytan and Griffith, 2007; Liguori et al., 2016; Carter et al., 2020). In high productivity regions, marine barite precipitates in the water column, once precipitated, it is exported to the seafloor where is preserved if conditions are oxygenated enough to prevent sulfate reduction (Paytan and Griffith, 2007). Thus, Ba enrichment in marine sediments is broadly used as a record of enhanced productivity, and to date numerous Ba profiles have been reported in diverse basins worldwide. Deserving mention as most exceptional examples of well-preserved Ba records are the Mediterranean basins (Weldeab et al., 2003; Van Beek et al., 2009; Martínez-Ruiz et al., 2000, 2015). In the Eastern Mediterranean basins in particular, the Ba content in sediments mirrors the deposition of sapropels (Van Santvoort et al., 1996; Wehausen and Brumsack, 1999; Martínez-Ruiz et al., 2003; Rohling et al., 2015). These are dark-coloured sediments with TOC concentrations >2% (Kidd et al., 1978), deposited cyclically over the last 13.5 Ma (Krijgsman et al., 1995; Lourens et al., 1996; Schenau et al., 1999; Hilgen et al., 2003; Rohling et al., 2015). In general, the thickness of sapropels varies 2-30 cm (Cramp and O'Sullivan, 1999; Marino et al., 2007; Moller et al., 2012), although in some sites of the Levantine basin (Hieke et al., 1999; Hemleben et al., 2003), the Napoli dome (Cane et al., 2002) and the Aegean Sea (Morigi, 2009) it can reach up to 120 cm. Importantly, post-depositional oxidation may affect the original thickness of sapropel layers, since organic carbon is easily oxidized. In such cases, Ba records evidence the original sapropel deposition, given by the Ba/Al ratio or algorithms as the Ba excess, which is the fraction of Ba in marine sediments that is not of terrigenous origin —it is also referred to as biogenic Ba, bio-Ba or Ba_{xs} (Paytan and Griffith, 2007). Sapropels were numbered in the Mediterranean from the most recent one, S1, to older sapropels (McCoy, 1974), and have also been classified according to insolation cycles (i-cycle). For instance, the sapropel deposited 1872 kyr ago is denoted as corresponding to i-cycle 182 coinciding with this insolation cycle (Hilgen, 1991; Lourens et al., 1996;

Emeis et al., 2000). Sapropels occur both in interglacial and glacial periods (Grant et al., 2016), and the conditions favoring their deposition have been linked to precession-forced summer insolation maxima in the northern hemisphere, which lead to latitudinal migration of the Intertropical Convergence Zone (ITCZ) and higher river runoff as a consequence of the African monsoons´ displacement northward, increasing precipitation over the Nile basin (Hennekam et al., 2014; Weldeab et al., 2014; Rohling et al., 2015; Grant et al., 2016). Besides the increase in river runoff, the deglacial freshwater input from the Atlantic contributed to water column stratification and high nutrient concentrations in surface layers (Grimm et al., 2015; Kotthoff et al., 2008; Spötl et al., 2010; Toucanne et al., 2015). Subsequently, higher nutrient and freshwater inputs increased primary productivity and its associated oxygen variations in bottom waters, resulting in enhanced organic carbon fluxes to the seafloor and seawater stratification allowing organic matter preservation (Emeis et al., 2003; Marino et al., 2007; De Lange et al., 2008; Rohling et al., 2015; Van Helmond et al., 2015). However, the debate about the mechanisms promoting the formation of these organic-rich sediments remains active. It was initially proposed that anoxia was the dominant mechanism leading to organic matter preservation (e.g., Calvert and Pedersen, 1990; 1992), yet the increase of primary productivity as a main forcing mechanism was also claimed (e.g. Bouloubassi et al., 1998; Van Helmond et al., 2015). At present, it is broadly accepted that a combination of both constitutes the important forcing mechanism (e.g., Emeis et al., 2000; Rohling et al., 2015). As increasing rainfall due to intense monsoonal activity led to a decrease in salinity in the surface water masses, the subsequent water stratification resulted in oxygen-depleted deep waters. Therefore, the physiography of the basin affected the total organic carbon, and sapropels deposited in deep basins show the highest TOC values: ranging from 1.2 to 3.4% for Holocene sapropels in the Ionian and the Levantine basins (Murat and Got, 2000; De Lange et al., 2008), up to 18% in Pleistocene sapropels (Murat, 1991), or even 30% in those deposited during the Pliocene (Emeis et al., 1996). In summary, increasing nutrient input resulted in enhanced primary productivity with the subsequent increases in organic matter export to the seafloor and organic matter accumulation (Rohling, 1991; Corselli et al., 2002). In many cases, the higher productivity also led to a bloom of N-fixing bacteria, which in turn gave rise to greater oxygen consumption, hence reduced oxygenation leading to greater organic matter preservation (Gallego-Torres et al., 2011; Bristow et al., 2017; Jickells et al., 2017; Bale et al., 2019). The fresh water input derived from the African monsoons activated fossil river/wadi systems along the North-African margin and small rivers around the Aegean and Adriatic Seas (e.g., Filippidi and De Lange, 2019), delivering larger nutrient inputs to the Eastern Mediterranean basin. Still, the major contribution was from the Nile, which was particularly high in the Levantine Basin (Hennekam et al., 2014; Ehrmann et al., 2016; Wu et al., 2016; Tesi et al., 2017; Zwiep et al., 2018). All these factors resulted in significant spatial variability for TOC content and also Ba content. Indeed, there is great variability throughout time. For example, compared to the most recent sapropel S1, the deposition of older sapropels S3 and S5 is characterized by stronger freshwater fluxes due to a more intense African monsoon, giving rise to the development of dense vegetation cover and therefore decreased erosion. Thus, Nile river runoff was higher during S1 deposition as compared to S5, although the influence of the Aegean and Adriatic riverine sediment input during sapropel S5 appears to be much higher because of precipitation seasonality (Wu et al., 2018). Global sea level was also variable throughout sapropel deposition, being lower during S3 than during S1 and S5 (Zwiep et al., 2018), the latter deposited in a fast sea level rise related to deglaciation phases (Capozzi and Negri, 2009). Such changes likewise influenced sedimentation rates that subsequently affected marine barite abundance in sapropels. In general, Ba contents increase with depth of the basins (Dymond et al., 1992, 1996; Von Breymann et al., 1992; Weldeab et al., 2003), reflecting the importance of physiography and basin hydrodynamics. Furthermore, sapropels show higher Ba content in sediments deposited in lower sedimentation rate settings, as enhanced sedimentation rates dilute the Ba content (Krom et al., 1991; Francois et al., 1995; Mercone et al., 2001; Moutin and Raimbault, 2002; Thingstad et al., 2005; Gallego-Torres et al., 2007; De Lange et al., 2008; Rohling et al., 2015). At this time, the Sahara dust contribution was negligible because of the development of dense vegetation during humid periods (Rohling et al., 2002, 2004, 2006; Hennekam et al., 2014; Grant et al., 2016; Wu et al., 2016, 2018; Zwiep et al., 2018).

Variations in paleoclimate and paleoceanographic conditions may have also been an important factor in the western Mediterranean basin, where sapropels are not recognized; though in the Alboran Sea basin organic rich-sediments with TOC content up to 1.05% mass have been recognized (Murat, 1999; Cacho et al., 2002; Martínez-Ruiz et al., 2000; Jiménez-Espejo et al., 2007; Rodrigo-Gámiz et al., 2011) —the so-called Organic-Rich Layers (ORLs). They have also been related to changes in oxygenation and productivity.

Within the context of the aforementioned spatial and temporal differences, this work aims to investigate sapropel depositional variability and to characterize marine barite distribution in the Mediterranean Sea, comparing barite and Ba records from different basins and time periods.

IV.1.1. Oceanographic setting

The Mediterranean Sea is subdivided into the western and the Eastern Mediterranean basins, which are in turn compartmentalized as diverse sub-basins (from West to East: the Alboran basin, South Balearic, Tyrrhenian, Ionian, Adriatic, Cretan, Mediterranean Ridge, Aegean, Levantine and Erathostenes Seamount). On the whole, the Mediterranean Sea has proven to be an exceptional natural laboratory for paleoenvironmental research, because its semi-enclosed nature and restricted water circulation (Roether et al., 1996; Pinardi and Masetti, 2000) makes this basin particularly sensitive to the effects of global changes (Durrieu de Madron et al., 2011; Lionello, 2012; Picotti et al., 2014; Martínez-Ruiz et al., 2015; Cortina et al., 2018). The current overall Mediterranean Sea circulation comprises relatively lighter surface Atlantic water passing through the Gibraltar Strait and travelling eastward, becoming a saltier and denser water mass, which sinks in the Levantine basin to undo its pathway westward as a deeper water body that flows out to the Atlantic. The surface Atlantic inflowing waters (0-200 m depth) travel eastward, reaching their highest salinity up to ~39 psu and temperatures up to 28 °C in the Levantine basin (Millot and Taupier-Letage, 2005; Pinardi et al., 2019). In the easternmost part of the basin, the cold winter winds provoke intense evaporation that causes surface water sinking by density, becoming Levantine Intermediate Water (LIW). The LIW does the reverse at intermediate depths (200-600 m) with a lower temperature of ~15.5 °C (Lascaratos et al., 1999; Millot and Taupier-Letage, 2005). The Mediterranean Deep Waters (MDW) are denser water masses, mainly formed in the southern Adriatic in the eastern basin (Eastern Mediterranean Deep Water, EMDW), and in the Gulf of Lion in the western (Western Mediterranean Deep Water, WMDW), with a mean temperature of ~13 °C (Roether et al. 2007; Tsimplis et al., 2008). The deep waters travel westward and the outflow is mixed with LIW, passing by the Strait of Gibraltar as Mediterranean Outflow Water (MOW) (Kinder and Parrilla, 1987; Sparnocchia et al., 1999; Astraldi et al., 2002). Variations in the MOW strength may play a significant role in the Atlantic Meridional Overturning Circulation (AMOC) through the outflow of more saline Mediterranean waters into the intermediate North Atlantic (Rogerson et al., 2006; Voelker et al., 2006). In addition, a reduced MOW leads to a density increase of the Atlantic waters at 1000 m and below, since the water mass replacing MOW is denser; this weakens the deep AMOC and leads to a significant warming of the Nordic Seas and surrounding areas through a subpolar gyre displacement eastward (Rahmstorf, 1998; Voelker et al., 2006; Swingedouw et al., 2019). In this framework, sapropels are a piece of the puzzle in global paleoclimate reconstructions and during their deposition, the decrease in deep and intermediate water formation may weaken the MOW and consequently affect the AMOC (Bahr et al., 2015; Voelker et al., 2006; Penaud et al., 2011).

IV.1.2. The Mediterranean sapropel record

As mentioned, oxygen depletion in deep waters and higher productivity are thought to be the major forcing mechanisms for sapropel deposition (Rohling et al., 2015; Grant et al., 2016; Amies et al., 2019; Wu et al., 2018; Zwiep et al., 2018). The intervals of sapropels deposition typically last between ~3 and ~8 kyr (Rohling et al., 2015; Grant et al., 2016) and they are recognized at different water depths. The shallowest sapropels are usually found ~300-400 m below the present sea level (Rohling and Gieskes, 1989; Murat and Got, 2000; Rohling et al., 2015), although in smaller basins such as the Aegean Sea the most recent sapropel S1 was found at 80-110 m water depth (Perissoratis and Piper, 1992; Siddall et al., 2003; Stanford et al., 2011). In terms of time scales, going from past to recent sapropel layers, old Pliocene sapropels are very rich in C_{org}, up to 30% (Emeis et al., 1996; Passier et al., 1999; Nijenhuis and De Lange, 2000; Waning and Brumsack, 2000), while Plio-Pleistocene sapropels show a

TOC-decreasing trend towards the present, from concentrations up to 18% to less than 2% (Calvert, 1983; Murat, 1991, 1999; Gallego-Torres et al., 2010). The most recent sapropels show lower TOC contents, for instance S5 has ~8% (Rohling et al., 2006; Gallego-Torres et al., 2007; Grant et al., 2016), and sapropel S1 was deposited during the last post-glacial rebound between ~6 and 10.8 kyr BP (De Lange et al., 2008; Hennekam et al., 2014; Tesi et al., 2017). Although it was proposed that S1 deposition was favored by anoxic conditions during the period of insolation maximum (Rossignol-Strick et al., 1982; Rossignol-Strick, 1985; Rohling et al., 2015; Tesi et al., 2017), recent studies point out that S1 —at least for shallower water columns— might also have been deposited during water renewal at intermediate depths (Tachikawa et al., 2015; Filippidi and De Lange, 2019). The deglaciation during S1 deposition led to a subsequent sea level rise, increasing the Atlantic-Mediterranean water exchange and decreasing salinity and the residence time of the Mediterranean waters (Rohling et al., 2015). The density decrease in surface waters interrupted deep-water formation and favored stagnation. It has been suggested that the significance of the sea level rise was greater for S1 than the monsoon effect, because compared to older sapropels, such as S5, the insolation maximum during its deposition was weaker (de Noblet et al., 1996; Grimm et al., 2015), and also the duration of the S1 deposition (4.8 kyr) is shorter than that of S5 (6.8 kyr) (Grant et al., 2016). In contrast, S5 is one of the best developed Quaternary sapropels. The buoyancy changes driven by monsoonal river runoff were significantly higher during S5 than during S1, and the effect of the sea level rise by deglaciation was lower compared to sapropel S1. Even though the Nile river inputs increased during S5 deposition, it may be that its contribution was limited during the deposition of this sapropel because of dense vegetation covering the Ethiopian Highlands (Hennekam et al., 2014; Wu et al., 2018). However, minor rivers surrounding the Aegean and Adriatic basin brought larger amounts of sediments into the Eastern Mediterranean (Wu et al., 2018; Amies et al., 2019), much higher during sapropel S5 than during S1. As already discussed, other than the enrichment in organic carbon, the Mediterranean sapropel record is essentially characterized by a significant Ba enrichment. Although only part of the precipitated barite accumulates in the underlying sediments (Dymond et al., 1992; Paytan et al., 1996), particulate Ba preservation in the Eastern Mediterranean basin is exceptional, which may be related and possibly due to sulfate saturation in pore waters derived from Miocene saline deposits formed during the Messinian salinity crisis (De Lange et al., 1990). This situation would have allowed barite to be preserved, being less affected by diagenetic dissolution (Nijenhuis and De lange, 2000; Filippidi and De Lange, 2019; Passier et al., 1999; Martínez-Ruiz et al., 2000). TOC content in sapropels mainly reflects input, preservation and dilution (Schwartzkopf, 1993; Tyson, 1995, 2001). In this regard, higher linear sedimentation rates (LSR) are expected to result in increasing TOC contents, because organic carbon would be buried fast, reducing exposure time and mitigating degradation effects, while low LSR would result in decreasing organic matter preservation, due to its oxidation (Heath et al., 1977; Müller and Suess, 1979; Coleman et al., 1979; Betts and Holland, 1991; Collins et al., 1995; Hartnett et al., 1998; Mayer, 1995). Nonetheless, high LSR bear a clastic dilution effect on TOC accumulation (Ibach, 1982; Stein, 1986; Canfield, 1989; Tyson, 1995, 2001, 2005; Schoepfer et al., 2015). It is moreover important to distinguish between LSR (sediment thickness per unit time, cm/kyr) and mass accumulation rates (MARs, mass deposited during a certain time on a unit area, g/cm^2/kyr), since sediments undergo compaction and LSR are not accurate enough for quantitative studies. MARs are calculated from LSR and dry bulk density (g/cm^3 of dry sediment per wet volume). An accurate and precise age model is critical for the calculation of MARs. In the Mediterranean Sea the LSRs increase westward, with an average of 3 cm/kyr in the Eastern Mediterranean (Cita et al., 1977; Thunell et al., 1977; De Lange et al., 2008; Gallego-Torres et al., 2010) and between 4.4 and 15 cm/kyr in the western basins (Grazzini, 1975; Masqué et al., 2003; Jiménez-Espejo et al., 2006; Pinardi et al., 2006; Zuñiga et al., 2007; Nieto-Moreno, 2012; Rodrigo-Gámiz, 2012). The highest values for MARs are found near the shoreline and nearby slopes. This is a particular important aspect regarding the Ba concentrations in sapropels, as they could be diluted by high sedimentation rates (Murat and Got, 2000). Even in relatively closely spaced cores, the MARs could be very different, which should be taken into account for calculating barite accumulation rates. The highest Ba_{bio}-MAR values during the Holocene are found in the Levantine basin (site 967), whereas for the Plio-Pleistocene sapropels they are found in the Ionian basin (site 964), especially during S5 deposition (see Figure 3). In general, there is temporal variability from the Pliocene to the Holocene sapropels, with a decreasing trend of Ba content towards the present with the exception of sapropel S5, with the highest concentrations. The Ba record is prominent within sapropels S1, S5 and S7, but it fluctuates in S6, which shows lower values, particularly in the Ionian basin and the Mediterranean Ridge. This may be related to higher sedimentation rates during S6 deposition (Castradori, 1993; Emeis et al., 2003). The lowest Ba_{bio}-MARs are recognized in the shallow sites (Dymond et al., 1992, 1996; Von Breymann et al., 1992; Weldeab et al., 2003). This is in agreement with the observed MARs both in the Holocene and Pleistocene sapropels (Table 4), even when sites are closely located, such as sites 966 (926 m depth) and 967 (2555 m depth).

IV.1.3. Mediterranean Organic-Rich Layers

During ODP Leg 161, dark sediments with an organic carbon content of <2% recognized in the western Mediterranean (Comas et al., 1996) were defined as ORLs. The relationship between eastern sapropels and ORLs in the western basins is still unclear. For the most recent ones, S1 and the last ORL, there is no correlation in age. The most recent ORL was deposited earlier than S1 in the Alboran Sea basin, between 14.35 – 8.9 kyr, during deglaciation and the early Holocene (Cacho et al., 2002; Martínez-Ruiz et al., 2015). These organic-rich sediments are thought to derive from reduced bottom circulation by changes in the buoyancy of the surface layer coupled with a reduction in the aspiration of the WMDW (Rogerson et al., 2008; Grant et al., 2016; Perez-Asensio et al., 2020). Such changes occurred during the Heinrich Event 1, including a decreasing trend in the seawater surface temperatures (Reguera, 2004) that affected surface water buoyancy and reduced the westward flux. Dense waters originated in the Gulf of Lion were injected at intermediate depth more often than at bottom depth, causing water stratification, with the subsequent isolation of the deep water mass, hence reinforcing the low oxygen conditions (Frigola et al., 2008; Rogerson et al., 2008). Oxygen depletion in the deep western basin is considered independent of the eastern basin (Rogerson et al., 2008; Rohling et al., 2015). The thickness of ORLs varies from a few to about 30 cm (Cramp and O'Sullivan, 1999; Marino et al., 2007; Moller et al., 2012), and these sediments do not show a significant Ba enrichment compared to sapropels, in some cases because of shallow depths. For instance, at site ODP 976 the shallow water column and high sedimentation rates led to low Ba content in the ORLs. Something similar occurred at 966 (Table 4), located in the Erathostenes Seamount (926 m. depth), where the Ba_{bio}-MARs are the lowest of the eastern basin because of the shallow water column. In general, similarly to organic carbon, low Ba contents are related to a shallow bottom likely due to shorter duration for barite to precipitate and dissolve in the water column (Singh et al., 2020), and/or to high sedimentation rates diluting Ba (Fagel et al., 2002), which reinforces the importance of the geological setting in the preservation of Ba records.

IV.2. Results

Diverse intervals were selected for the study as representative examples to analyze Ba content in Mediterranean sapropels (Figs. 1 and 2, Tables 1 to 4). These intervals correspond to cores recovered during ODP-Leg 160. From west to east, the drilling sites from this Leg are located in the Ionian basin (site 964), the Mediterranean Ridge (site 969), the Erathostenes Seamount (site 966) and the Levantine basin (967). Site 964 is the deepest (3658 mbsl), located in the Ionian basin and affected by the marine currents flowing through the Sicily Strait. Site 969 (2200 mbsl) constitutes the centermost location of the eastern basin, whereas site 966 (926 mbsl) is located in a pelagic high, and site 967 (2555 mbsl) is influenced by the detrital Nile River inputs. Sites 964 and 969 comprise Holocene to Pliocene sapropels; sites 966 and 967 record Holocene to Pleistocene sapropels.

Table 1- Ba concentrations (ppm) and Ba/Al ratios in Pliocene to Holocene sediments from different Mediterranean Sea sub-basins.

	Alboran basin	South Balearic basin	Tyrrhenian basin	Ionian basin	Adriatic basin
Ba (ppm)	226-680	159-324	21-631	167-2732	14-336
Ba/Al	[1] 0.0020	[3] 0.0055		[3] 0.0130	[5] 0.0058
	[2] 0.0033			[3] 0.0220	
	[3] 0.0034			[5] 0.0140	
	[4] 0.0037			[7] 0.0180	

	Cretan basin	Mediterranean Ridge	Aegean basin	Levantine basin	Erathostenes seamount	n
Ba (ppm)	143-3500	130-4500	168-677	187-700	127-1260	75
Ba/Al	[1] 0.0050	[1] 0.0040	[1] 0.0050	[1] 0.0020	[1] 0.0030	
	[1] 0.0060	[5] 0.0100	[5] 0.0250	[5] 0.0050	[5] 0.0010	
	[5] 0.0029	[7] 0.0310			[6] 0.0019	
					[7] 0.0060	
					[7] 0.0200	

References: Cita et al., 1977; Calvert, 1983; Robertson, 1990; Leoni et al., 1991; Pruysers et al., 1991; Van Os et al., 1991; Thomsom et al., 1995; Van SantVoort et al., 1996; Dolenec et al., 1998; Wehausen and Brumsack, 1998; Martínez-Ruiz et al., 2000, 2003; Warning and Brumsack, 2000; Böttcher et al., 2003; Weldeab et al., 2003; Arnaboldi et al., 2007; Gallego-Torres et al., 2011; Rodrigo-Gámiz et al., 2011; Rozic et al., 2011; Saccà et al., 2011; Karditsa et al., 2014; Tanaskovski et al., 2014; Möbius et al., 2015; this work. n= number of total cores. [1] Weldeab et al., 2003; [2] Moreno et al., 2004; Sánchez-Vidal et al., 2005; [3] Martínez-Ruíz et al., 2003; [4] Reitz et al., 2004; [5] Mercone et al., 2000; [6] Van Os et al., 1994; Wehausen et al., 1998; [7] this work.

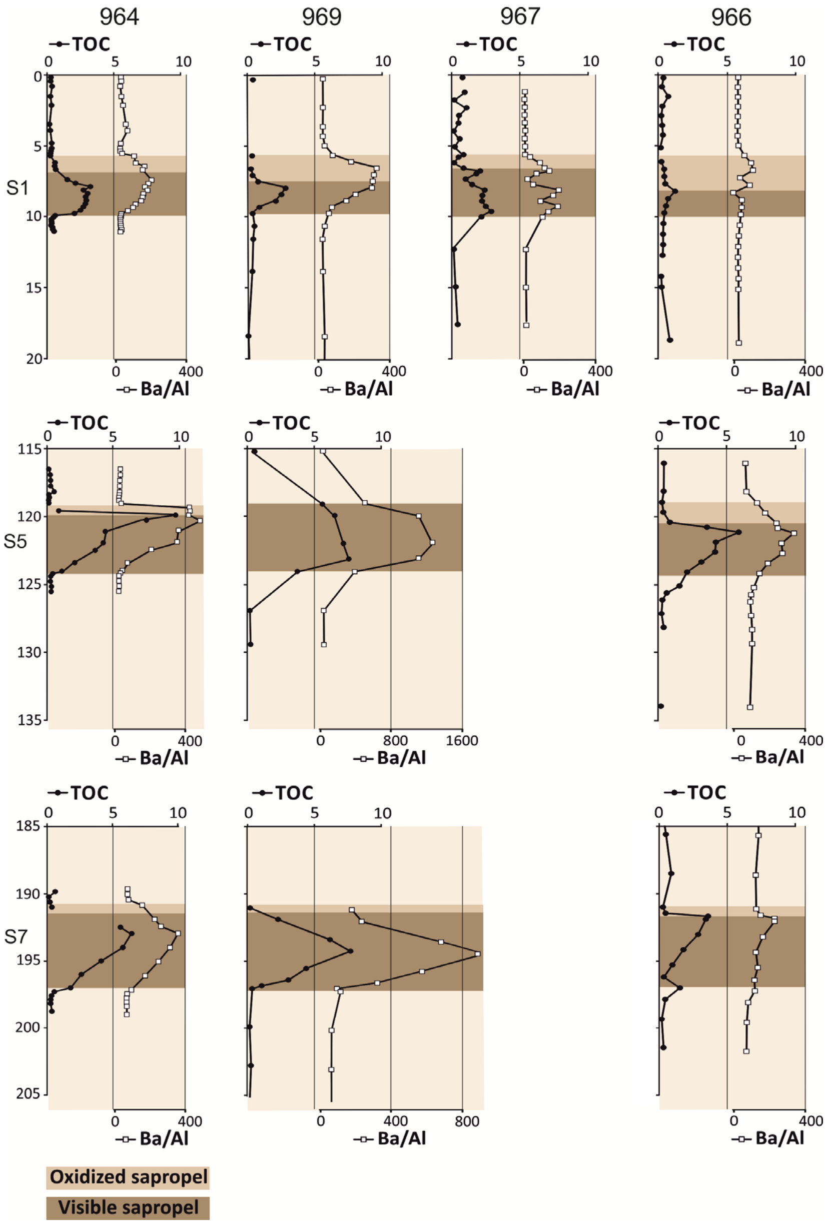

Fig. 1. Total organic carbon (TOC) (%) and Ba/Al ratio profiles in the selected intervals analyzed in this thesis. Age (ky) in vertical scale (modified from Gallego-Torres et al., 2010). Dark brown shading corresponds to visible sapropel, based on TOC concentration and dark colored sediments. Light brown shading represents the oxidized sapropel intervals, determined by the offset between Ba/Al and TOC increases.

Selected Plio-Pleistocene sapropels are shown in Table 5. As mentioned in Chapter III, barite crystals were extracted following a leaching procedure (Paytan et al., 1996; Eagle et al., 2003). Afterwards, the sample was washed and the insoluble residue was observed under HRSEM to determine barite morphologies and chemical composition. In all the analyzed samples the crystals showed the typical barite morphologies alike those seen in sediment trap samples (Dehairs et al., 1980; Bishop, 1988; Paytan et al., 2002), the ovoid, spherical and sub-euhedral shapes, and sizes <5 μm (Figure 3), which support its formation in the water column (Dehairs et al., 1980; Bishop, 1988; Paytan et al., 2002; Griffith and Paytan, 2012; Martínez-Ruiz et al., 2019). Indeed, it has been demonstrated that Pliocene and Pleistocene barite crystals extracted from the same study sites as in this work (ODP Sites 964, 966 and 967) were authigenic (Paytan et al., 2004).

Table 2- Ba concentration (ppm) in Mediterranean sapropels and ORL’s.

	Age (ky)	i-cycle	Alboran basin	South Balearic basin	Tyrrhen. basin	Ionian basin	Cretan basin	Medit. Ridge	Levant. basin	Erathost. seamount	**n**
S1	8	2	390-680			300-1200	362-1100	855-1269		317-1003	16
S3	81	8								960	1
S4	102	10					362	1400		818	3
S5	124	12				548-2732	362-3500	3406-4500	700	1260	10
S6	172	16				429-548	362-2600	561-2500	500	594-885	11
S7	195	18				548-2122		2250		761	5
S8	217	20				548-1100				721	4
Pleistoc.*				210-324	522-611	1735-1889	856-1269	856-3305	1261-1273	563-1072	10
Pliocene*						179				270	2

References: Pruysers et al., 1991; Van Os et al., 1991; Thomsom et al., 1995; Van SantVoort et al., 1996; Martínez-Ruiz et al., 2000, 2003; Weldeab et al., 2003; Arnaboldi et al., 2007; Gallego-Torres et al., 2011; Möbius et al., 2015; this work. n= number of total cores. *Calvert, 1983; Cita et al., 1977; Arnaboldi et al., 2007; **Wehausen and Brumsack, 1998.

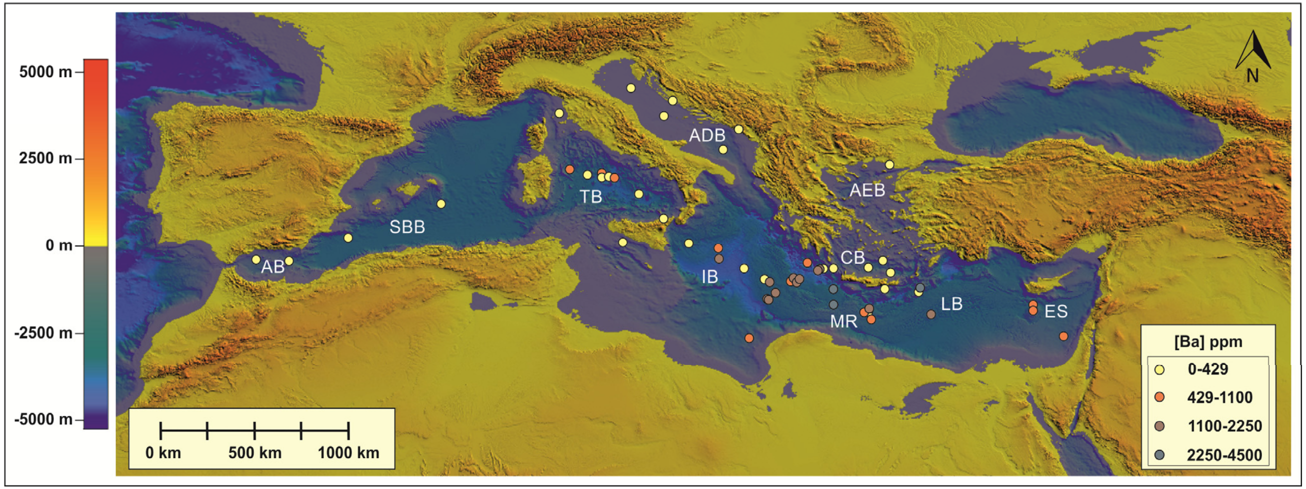

Fig. 2. Average barium concentrations in organic-rich layers (ORLs) and sapropels from the Mediterranean Sea. References: Ryan et al., 1973; Cita et al., 1977; Calvert, 1983; Robertson, 1990; Van Os et al., 1991; Leoni et al., 1991; Pruysers et al., 1991; Thomson et al., 1995; Van Santvoort et al., 1996; Dolenec et al., 1998; Wehausen and Brumsack, 1998; Martínez-Ruiz et al., 2000, 2003; Passier et al., 1999; Böttcher et al., 2003; Weldeab et al., 2003; Arnaboldi et al., 2007; Gallego-Torres et al., 2010, 2011; Rodrigo-Gámiz et al., 2011; Rožič et al., 2012; Saccà et al., 2011; Karditsa et al., 2014; Tanaskovski et al., 2014; Möbius et al., 2015; this work. **AB:** Alboran basin, **SBB:** South Balearic basin, **TB:** Thyrrenian basin, **IB:** Ionian basin, **ADB:** Adriatic basin, **CB:** Cretan basin, **MR:** Mediterranean Ridge, **AEB:** Aegean basin, **LB:** Levantine basin, **ES:** Erathostenes seamount.

Table 3- Ba/Al ratios in Mediterranean sapropels and ORL's.

	Ba/Al						
	Alboran basin	South Balearic basin	Ionian basin	Cretan basin	Mediterranean Ridge	Levantine basin	Erathostenes seamount
S1	[3, 4, 5, 6] 0.0020-0.0037	[3] 0.0055	[1, 2, 3] 0.013-0.022	[2, 4] 0.029-0.060	[1, 2, 4] 0.010-0.031	[2, 4] 0.002-0.005	[1, 2, 7] 0.001-0.020
S3							[1] 0.004-0.017
S4					[1] 0.044		[1] 0.015
S5			[1] 0.043		[1] 0.127		[1] 0.029
S6			[1] 0.009		[1] 0.018		[1] 0.015-0.022
S7			[1] 0.034		[1] 0.093		[1] 0.018
S8			[1] 0.021				[1] 0.018
i-c 152					[1] 0.062		
i-c 156					[1] 0.093		
i-c 160					[1] 0.045		
i-c 170							[1] 0.017
i-c 172							[1] 0.014
i-c 178							[1] 0.013
i-c 180							[1] 0.021
i-c 182							[1] 0.018
i-c 272			[1] 0.064				
i-c 280					[1] 0.030		
i-c 282			[1] 0.033				
i-c 284			[1] 0.024		[1] 0.028		
i-c 286			[1] 0.048				

[1] This work; [2] Mercone et al., 2000; [3] Martínez-Ruíz et al., 2003; [4] Weldeab et al., 2003; [5] Moreno et al., 2004; Sánchez-Vidal et al., 2005; [6] Reitz et al., 2004; [7] Van Os et al., 1994; Wehausen et al., 1998.

Table 4- Ba concentration (ppm) and Ba$_{bio}$-MAR's from the studied sapropel intervals (Gallego-Torres et al., 2010).

Site	Region	Water depth m	Sapropel	Age ky BP	Sed. rate cm/ky	Ba ppm	Ba/Al $\cdot 10^{-4}$	MAR $\cdot 10^{3}$ mg/cm^2/ky	Ba$_{bio}$-MAR mg/cm^2/ky
964	Ionian basin	3658	S1	8	4.14	864	184.4	5.14	4.44
			S5	124	5.93	2732	432.9	6.99	19.09
			S6	172	6	429	86.3	8.06	3.46
			S7	195	4.04	2122	337.0	5.23	11.11
			S8	217	3.02	1100	207.6	4.30	4.73
			i-c 272	2828	2.55	3094	644.0	4.36	13.47
			i-c 282	2943	2.41	1335	326.7	3.30	4.41
			i-c 284	2965	2.87	1083	238.4	3.71	4.02
			i-c 286	2989	2.50	1893	484.7	3.20	6.05
969	Med. Ridge	2200	S1	8	4.39	1051	307.8	5.54	5.82
			S4	102	4.36	1400	438.5	5.17	7.24
			S5	124	4.24	3406	1265.5	4.81	16.39
			S6	172	2.62	561	182.6	3.25	1.830
			S7	195	2.64	2250	934.2	2.98	6.717
			i-c 152	1564	2.80	1951	616.9	3.37	6.57
			i-c 156	1603	2.63	1319	927.8	3.25	4.282
			i-c 160	1642	2.75	1467	452.1	3.37	4.94
			i-c 280	2921	1.88	1366	296.8	2.14	2.93
			i-c 284	2965	2.37	1259	282.0	2.93	3.68
967	Levantine basin	2555	S1	8	5.36	1003	204.3	7.60	7.62
			S3	81	5.64	960	172.4	7.77	7.46
			S4	102	5.9	818	154.2	8.76	7.17
			S6	172	4.67	885	216.7	6.93	6.13
			i-c 170	1736	2.20	883	177.4	3.01	2.65
			i-c 172	1757	2.58	596	139.1	3.93	2.34
			i-c 178	1829	2.42	639	133.1	3.57	2.27
			i-c 180	1851	2.30	1022	205.7	3.22	3.28
			i-c 182	1872	2.18	1146	182.4	3.03	3.47
966	Erath. seamount	926	S1	8	4	317	64.0	5.09	1.62
			S3	81	2.6	266	44.6	3.76	1.00
			S5	124	2.35	1260	291.9	2.86	3.61
			S6	172	2.19	594	150.6	2.88	1.71
			S7	195	2.24	761	182.2	2.93	2.23

Table 5. Sapropel intervals selected for barite separation.

Core	Section (cm)	Location	Age
969A 1H 03	78-80, 96-98	Mediterranean Ridge	Pleistocene
969D 9H 01	30-32	Mediterranean Ridge	Pliocene
964 9H 05	89-91	Ionian basin	Pliocene
967D 2H 3A	80.5-82.5	Levantine basin	Pleistocene
967A 9H 06	120-122	Levantine basin	Pliocene
966D 5H 06	38-40	Erathostenes seamount	Pliocene
966B 1H 03	132.5-143	Erathostenes seamount	Pleistocene

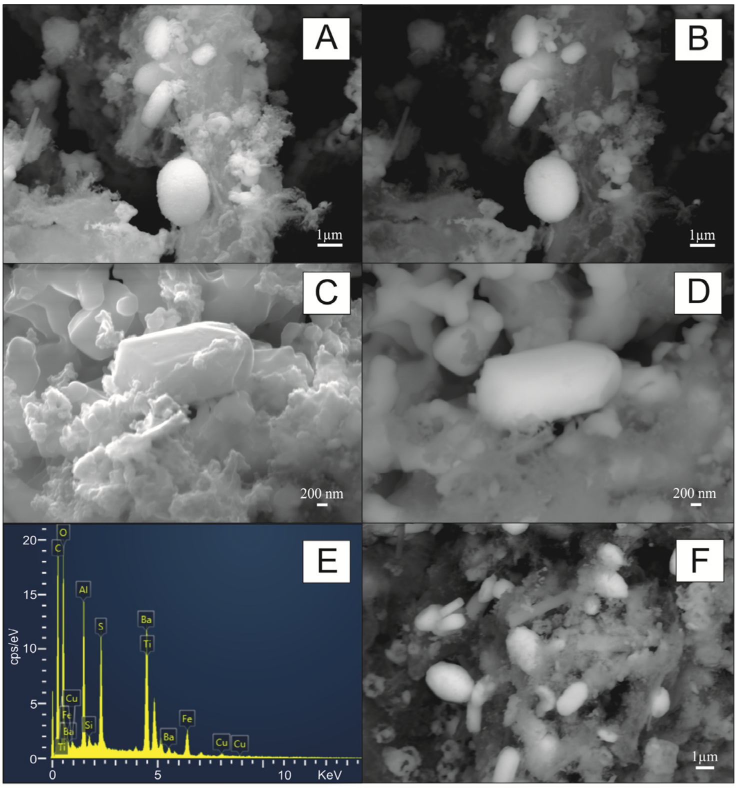

Fig. 3. Scanning electron micrographs showing barite crystals extracted by the leaching procedure (Eagle and Paytan, 2003) from core intervals from sites 966 and 969. Photographs were obtained under two different SEM observation modes: Secondary (left) and backscattered electron mode (right). A and B: Barite from sample 969D 9H 01 (see table 3 from Chapter III). C, D and F: Barite from sample 966D 5H 06 (see Table 3 from Chapter III). E: X-ray spectroscopy spectra from a large barite crystal representative of the chemical composition of the most barite crystals present in the samples.

IV.3. Discussion

Spatial Ba distribution in marine sediments

Understanding the Ba distribution in marine sediments and hence the productivity variations linked to climate variability in the Mediterranean Sea is essential to shed further light on the impact of global climate changes in this region. Although many proxies have been used for climate reconstructions in the Mediterranean (e.g., Martínez-Ruiz et al., 2015 and references therein), barium proxies stand as a reliable tool for productivity reconstructions in the sedimentary record when sufficiently oxygenated conditions allow for barite preservation (Rutsch et al., 1995; Paytan et al., 1996; McManus et al., 1999). Ba_{excess} (total barium minus the fraction associated with terrigenous material) usually includes other phases (carbonates, opal, oxides and ferromanganese oxyhydroxides); but to study productivity in Mediterranean Sea sediments, we focused on the proxy Ba_{bio}, as it can be considered practically equivalent to barite precipitated in the water column and accumulated in sediments underlying areas of high productivity (Eagle et al., 2003), which are precisely the characteristics of the studied sediments. In the Mediterranean Sea, Ba spatial variability —both in the water column and in sediments— reflects marine barite distribution and therefore productivity. Although Ba records have been commonly used for reconstructing productivity in the Eastern Mediterranean (Ehrmann et al., 2016; Wu et al., 2016; Tesi et al., 2017; Zwiep et al., 2018), it should be taken into account that sedimentation rates can affect Ba accumulation, as this element can undergo significant dilution. Indeed, near the Tyrrhenian coastline and in the shallow Adriatic sub-basins, high sedimentation rates have resulted in low Ba content; whereas in deeper sites with lower sedimentation rates, higher Ba contents are found. Coeval sapropels have shown that water depth also affects Ba_{bio}-MARs, as occurring in some sites having a nearby location, such as site 966 in the Erathostenes Seamount (a pelagic high with a water column of 900 m depth) and site 967 (2700 m water depth) with Ba_{bio}-MARs of 1.6161 mg/cm^2/kyr and 7.6214 mg/cm^2/ky, respectively, during S1 deposition. Schenau et al. (2001) made the same observation in surface sediments of the Arabian Sea, showing a linear Ba increase in deeper basins, which can be extrapolated to carbon concentrations (Murat and Got, 2000). Thus, the strong correlation of Ba_{bio}-MARs and water column depth suggests enhanced barite formation at deeper sites and its subsequent accumulation in sediments. This should also be taken into account when comparing coeval sediments deposited at different water depths. In general, physiography and water depth are also important factors conditioning Ba spatial distribution. Bottom-current directions and velocities can also indirectly be affected by bottom morphology, such as slope gradient changes, bathymetric highs, eddies and gyres interacting with the water masses, which would in turn affect the eutrophic zone isotherms punctually increasing nutrients and therefore primary production in oligotrophic areas (Owens and Hogg, 1980; Candelario et al. 2008; Schroeder et al., 2012). In summary, detrital mass accumulation rates, water depth and the physiography of the basin would locally affect the Ba concentration in sediments (Goldberg and Arrhenius, 1958; Schmitz, 1987; Dymond et al., 1992). Despite the effects of these factors, the Ba distribution in the Mediterranean Sea sediments (Figure 3) correlates well with the TOC, showing an eastward increase, and accordingly the highest Ba concentrations in the eastern basin. This reflects the primary productivity, whose variations are mainly due to the Nile river contribution in the eastern Mediterranean (Hennekam et al., 2014; Van Helmond et al., 2015; Wu et al., 2016), which delivered vast amounts of nutrients to the oriental basin (which are related to high evaporation in the surface water mass). Data complied here show that the spatial variability of Ba_{bio}-MAR (Table 4) —especially during the Holocene— follows the regular trend of productivity in the Mediterranean Sea, increasing eastward, except at the Erathostenes Seamount (site 966) because of the shallow water column. During the Pleistocene this trend is also observable from the Ionian (site 964) to the Levantine (site 967) basin, but it is not so clear at the Mediterranean Ridge (site 969), where the water depth difference with respect to sites 964 and 967 cannot account for the huge difference in Ba_{bio} content (which is much lower in site 969). This could point to spatial gradients of nutrient availability and surface water productivity in the Eastern Mediterranean at the times of S5, S6 and S7 deposition, as reported for sapropel S5 by Weldeab et al. (2003). Indeed, during S5 deposition, seafloor anoxia developed with different timings at different locations, reflecting spatial differences in organic carbon fluxes to the seafloor, signaling spatial export productivity gradients and/or spatial ecosystem differences (Casford et al., 2003; Weldeab et al., 2003; Bianchi et al., 2006; Rohling et al., 2006; Moller et al., 2012). The differences in nutrients and therefore spatial variability of productivity were partly explained by Rohling and Gieskes (1989), who proposed the shoaling of the pycnocline to the lower part of the photic layer during S5. River inputs could explain the moderate productivity increase during S5 at some sites (Wehausen and Brumsack, 1999), although for areas with less riverine influence further mechanisms of nutrient delivery into the photic zone would be required. While in south Crete high primary productivity was recorded, in the southeast and southwest areas the upward nutrient mixing was likely moderated by weak wind mixing (Weldeab et al., 2003). Furthermore, Moller et al. (2012) proposed that the eastern Mediterranean is generally more sensitive to development of deep-sea anoxia than the western basin because of differences in the efficiency of deep water renewal. The highest values for Ba_{bio}-MARs during the Holocene are found at the Levantine basin (site 967), whereas during deposition of S5 and S7 in the Pleistocene the highest Ba_{bio}-MARs are located at the Ionian basin (site 964) and the Mediterranean Ridge (site 969).

Temporal Ba distribution: changes in productivity and preservation

At present, the Mediterranean Sea is poor in nutrients, especially in nitrogen and phosphorus. This lack of nutrients, along with other factors —mainly salinity and temperature triggering density changes and water masses stratification— gave rise to an oligotrophic basin in the western and an ultra-oligotrophic one in the eastern Mediterranean (Krom et al., 2003, 2010; Thingstad et al., 2005; Zohary et al., 2005; Krom et al., 2010; Pujo-Pay et al., 2011). Primary production decreased eastward from 232 to 20 gC/m^2 yr (Siokou-Frangou et al., 2010; Yogev et al., 2011). The highest values of primary productivity are found in the western basin, varying from 86 to 232 gC/m^2 yr (Marty and Chiaverini, 2002); meanwhile, in the eastern basin they vary from 20 to 80 gC/m^2 yr, the lowest values being located at the Levantine Basin (Psarra et al., 2005; Siokou-Frangou et al., 2010; Yogev et al., 2011). The nitrate to phosphate ratio (28:1) in the EMDW does not follow the Redfield ratio (16:1) for the world ocean (Krom et al., 1991). In the eastern basin there is very little organic matter production and oxygen is not completely consumed, so denitrifying bacteria cannot act and the excess of nitrate to phosphate remains (Krom et al., 2016). Yet the Mediterranean Sea´s situation has significantly varied over time. Past frameworks entail periods of high productivity, from Miocene to Holocene sediments. Although sapropel formation is controlled by cyclic changes in productivity and preservation (Higgins et al., 2010), the degree of importance of each one has been debated or is still under debate. During the Miocene and Pliocene, high productivity and preservation were apparently of equal importance (Menzel et al., 2003; Gallego-Torres et al., 2011; Vasiliev et al., 2019). In the Pliocene and Early Pleistocene, when deep-water circulation in the eastern basin was slower, the restricted water ventilation enhanced sapropel preservation along with high productivity (Menzel et al., 2003; Gallego-Torres et al., 2011). In contrast, Late Pleistocene and Holocene sapropels show greater productivity than preservation (De Lange et al., 2008; Rohling et al., 2015; Van Helmond et al., 2015; Zwiep et al., 2018). Overall, Mediterranean Sea sediments draw a downward organic matter trend to the present owing to lesser productivity, and the Ba content within sapropels evolves along the same line because of its strong link with productivity. The S6 is a Pleistocene exception, showing the lowest values of Ba_{bio}-MARs (Table 4); the underlying reasons are a moderate increase in productivity, likewise reported for S8 (Emeis et al., 2003), and higher sedimentation rates diluting Ba, also observed for TOC (Gallego-Torres et al., 2010). In the Eastern Mediterranean, such unfavorable conditions for S6 and S8 deposition during glacial periods implied a cold, arid climate with a low sea level, alternating with slightly warmer climatic shifts (Triantaphyllou et al., 2009). The contrast with S5 and S7 is remarkable: they show the highest Ba accumulations (Table 4). This suggests that the differences in conditions for sapropel deposition between glacial (S6, S8) and interglacial (S5, S7) periods are more important than previously thought.

Barite from sapropels

Barite extracted from the studied sapropel intervals reveals the usual sizes and morphologies of authigenic marine barite precipitated in the water column (Figure 3). Furthermore, $BaSO_4$ extracted from Mediterranean Holocene sapropels has been described as authigenic (Martínez-Ruiz et al., 2000), its origin confirmed by the isotopic S signature corresponding to seawater (Paytan et al., 2004). This is concordant with the fact that marine barite precipitation occurs mainly in the upper part (~200 – 1000 m) of the water column, where organic aggregates exist and decompose (Goldberg and Arrhenius, 1958; Bishop, 1988; Bernstein et al., 1992; Ganeshram et al., 2003; Paytan and Griffith, 2007; Griffith and Paytan, 2012; Carter et al., 2020). Bacteria are the most abundant microorganisms involved in organic matter decomposition, colonizing sinking particles that are converted into small substrates available for cellular metabolism (Arnosti, 2011). Thus, there is a coupling between sinking particles and free living bacteria in the mesopelagic (Cho and Azam, 1988; Smith et al., 1992) and bathypelagic layers (Nagata et al., 2000); the same happens with marine barite, linked to a higher bacterial activity in the mesopelagic zone (Dehairs et al., 2008; Jacquet et al., 2011; Planchon et al., 2013) and therefore to organic matter decay.

Under nutrient limitations —that is, the Mediterranean Sea situation— the labile carbon from phytoplankton production may be first respired for bacterial basal metabolism (Carvalho et al., 2017). In this basin, bacterial respiration on average represents 62% of the total community respiration, a value slightly higher than in open oceans, according to the general oligotrophic nature of the Mediterranean Sea (González-Benitez et al., 2019). Such a percentage means that bacterial activity is very substantial and therefore any variation in its role decomposing organic matter in the water column may affect barite formation. In fact, changes in productivity throughout the Mediterranean Sea´s history are reflected in sapropel deposition and barite content in these sediments (Martínez-Ruiz et al., 2000; Gallego-Torres et al., 2011; Rohling et al., 2015; Zwiep et al., 2018). Moreover, barite in not altered during post-depositional oxidation, remaining in the sediments for detection of the actual layer thickness, while organic matter may have been oxidized (Thomson et al., 1995; Van Santvoort et al., 1997; Paytan et al., 1996; Thomson et al., 2006). As mentioned, the good preservation of barite in the Mediterranean sapropel layers can be linked to sulphate saturation in pore waters, which may be promoted by a high saline background in the basin originated by Messinian deposits (De Lange et al., 1990).

IV.4. Conclusions

We have analyzed and compared Ba data reported for Mediterranean Sea sediments, finding extensive evidence of spatial productivity gradients as well as major temporal variability in barite accumulation. In general, sapropel formation occurs during high productivity episodes associated with monsoon maxima, during which strong water mass stratification may also led to deep water oxygen depletion. However, overlying variability —imposed by spatial differences, depth and sedimentation rates within each sapropel event— distinctly show that actual sapropel deposition during such events is also ruled by a dynamic system constrained by interactions between physiography, seawater circulation, detrital fluxes and biogeochemical processes that also influenced organic matter accumulation. Thereupon, the key to unveil the exact mechanisms behind sapropel deposition at each particular region also lies in further studies, needed to correlate Ba data from Mediterranean Sea sediments and the water column, to arrive at a better understanding of the regional and temporal variability. In the next chapter we therefore analyze and discuss the spatial Ba distribution in the Mediterranean Sea water column.

Chapter V. Barium concentration in the water column along a Mediterranean Sea transect

This chapter will be extedended with further information and submitted for publication in an international journal in collaboration with Prof. Gert de Lange, who was the chief scientist of the cruises providing the dissolved Ba data included in this thesis (1999 “Smilable” with RV Logachev; 2010-2011 “Ristretto e Lungo” with RV Meteor; 2000 “Pasap”, 2008 “Doppio”, 2009 “Macchiato” and 2012 “Cortado” with RV Pelagia. Respective codes: SL, RL, PS, DP, MP, CP).

V.1. Introduction

In the water column, barium shows a nutrient-like profile reflecting Ba biogenic uptake for barite precipitation ($BaSO_4$) in surface waters while Ba release through partial dissolution of this mineral in the deep layers (Chan et al., 1976; Dehairs et al., 1980; 1991; Collier and Edmond, 1984; Von Breymann et al., 1992; Hoppema et al., 2010). Although most modern oceans are rich in sulphates (~28 mM at salinity of 35 psu), they are in general undersaturated with respect to barium, showing concentrations that usually range 60-140 nM in deep waters to 30-60 nM in surface waters, with the exception of areas near river inputs or with intense upwelling increasing Ba concentration (Chan et al., 1976; Elderfield, et al., 1996; Jeandel et al., 1996, 2000; Monnin et al., 1999; Jacquet et al., 2005). The Ba removal in surface waters is relatively slow compared to its supply, besides this element is progressively concentrated with aging of deep water masses during their pathway from the North Atlantic to the Pacific (Rushdi et al., 2000; Jacquet at al., 2005; Sternberg et al., 2005). The Atlantic shows Ba concentrations ranging from 4 µg/l in surface waters to roughly 18 µg/l in the deep layer, whilst the Pacific ranges from 8 to 31 µg/l (Wolgemuth and Broecker, 1970; Broecker et al., 1976; Bernat et al., 1972; Chan et al., 1976; Sugiyama et al., 1984; Horner et al., 2015; Bates et al., 2017; Le Roy et al., 2018). The Southern Ocean and the Indian Ocean are almost in the same range than the Atlantic, with maximum barium concentrations up to 16 µg/l at depth (Jeandel et al., 1996; Jacquet et al., 2004, 2007; Singh et al 2013; Pyle et al., 2018). Despite of the Ba undersaturation in seawater, marine barite precipitates in the mesopelagic zone where organic matter decomposition occur (Goldberg and Arrhenius, 1958; Church and Wolgemuth, 1972; Bishop, 1988; Ganeshram et al., 2003; Dehairs et al., 2008; Jacquet et al., 2011; Griffith and Paytan, 2012; Planchon et al., 2013). For decades scientists have been trying to unveil which are the mechanisms of marine barite precipitation under subsaturated conditions and they first attributed to some microorganisms (e.g., Acantharia) the Ba release, although their abundance was not enough to explain barite content in sediments nor in seawater (Fresnel et al., 1979; Gooday and Nott, 1982; Bernstein et al., 1992; Bertram and Cowen, 1997). In the last years bacteria have arisen as promising microorganisms for the Ba contribution (González-Muñoz et al., 2012; Martinez-Ruiz et al., 2018), but the biotic barite precipitation mechanisms are still partially understood. On the other hand, there are several geochemical sources contributing to the Ba input in seawater: rivers, dust, hydrothermal systems, cold seeps, submarine groundwater discharge (SGD), desorption from remineralization processes and diffusion from sediments (Elderfield and Schultz, 1996; Monnin et al., 2001; Hsieh et al., 2017; Bridgestock et al., 2018). Rivers provide large Ba inputs to the oceans with an average global flux from 2 to 6.14 nmol/cm^2 yr (Von Damm et al., 1985; Elderfield et al., 1996; Dickens et al., 2003; Das et al., 2006). Although with a modest contribution, submarine groundwater discharge has been recently estimated to contribute with up to the 25% of the river input, about 0.46 nmol/cm^2 yr (Mayfield et al., 2021). In North America, on the southern shore of Cape Cod, it has been demonstrated that SGD is a major source of dissolved Ba to the ocean (Charette et al., 2015). Nevertheless, the concentration of dissolved Ba in rivers and groundwater varies and their influence decreases away from the coast, this is reflected in spatial variations in seawater (Dehairs et al., 1987; Jeandel et al., 1996; Moore, 1997; Jacquet et al., 2005, 2007; Guay et al., 2009; Santos et al., 2011; Mayfield et al., 2021). Indeed, near coastal areas the Ba concentrations reach up to 28 μg/l, being higher than in the pelagic water column (Carter et al., 2020 and references therein). In areas of low productivity, the SGD can be also a source of iron, affecting to adsorption of dissolved Ba into metal oxides that sink in association with organic debris (Sajih et al., 2014; Grissom, 2015). Then, barium will be desorbed enhancing an enrichment of the dissolved phase near the oxygen minimum zone. Regarding the Ba contribution from cold seeps and hydrothermal systems, it has been calculated about 1.4 to 5.6 μg/l

(Edmond et al., 1979; Von Damm et al., 1985; McQuay et al., 2008), about the 2%-13% of the total Ba input to the ocean. Nevertheless, there are many uncertainties both for SGD, hydrothermal systems and cold seeps since they are not easily detectable and it is difficult to quantify the total contribution of these systems. Moreover, the precipitation of barite mounds and chimneys in cold seeps and hydrothermal systems consume most the dissolved Ba expelled and their fluxes may have been overestimated. In regard to the Ba input from dust lithogenic fluxes, some researchers argue that an increase of this input because of human activities could be significant (Guerzoni and Chester, 1996), whilst other authors have determined that the inputs of particulate barium from this source are despicable (Piela et al., 2012). Taking the global ocean as a whole, it is difficult to evaluate and quantify all sources as compared to the restricted ocean basin setting of the Mediterranean, whose seawater masses are very well defined. The major lithogenic contributions to the Mediterranean Sea are traditionally attributed to rivers and dust. In the eastern basin, the Nile River is the most relevant (Weldeab et al., 2014). However, although rivers seem to be important sources there are few studies focusing on their total contribution to the western Mediterranean basin (Roy-Barman et al., 2019). In turn, river sources contribute only 3.5% of the atmospheric input (Chou and Wollast, 1997), and their input can be considered negligible in view of estuarine removal processes (Maring and Duce, 1987). The dust input was also suggested as a substantial Ba source into the Mediterranean Sea (Hydes et al., 1988) but its contribution to the dissolved Ba in the Mediterranean Sea was overestimated by Dehairs et al. (1987) and has been demonstrated as negligible by Weldeab et al. (2014). Dehairs et al. (1987) also concluded that the internal cycling of Ba is of minor importance if compared to the fluxes throughout the system and Jacquet et al. (2016) confirmed this by studying a high resolution transect of dissolved Ba in the Mediterranean Sea and conclude that the dissolved Ba distribution is mainly driven by hydrodynamics. These authors pointed to high dissolved Ba contents in deep waters as a reflection of particulate Ba dissolution in undersaturated waters, enhanced by the residence time and low ventilation of deep waters. Additionally, the hydrothermal Ba fluxes may be substantial, as well as the sediment-related input source arriving to the deep water; but as previously mentioned these fluxes are difficult to estimate. Along these lines, in this chapter we provide high resolution barium profiles from the water column through a Mediterranean Sea transect (see Figure 1 of Chapter III), also compared with those from literature. Despite research efforts, Ba fluxes are still fairly unknown and it is crucial to evaluate in more detail the processes driving Ba fluxes in order to constrain the model and to understand how global variations affect to the Ba cycle and marine barite accumulation.

V.1.1. Oceanographic setting

The Atlantic inflowing water travels through the Mediterranean Sea in a surface layer (0-200 m depth) with salinity increasing eastward from 35 to ~39 psu, and a temperature of 15-28 °C (Marullo et al., 1999; Millot and Taupier, 2005; Rohling et al., 2009). This Modified Atlantic Water (MAW) passes through the Sicily Strait, although part of it remains in the western basin to contribute to the deep water formation. The flux of MAW passing the Sicily Strait is estimated in 1-3 Sv (1 Sv = 10^6 m^3/s. Millot, 1999). When the MAW reaches the Levantine basin, during winter, under cold and dry air conditions, a decreasing surface water temperature generates denser water that sinks and turns into Levantine Intermediate Water (LIW) near the Rhodes gyre. The mean annual production of LIW is of 1 Sv (Ovchinnikov, 1984; Lascaratos, 1993; Tziperman and Speer, 1994; Lascaratos and Nittis, 1998). The LIW spreads westward at intermediate water depths (200-600 m depth) with a temperature of ~15.5 °C (Millot, 2013 and references therein). In the western basin, the LIW continues as Mediterranean Intermediate Water (MIW), being slowly admixed with adjacent watermasses (Emelianov et al., 2006). Meanwhile, the Mediterranean Deep Water (MDW) is formed by denser water with salinity up to 39 psu and temperature of 12.7 to 13.6 °C. The MDW formation depends on its location in the Mediterranean Sea, so that: a) the Eastern Mediterranean Deep Water (EMDW) is usually formed the southern Adriatic Sea (0.1-0.4 Sv) but sometimes its formation has been located in the Aegean Sea (Roether et al. 1996, 1998, 2007; Lascaratos et al., 1999; Tsimplis et al., 2008); whereas b) the Western Mediterranean Deep Water (WMDW) comes from the Gulf of Lion (Leaman and Schott, 1991). The volume of WMDW varies widely depending on the method used in its estimation (0.4.3 Sv). Finally, a mixture of MIW and WMDW, known as Mediterranean Outflow Water (MOW), flows through the

Gibraltar Strait (Kinder and Parilla, 1987; Sparnocchia et al., 1999; Astraldi et al., 2002). A general scheme of the water masses circulation in the Mediterranean Sea is modeled in Figure 1. Regarding volumes of deep (DW, 600-1500 m), intermediate (IW, 200-600 m) and surface (SW, 0-200 m) water masses of the eastern and western Mediterranean basins, they can be easily derived from the often cited data in Bethoux and Gentili (1999), obtaining a reasonable aproximation. The water masses derived from the model are: SW_{mass}= 1.6 x 10^{17} kg; IW_{mass}= 3.3 x 10^{17} kg; DW_{mass}= 7.85 x 10^{17} kg for the western Mediterranean Sea and SW_{mass}= 3.65 x 10^{17} kg; IW_{mass}= 6.2 x 10^{17} kg; DW_{mass}= 14.4 x 10^{17} kg for the eastern Mediterranean Sea. From these data we can calculate volumes by multiplying by the seawater density, thus for the western basin we obtain volumes of 155 x 10^{12} m^3 (SW), 320 x 10^{12} m^3 (IW) and 760 x 10^{12} m^3 (DW), whereas in the eastern basin these values are of 355 x 10^{12} m^3 (SW), 603 x 10^{12} m^3 (IW) and 1400 x 10^{12} m^3 (DW). The volumes of surface and intermediate waters in the western basin fit well with those from Powley et al. (2016). Compared with oceans, the residence time of the Mediterranean water masses is relatively short overall. The MAW remains between 2.2 to 10 years (Chou and Wollast, 1997; Roether et al., 1998; Migon, 2005; Van Cappellen et al., 2014), while the LIW residence time varies between 8.7 and 25 years (Ovchinnikov, 1983; Stratford and Williams, 1997; Roether et al., 1998, Van Cappellen et al., 2014). Times for the WMDW are reportedly from 2 to 30 years (Bethoux and Copin-Montegut, 1984; Bethoux et al., 2002; Schroeder et al., 2008), and even up to 80 years according to the Bèthoux model. However, our water samples are more coherent with a residence time of 30 years for the WMDW since they were recovered during a post-Eastern Mediterranean Transient (EMT) period. The EMT was a hydrodynamic event caused by a combination of high-salinity waters intruding into the Aegean Sea between 1991 and 1993, in a context of particularly strong winters (Roether et al, 2014). The deep-water formation rates during the EMT were therefore higher, about 1.0-3.0 x 10^6 m^3/s rather than 0.3 x 10^6 m^3/s (Roether and Schlitzer, 1991; Lascaratos, 1993; Béranger et al., 2009; Theocharis, 2009). For the EMDW, a residence time between 50 and 150 years has been put forth (Bethoux and Copin-Montegut, 1984; Schlitzer et al., 1991; POEM-Group, 1992; Roether and Well, 2001; Bethoux et al., 2002; Theocharis, 2009), but again, here we should adopt a coherent number. Consequently, a post-EMT residence time of ~100 yr for the EMDW seems more realistic for our sampling periods.

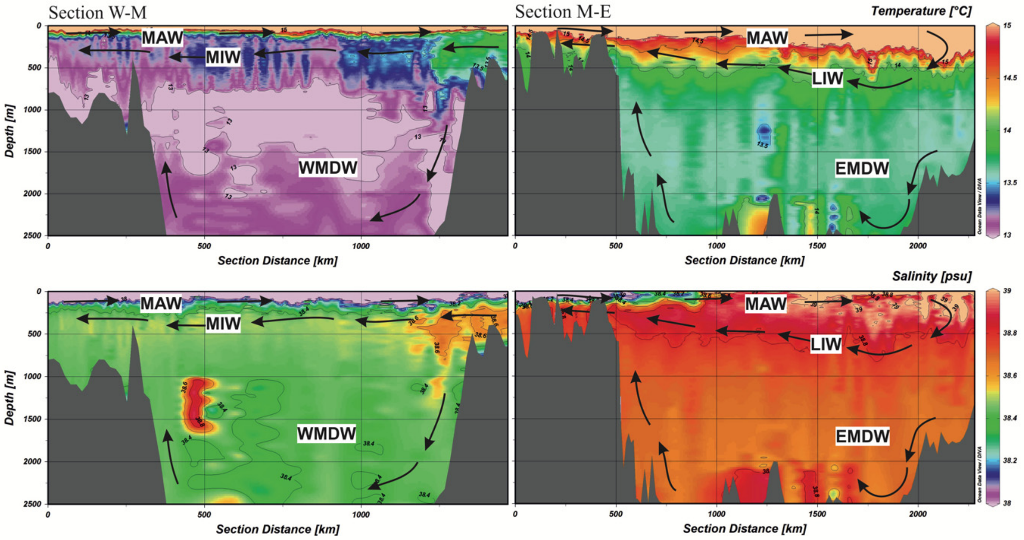

Fig. 1. Modelling of water mass circulation along the studied Mediterranean Sea transect (see Fig.1 Chapter III). Data from MedatlasII database (Fichaut et al., 2003. MEDAR group) and World Ocean Database (Boyer et al., 2018) plotted together using the Ocean Data View software (Schiltzer, 2013). The acronyms MAW, LIW, MIW, WMDW and EMDW correspond to Modified Atlantic Water, Levantine Intermediate Water, Mediterranean Intermediate Water, Western Mediterranean Deep Water and Eastern Mediterranean Deep Water, respectively.

In the Mediterranean Sea there are potential mixing zones that can slightly modify the typical temperatures and salinity of the waters bodies in some regions due to an intensive water mixture underway, i.e. the Atlantic inflow in the western basin and the Sicily Channel. The obtained salinity and temperature profiles (Figure 2) are consistent with the physical and chemical characteristics of the inflowing and eastward-moving Atlantic surface waters (MAW), relatively warm, as well as those of the westward-flowing bodies of intermediate and deep waters (LIW, MIW and MDW), which are denser and therefore colder and saltier. The anti-estuarine circulation of the Mediterranean Sea relies in the excess of evaporation over precipitation and river inputs. This renders in a water budget deficit that enhance the input of Atlantic water to balance the sea level and also it increase the density in the eastern basin forcing the vertical mixing that leads to deep water formation, which increase the pressure that in turn leads to outflow the deep water masses to the western Mediterranean Sea and then to the Atlantic (Sánchez-Román, 2008; Soto-Navarro, 2012). About 0.78 Sv (1 Sv = 10^6 m^3/s) of MOW passes thought the Gibraltar Strait to the Atlantic Ocean whereas the overlying Atlantic waters inflow is about 0.83 Sv (Powley et al., 2016). In general, the Mediterranean watermass budgets seem to be in balance, although some uncertainties remain for the Sicily Strait due to the lack of data in this region. Recently, García-Solsona et al. (2020) used rare-earth elements and Nd isotopes as water masses tracers and concluded that the contribution of deep water from the eastern Mediterranean passing through the Sicily channel to the western basin could reach 70% of the deep waters in the Tyrrhenian Sea, a value higher than previously calculated. In view of the water balance a net-transfer of water across the Sicily Channel from west to east of 0.02 Sv would be needed. An important number of researchers have tried to assess the water budget in the Mediterranean Sea with the available data and models (Bethoux, 1979; Peixoto et al., 1982; Bryden and Kinder, 1991; Harzallah et al., 1993; Gilman and Garrett, 1994; Castellari et al., 1998; Angelucci et al., 1998; Bethoux and Gentili, 1999; Boukthir and Barnier, 2000; Mariotti et al., 2002; Soto-Navarro et al., 2010), but their approaches are always shaded by many uncertainties depending on the methodology, sources of the datasets and temporal variability. As example, the studies of river inputs provided a mean annual flux between 8.1-16 x 10^3 m^3/s for the whole Mediterranean (Tixeront, 1970; Ovchinnikov, 1974; Margat, 1992; Boukthir and Barnier, 2000; Struglia et al., 2004). This has been related to the construction of dams in recent decades as well as the increase in the fluvial catchments for irrigation. Several authors have also mentioned the potential underestimation of river fluxes since not all rivers are included in some estimates. In addition, subterranean estuaries are generally overlooked, but they may contribute up to 50% of the river waterflux, and for dissolved species this represents an amount possibly equal to that transported by rivers. For the Black Sea, several numbers have also been proposed for the mean net inflow, from 5.8 x10^3 m^3/s to 9.6 x 10^3 m^3/s (Tolmazin, 1985; Unluata et al., 1990; Beşiktepe et al., 1994, Bethoux and Gentili, 1999; Karnaska and Maderich, 2008). Despite certain differences, the high evaporation in the Mediterranean basins has been estimated by several authors e.g., using the salt-balance at Gibraltar, or derived from long time series of satellite evaporation/precipitation observations, or from direct precipitation rates summarized per country (e.g., Bryden and Kinder, 1991; Mariotti et al., 2002). The values mostly fall in the range 0.035 +/- 0.05 Sv. These estimates have provided the most consistent picture for the average water budget over a few decades, which proves important for future calculations of detrital fluxes and barium budgets.

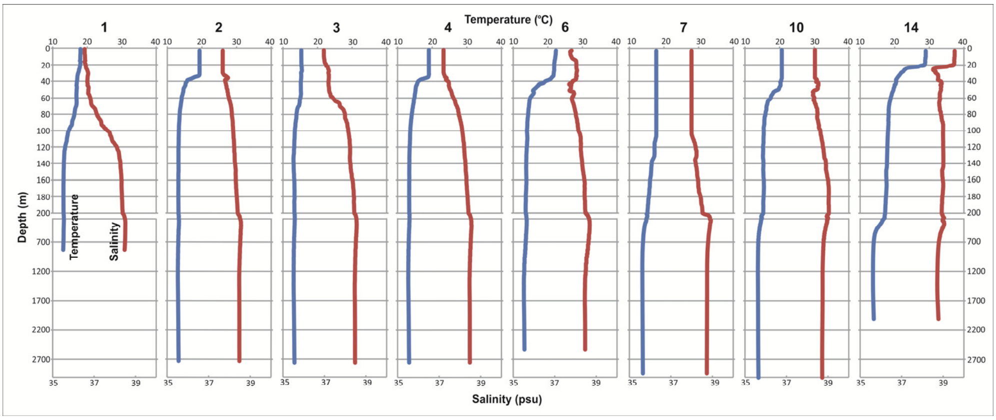

Fig. 2. Salinity and temperature profiles of the stations plotted in map from Figure 1, Chapter III, located in the studied Mediterranean Sea transect.

Large-scale patterns of atmospheric variability exert a potential influence on the Mediterranean Sea. This is the case of one of the most important climate variability indicators, the North Atlantic Oscilation (NAO), which according to Mariotti et al. (2002) appears to be closely related to decadal and interdecadal variability of the Mediterranean winter moisture deficit. The NAO is a fluctuating climatic phenomenon in the North Atlantic Ocean resulting from atmospheric pressure differences at sea level between the Icelandic Low and the Azores High (Hurrell and Deser, 2009). In particular, the increase in Mediterranean water deficit from the 1970s to the 1990s corresponds to a switch from a low to high NAO index. This climatic phenomenon has a major influence on precipitations and temperatures in the Mediterranean Sea basin (Hurrell 1995; Rodwell et al., 1999; Mariotti et al., 2002) and therefore affects evaporation and river runoff (Tsimplis and Josey, 2001; Struglia et al., 2004; Milly et al., 2005), although the latter also has an increasingly significant anthropogenic component (enhanced irrigation, reduced run-off). The excess evaporation and consequential Mediterranean Water Deficit (MWD) must, of course, be on average similar to the water balance at Gibraltar. Thus, on inter-annual to inter-decadal timescales, during the period 1948 – 98, the Atlantic water influx through Gibraltar must have adjusted to compensate for the enhanced Mediterranean water deficit. Initially, precipitation would have been mainly responsible for this —no significant correlation with the NAO is found for evaporation. Even so, variability in Mediterranean excess evaporation has been also favored by variations in other climate patterns. Josey et al. (2011) concluded that the NAO has minor impact on the winter mean heat budgets, whereas the East Atlantic pattern (EA), less known but with an effect five times greater than NAO in the whole Mediterranean, bears a strong impact especially evident in the western basin (Josey and Marsh, 2005; Josey et al., 2011). The negative EA pattern influence upon the Mediterranean Sea is associated with cold and dry air from the northeast increasing the sea-atmosphere temperature and moisture gradients, leading to a greater heat loss. The positive phase of the East Atlantic/West Russian pattern (EA/WR) moreover leads to higher latent heat losses in the Eastern Mediterranean, since it enhances northerlies over the eastern basin and southerlies over the western one (Krichak and Alpert, 2005; Josey et al., 2011). Some authors have suggested the existence of its own pattern in the Mediterranean Sea between the eastern and the western basins, the Mediterranean Oscillation (MO, Conte et al., 1989). This index of local character could have a more specific influence in the basin processes (Supić et al., 2004; Gomis et al., 2006), although it remains poorly studied. The MO is an oscillation of sea level pressure anomalies in the Central and Western Mediterranean that correlates well with NAO in winter but it seems that the MO influence in the Mediterranean is more noticeable than the NAO in all seasons (Criado-Aldeanueva and Soto-Navarro, 2020) and its negative phase is linked to increased rainfall and intensified evaporation in the Levantine basin. In the whole, there are three patterns that could influence the ocean-atmosphere heat exchanges during winter in the Mediterranean Sea: the EA, the EA/WR and the MO patterns. The NAO plays a secondary role. The warming and heightened net evaporation over the Mediterranean Sea surface in recent decades was predicted through atmospheric models based on re-analysis of observational data (Mariotti et al., 2008, 2015; Somot et al., 2008). In adittion, an increasing salinity trend has been predicted for the whole basin (Macias et al., 2015; Adloff et al., 2015; Maier et al., 2019), which could dramatically affect the overturning circulation of the Mediterranean Sea.

V.1.2. Ventilation and primary productivity (nutrients)

Besides productivity, for predicting the response of oceans to climate change it is also necessary to understand the temporal and spatial variability of nutrients distribution and ocean ventilation. Ocean ventilation is linked to heat transport, salinity, dissolved gases and nutrients (Luyten et al., 1983; Khatiwala et al., 2012). Although the Mediterranean Sea is well ventilated in comparison to the global ocean because of the relatively fast deep water formation and the short renewal timescale of the water masses (Li and Tanhua, 2020), a slowly ventilated water body is found between 1200 m and 2600 m depth. As mentioned in section V.1.1 (see references there), the residence time of the water masses in the Mediterranean Sea have been estimated in and average of 17 years for the LIW, 100 yr for the EMDW and 16 years for the WMDW (Bethoux and Copin-Montegut, 1984; Bethoux et al., 2002; Schroeder et al., 2008). Such ventilation ages affect to the nutrients content and dissolved oxygen in the different Mediterranean basins. The present Mediterranean is oligotrophic due to a lack of nutrients, especially of inorganic phosphorus and nitrogen, which is the main limitation for primary production (e.g., Krom et al., 1991, 2010; Thingstad et al., 2005). Because of this limitation the oxygen input by deep-water formation may exceed the deep oxygen demand, which could explain why the obtained profile of average dissolved O_2 (Figure 4) in the intermediate and deep waters shows a slight increase eastward. Although the Levantine basin is ultraoligotropic, the dissolved oxygen produced by primary production would not be consumed at the same rate of its production and this would increase oxygen concentration in deep waters. Also, fast deep water formation could increase oxygen concentrations. A significant dissolved oxygen maximum is located in surface waters of the Levantine basin, which has been reported by several authors (e.g., Schlitzer et al., 1991; Krom et al., 1992; Yacobi et al., 1995) and it has been attributed to the fast layering of the water mass, preventing gas exchange and keeping the oxygen concentration constant (Kress and Herut, 2001). Along to the rapid deep-water formation, other authors invoke changes in solubility and aerobic respiration kinetics due to climate warming as enhancers of high O_2 levels (Powley et al., 2016). Indeed, O_2 consumption rates in the intermediate waters along the whole basin are lower than the global average mesopelagic value (Williams, 2014), as well as nutrients in the present Mediterranean Sea, which level is also low in comparison to the world ocean. In the Mediterranean, nutrient concentrations entering through the Gibraltar Strait are low because the subtropical Atlantic water has low nutrient content. The atmospheric and terrestrial inputs of phosphate and nitrate are not significant enough to increase the nutrient levels in the MAW, therefore, the Mediterranean Sea surface layer remains oligotrophic. In the water column, nutrients are thought to be primarily influenced by precipitation and subsequent remineralization of settling organic debris; their concentration profiles versus water depth reflect uptake in surface waters and release in deep waters (Figure 3). As precipitation is thought to take place in all surface waters, albeit at low rates for the Mediterranean (Chow and Goldberg, 1960; Church and Wolgemuth, 1972; Monnin et al., 1999; Rushdi et al., 2000), the continuous remineralization of organic matter produces increasing nutrient concentrations with age in the deep water mass. This has been clearly demonstrated for Atlantic-Pacific deepwaters (e.g., Broecker et al., 1980). In this study, the increasing nutrients concentration for the Mediterranean Intermediate Water flowing from east to west, and the average nutrient level for Mediterranean Deep Waters in the eastern and western Mediterranean, concur with these general processes (Figure 4) as well as with the data reported by other authors (Lazzari et al., 2016). In addition, the rather moderate concentration gradient is in line with an oligotrophic —i.e. low-productivity— Mediterranean and with the relatively modest ages for this water mass compared to those in the Atlantic-Pacific system. Whereas the Atlantic inflow brings few nutrients, the MOW could export them, making the anti-estuarin circulation and net nutrient removal render the Mediterranean into a marine desert overall. Notwithstanding, Crispi et al. (2001) showed through numerical simulations that an anti-estuarine circulation do not fully explain the oligotrophy of the Mediterranean Sea. Phosphate is a primary productivity-limiting nutrient in the Mediterranean Sea, and its deficit in this basin is not only due to the depleted Atlantic inflow, but also because the armospheric input provide N in excess, which leads to P deficit in seawater. This P deficiency relative to N in atmospheric deposition it is reflected in the unbalanced Redfield ratio in the Mediterranean Sea (Durrieu de Madron et al., 2011; Huertas et al., 2012). The nitrate/phosphate ratio for the intermediate waters given by our data (N:P=20-28:1) confirms the ultra-depletion of phosphate in the Mediterranean Sea, which matches previous reports (N:P=20-27:1) as very far from the world ocean´s typical Redfield ratio (N:P=16:1). Nitrate and phosphate profiles reach their maxima at depth where the maximum bacteria degradation occurs, these nutrients are regenerated faster than silica, whose profiles are strongly correlated with those from Ba (e.g., Jacquet et al., 2005, 2007); accordingly, it would be expected barium to be depleted in surface as much as silica, which is also corroborated by our data. Silica is also the main scavenger of Al, which it is used to predict iron inputs from dust fluxes into the euphotic zone (e.g., Han et al., 2008; Han, 2010; Measures et al., 2005, 2010) since iron is essential for phytoplankton photosynthesis and its concentration in seawater has a direct impact on primary productivity (e.g., Boyd et al., 2007; de Baar et al., 2005). On the other hand, Al impedes the solubility of sedimentary biogenic Si, with the subsequent effect that this would have in productivity (e.g., Emerson and Hedges, 2006; Sarmiento et al., 2006). The major Al source in the Mediterranean Sea is the Saharan dust, which in contact with water dissolves between 1-15% of Al (e.g., Maring and Duce, 1987; Measures et al., 2005). Dissolved Al is attached to solid surfaces of sinking particles (Bacon and Anderson, 1982; Goldberg, 1954; Bruland and Lohan, 2006) and during its pathway to the seafloor Al desorbs or it is released by dissolution of the biogenic carrier particles, increasing its concentration with depth as corroborated by our data (Figure 3). The Al concentrations are used to determine the detrital fluxes to the basin, which is the first step to estimate Ba fluxes in the Mediterranean Sea.

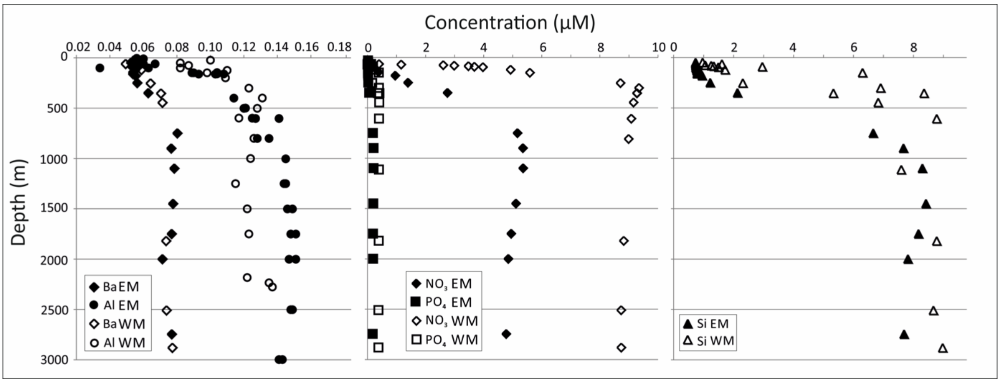

Fig. 3. Ba, Al, NO3, PO4 and Si average concentrations in the water column both in the western Mediterranean (Site 1) and in the eastern (Site 14). See Figure 1, Chapter III.

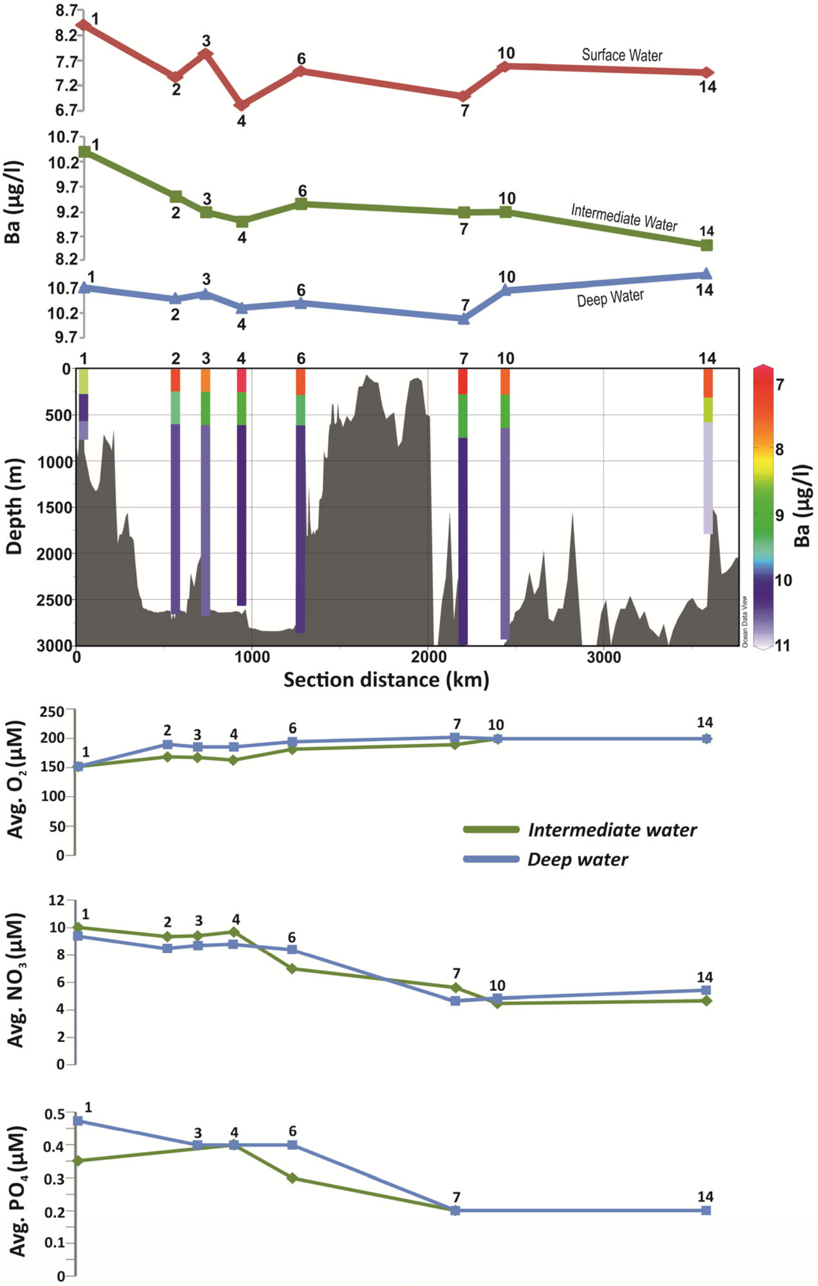

Fig. 4. Average concentrations of Ba (diagram on top), O_2, NO_3 and PO_4 in water masses from stations along the studied Mediterranean sea transect (See Figure 1 in Chapter III). Note that avg. Ba concentrations are plotted with points indicating the station with a number above.

V.1.3. Barium distribution in the water column

The Ba distribution in the water column, with relatively low dissolved Ba in surface waters, reflects the productivity-related Ba-uptake processes and enrichment in deep waters due to partial dissolution of barite (Bernat, 1972; Dehairs et al., 1987; Jacquet et al., 2016; Roy-Barman et al., 2019). High dissolved Ba concentrations are correlated with the Apparent Oxygen Utilization (AOU), a proxy to cumulative net community respiration, linked to bacterial activity and organic matter remineralization (Arraes-Mescoff et al., 2001). The AOU maxima values in the Mediterranean Sea are located in the mesopelagic layer, where barite precipitation takes place. Here marine barite precipitation needs Ba saturated microenvironments linked to organic matter decay to precipitate (e.g., Goldberg and Arrhenius, 1958; Bishop, 1998; Bernstein et al., 1992, 1998), and the high dissolved Ba concentrations are maintained despite barite precipitation because the saturation index for Ba in the Mediterranean Sea is ultra low. Thus, any barite crystal sinking to the seafloor would be immediately dissolved (Jacquet et al., 2016). The Mediterranean Sea can be considered a sort of mini-ocean where surface waters flow from Gibraltar to the most eastern part, and deep waters travel from east to west to flow out into the Atlantic. Accordingly, we would expect to see a consistent, gradual increase in the dissolved Ba concentration in the deep water flowing from east to west, if substantial productivity-related Ba-removal from surface water and release into deep water took place. If no appreciable sources of dissolved Ba occur (rivers, upwelling, dust) while this water flows to the eastern Mediterranean basin, then surface water productivity related processes would even further decrease the surface-water Ba content. In surface waters, the dissolved Ba concentrations reflect the low primary productivity in the oligotrophic present-day Mediterranean. In contrast, there are rather constant deep-water Ba concentrations from east to west, though the Ba-enriched levels in deeper waters relative to surface waters are more difficult to explain. The dissolved Ba enrichment is coincident with the Oxygen Minimum Layer (OML), a layer with ultra-low oxygen concentrations (185 µmol/ kg. Coppola et al., 2018) located from about 200 to 1300 m in the Mediterranean Sea (Millot and Taupier-Letage, 2005). The oxygen consumption in the OML is fuelled by organic matter decomposition exceeding the O_2 input through physical processes such as upwelling and diffusion (Packard et al., 1988; Tanhua et al., 2013). The dissolved Ba increase in this layer it is related to the organic matter decay and the subsequent partial dissolution of marine barite during its pathway to the seafloor. Although productivity in itself cannot explain the Ba increase with depth in the Mediterranean Sea, the residence time of intermediate and deep waters could account for the gradual increase of Ba since the RT of this element is about 10,000 years (Chan et al, 1976), much higher than those of Mediterranean seawater masses. Similarly to the increase of nutrient content with water mass age, an increase in barium has been reported for the Atlantic-Pacific deep water system (e.g., Broecker et al., 1980; Clarke and Coote, 1988; Pérez et al., 1993; Rushdi et al., 2000). Compared to the latter system, the increase in barium in Mediterranean intermediate waters is very modest, which is in line with the low productivity, hence low organic matter fluxes and barite precipitation, and the modest age of this water mass. The long residence time of Ba in seawater compared to the relatively short residence time of the Mediterranean water masses, evidences that the high Ba concentrations observed in deep-water must be related to hydrology and to average biomineralization and dissolution processes over many seasonal cycles, which is concordant with the general idea of a consistent and gradual increase of dissolved Ba concentrations in deep waters from North Atlantic to the North Pacific. In general, oceans (e.g., the Pacific) are undersaturated in barite (Monnin et al., 1999), but the Pacific Ba concentrations are the double than in the Mediterranean Sea, meaning the Mediterranean Deep Water is even more undersaturated relative to barite than other oceans. Furthermore, the Mediterranean salinity —thus sulphate concentration— is ~10% higher than in the Pacific, and therefore the saturated Ba concentration could be ~10% lower. The Ba concentration in deep waters could also vary depending on where the deep-water formation takes place (Adriatic or Aegean) due to the presence of possibly unknown Ba-sources, which should be further investigated. To explore barium systematics, i.e. biological processes and provenance, in a more quantitative way, it is first necessary to assess barium sources and fluxes, a matter discussed in the next sections.

V.1.4. Barium sources in the Mediterranean Sea

After more than four decades of research, the Ba cycle is just beginning to be untangled. The main Ba inputs to the Mediterranean Sea come from the Atlantic Ocean, whose inflowing water contains rather low Ba concentrations. If no appreciable sources of dissolved Ba occur, a depletion of this element could be expected in the surface layer eastward. It is known that evaporation, and therefore salinity increases, however, these do not suffice to explain barite accumulation in the Mediterranean basin since only accounts for a rise of 8% of the dissolved Ba (Jacquet al., 2016), thus, additional sources are required. The plausible Ba sources in the Mediterranean Sea are the Saharan dust, rivers, the Black Sea, hydrothermalism and cold seeps, diffusion from sediments, and even Messinian evaporites. Although some microorganisms may be behind the Ba release, it is difficult to quantify them and the precipitation mechanisms involved are still poorly understood. It is assumed that they have a minor contribution until further research reveals the opposite. Despite efforts made to explain Ba concentrations in the Mediterranean basin through the above sources, there are no sound estimates and no Ba balance in this basin, as the contribution of rivers and dust cannot be clearly depicted to date. Recently, some authors have been proposed for first time a quantitative Ba budget scheme for the western Mediterranean Sea (Roy-Barman et al., 2019). Among other reasons, the absence of quantitative approximations for the Ba budgets in the Mediterranean Sea it is because the Ba sources are difficult to evaluate given the uncertainties in the actual Ba desorption percentage, there is a wide range of estimates and a lack of data from rivers and other regions. Although submarine groundwater fluxes have been recently claimed to be more important than previously thought, yet many uncertainties remain regarding groundwater discharge as well as the Messinian evaporites contributions or the actual Ba input from rivers. We further suggest that the Ba released from human activities should not be underestimated given their increasing impact on Earth and climate change. In the following paragraphs each source will be addressed in greater detail.

Atlantic Ba input

The Atlantic Ocean is the source that introduce more water to the Mediterranean basin (0.83 Sv, Powley et al., 2016). The mean dissolved Ba concentration in the Atlantic surface layer is of 6.1 µg/l (43 ± 3 nmol/kg. Grissom, 2015), which is in line with the Ba concentration found in this water mass once have passed throught the Gibraltar Strait (40–47 nmol/kg. Jacquet et al., 2016; Roy-Barman et al., 2019). Our data in station 1 located in the Alboran Sea, near the Gibraltrar Strait entrance, show a mean dissolved Ba for the upper layer (0-200 m) of 8.3 µg/l, within the range of the reported by Jacquet et al. (2016) and Bernat et al. (1972), who give 5.3 µg/l (38 nmol/kg) and 11.3 µg/l respectively. On the other hand, the average Ba concentration in the MOW, below 200 m water depth, is of 10.6 µg/l at the same site, which is in line with the range of 9.9–10.6 µg/l (~70–75 nmol/kg) given by Jacquet et al. (2016). In the water column, the Ba increase with depth it is related to barite dissolution by pressure changes and the ultra-low saturation index of Ba in the Mediterranean Sea (Jaquet et al., 2016), although the Ba excess in the MOW may be due to lateral accumulation after several remineralization cycles during the water mass pathway westward, first as intermediate and deep water in the eastern basin, and finally as MOW in the western. This is in agreement with the reported for other oceans, such as the Ba increase from the Atlantic to the Pacific related to the ageing of the water mass (Rushdi et al., 2000). Nevertheless, the Mediterranean water masses have a relatively short residence time compared to the mentioned oceans, and other sources have to be invoked to account for the excess of dissolved Ba in this basin.

Ba from the Black Sea

The Black Sea is currently the largest anoxic basin in the world due to its restricted water circulation. There is a salinity gradient between the surface water fed by riverine fluxes and the intermediate and sulphide-rich deep water masses ventilated by the highly saline input from the Mediterranean Sea (Murray et al., 1991), which creates a permanent halocline and prevents oxygenation (Özsoy and Ünlüata, 1997). Primary production in the Black Sea is sustained by the nutrients input from major rivers (Murray et al., 1991; Sur et al., 1996; Yiğiterhan and Murray, 2008) despite the inconsistency of such input in past decades (Codispoti et al., 1991; Tuğrul et al., 1992;

Cociasu et al., 1996; Konovalov and Murray, 2001; Cociasu and Popa, 2002). Although data on dissolved Ba in the Black Sea water column are very scarce, it is known that the Ba vertical profile shows an increase with water depth, as in other seas and open oceans (Falkner et al., 1993). Furthermore, its dissolved Ba concentrations are much higher than in any other marine environment: from 130 – 310 nM/l (17.8 – 42.6 µg/l) in surface waters to 450 – 500 nM/l (61.8 – 68.7 µg/l) in deep waters, and up to twice the value for barite (Falkner et al., 1991, 1993; Moore and Falkner, 1999). In the Black Sea, the dissolved Ba maximum it is located near the redox interface, where microbial activity promotes organic matter deccay and barite dissolution rates are high because of the sulfate reducing conditions. The dissolved Ba concentration of the Marmara Sea inflowing waters to the Black Sea is of 115 ± 5 nM (15.8 µg/l. Falkner et al., 1993). Regarding the Black Sea output to the Mediterranean, Dehairs et al. (1987) made a theorical estimation of 1.8 x 10^{16} µg/yr, which is over three times the input. The sources of Ba to Black Sea surface waters are mainly attributed to upwelling, groundwater discharge and rivers. Moore and Falkner (1999) were the first to report data for the concentrations of dissolved Ba in rivers draining into the Black Sea, showing a range of 215 – 405 nM/l (29.5 – 55.6 µg/l), which matchs with the dissolved Ba outflowing to the Mediterreanean Sea. Nevertheless, the Ba cycle in the Black Sea has not been steady during the past few decades because of anthropogenic impact; high nutrient inputs owing to human contamination have given rise to high productivity, while dams decrease detrital influx from rivers. The theorical estimates for Ba input from the Black Sea to the Mediterranean made by Dehairs et al. (1987) should be confirmed with the modern datasets and having also into account other Ba sources in its balance since there is recent evidence of submarine groundwater discharge in the Black Sea (Schubert et al., 2017).

Ba dissolved in riverine

The major detrital inputs to the Mediterranean Sea have been traditionally attributed to rivers and dust. Estimating riverine detrital input into the ocean is hampered by a lack of consistent long-term data for most small river systems and because of recent changes due to the damming of most large rivers. Total riverine discharge to the basin is currently estimated around 0.3 x 10^{12} m^3/yr (Ludwig et al., 2009). Although riverine discharge is considered as a main continental source of trace metals to the ocean (Oelkers et al., 2011; Jeandel and Oelkers, 2015), rivers are not significant Ba sources in the Mediterranean Sea since their contribution to the Ba fluxes is < 3%. It is believed that only major rivers could account for Ba release in deep-water formation areas. In general, 10% of the fluvial discharge reaches the deep basin while the remaining 90% is deposited in deltaic and continental shelf areas (Martin et al., 1989). The Rhône (1550 ± 200 m^3/s) is the main river providing sediment fluxes to the western Mediterranean basin and the fraction of suspended matter exported beyond the coastal zone, thus between 1.5 and 10% actually makes it to the deep-sea. Roy-Barman et al. (2019) calculated average dissolved Ba fluxes along the Rhône River at 1.1 x 10^7mol/yr (1.5 x 10^6 kg/yr). Neglecting the contribution of other rivers and assuming 100% of particle dissolution, these authors assessed a total dissolved Ba input by rivers to the western basin at (6.0 ± 1.3) x 10^7 mol/yr (8.2 x 10^6 kg/yr), which roughly fits the estimate of Dehairs et al. (1987). In the eastern basin, mainly the Rosetta and Damietta branches of Nile River discharge freshwater into the Mediterranean Sea and form the Nile delta. The Nile flow is very fluctuant: the average total annual Nile discharge in the period 1959-1964 reached 42.9 km^3, whereas after the Aswan dam in 1964 the flow to the estuary decreased to just 18-21 km^3 (Halim, 1991). The Nile freshwater reaching the Mediterranean annually amounts to 2.5 – 4 km^3 (Halim, 1991). The Rosetta Nile branch influx constitutes the main winter source of nutrients in the coastal area off the Nile delta (Hamza, 2005). El-Bouraie et al. (2010) reported an averaged dissolved Ba concentration of 0.204 mg/l (0.5 – 0.8 x 10^6 kg/yr) in surface waters close to the mouth of this branch (after Aswan dam). Nevertheless, the Ba input from rivers may be underestimated since there are a large number of unmonitored coastal basins and minor rivers have been ignored. As example, in the western Mediterranean only the Rhône river has been considered as Ba source but other French, Spanish, Algerian, Tunisian and Italian rivers could also account since Wang and Polcher (2019) found that more freshwater (40 – 60%) flows into the Mediterranean Sea than previously estimated, also pointing to the neglected submarine groundwater discharge. In the central Mediterranean and the eastern basin not only the Po and Nile but other Italian rivers and unaccounted subterranean rivers should be taken into account.

Ba dust related sources

Along with river contributions, atmospheric deposition has been traditionally invoked as major source of trace metals in the ocean (Duce et al., 1991; Jickells, 1995; Bowie et al., 2002; Mahowald et al., 2005; Mackey et al., 2012). Large amounts of Saharan dust are deposited both in the western and the eastern Mediterranean surface waters by wet (rainfall) or dry deposition and it has been considered a dominant Ba input to the Mediterranean basin (e.g., Dehairs et al., 1987; Bonnet and Guieu, 2006) despite being a source with high intra-annual and inter-annual variability (Avila et al., 1998; Guieu et al., 2010). Indeed, both in the western and eastern Mediterranean Sea, the fluxes of many trace elements from dust to the surface layer are much higher than those from the Rhône and Po rivers (Guieu et al., 1997; Guerzoni et al., 1999). The average present-day Saharan dust fluxes vary from ~1 mg/cm^2 yr in the western Mediterranean Sea up to 3.5 mg/cm^2 yr in the eastern basin (Herut and Krom, 1996; Guerzoni et al., 1999; Guerzoni and Molinaroli, 2005), the last one is almost double than the 2 mg/cm^2 measured in sediment traps reported by Rutten et al. (2000), and account for 10 - 20% of the recent deep-sea sedimentation in the western Mediterranean (3 -15 mg/cm^2 yr). Dulac et al. (1996) and Moulin et al. (1997) compared Meteosat data with measured aerosols at the Corsica site for a six-month period, finding that the average measured aerosol flux was roughly 0.9 mg/cm^{2}yr, whereas Meteosat data yielded an average of 0.8 mg/cm^2 yr (0.4 - 1.1 mg/cm^2 yr) for the same period. They also compared the atmospheric fluxes derived from Meteosat with dust fluxes measured in sediment traps for a period of two years. The Meteosat gave a similar range both years, 0.35 - 1.1 mg/cm^2 yr, while the sediment trap data showed different fluxes, 1.2 and 0.9 mg/cm^2 yr. To explain these variations, these and other authors (Heimbürger et al., 2014) point to several short-term hydrological and biological processes controlling the downward flux of particles in the water column. Atmospheric deposition cannot explain by itself the downward transport, especially in the eastern Mediterranean, and it is rather a conjunction of dense water convection and surface primary productivity leading to the formation of aggregates that facilitates the transport of particulate material to the bottom (Theodosi et al., 2019). Guerzoni et al. (1999) estimated annual dust fluxes of 8 – 12 – 35 g/m^2 for the Western Mediterranean (WM), Central Mediterranean (CM) and Eastern Mediterranean (EM), respectively. Guerzoni and Molinaroli (2005) used these values to arrive at a total annual dust flux for the whole Mediterranean of ~40 x 10^6 tons, pointing out that these fluxes amount to one third of riverine sources. In general, it is assumed that mineral dust aerosols dissolve in the ocean surface in a range of 1.5-5% (Measures et al., 2008). Hydes et al. (1988) assumed that 10% of the dust input is Al, and of this percentage 5% is finally dissolved in the ocean (deduced from Pacific Ocean: Maring and Duce, 1987; Prospero et al., 1989), assuming this in a steady state model framework that considers negligible the influence of rivers and sediments for the Al contribution. This was corroborated by Chou and Wollast (1997) with a sophisticated data set and budget calculations for the western Mediterranean; and Moreno et al. (2006) arrived at a close estimate of 6% total Al flux (reported as 12% Al_2O_3). The same fractional solubility can be considered both for Al and Ba (Weldeab et al., 2014), thus 5%. Although other authors proposed much higher percentages —such as Dehairs et al. (1987), who gave 75% of Ba from dust finally dissolved in seawater— a Ba fractional solubility in agreement with the range 1.5-5% previously reported is the most reasonable (Desboeufs et al., 1999; Measures et al., 2008; Hsu et al., 2010). Still, Roy-Barman et al. (2019) considered the percentage given by Dehairs et al. (1987) and derived a Ba flux of 0.6 ×10^8 mol/yr from dust finally disolved in the western Mediterranean, but they coud not justify the Ba loss due to water circulation. In general, it has been concluded that dust fluxes are negligible and the dissolved Ba distribution is mainly driven by hydrodynamics (Weldeab et al., 2014; Jacquet et al., 2016).

Submarine groundwater discharge

Submarine Groundwater Discharge (SGD) is known to play an important role in the biogeochemical cycles of marine environments. These fluxes comprise both fresh meteoric groundwater and recirculated seawater through permeable marine sediments. The location and quantification of SGD inputs are challenging, but the combination of different methods —aerial thermal infrared images, electromagnetic techniques, salinity and temperature measurements, Rn and Ra tracing or direct measurement of the spring outflow by seepage meter— allow for an approximation to the total groundwater outflow (e.g., Ollivier et al., 2008; Moore, 2010; Mejías et al., 2012; Bejannin et al., 2017; Bejannin et al., 2020). SGD has been estimated up to 4 times higher than riverine discharge into the oceans (Kwon et al., 2014), about 0.7–6% of the global river discharge (Zektser, 2006; Beck et al., 2013; Zou et al., 2019; Luijendijk et al., 2020). SGD is a significant source of chemical elements (Moore et al., 2008; Beusen et al., 2013; Trezzi et al., 2016; Cho et al., 2018; Robinson et al., 2018) and it is a Ba-rich reservoir. The concentration and desorption of Ba during marine water intrusions are being alike to those from Ra (Li and Chan, 1979; Moore, 1996; Rama and Moore, 1996), meaning a Ba release about ten times higher than from rivers (Shaw et al., 1998). Indeed, the Ba fluxes from SGD are ranging to 5–57% of the riverine flux (Gaillardet et al., 2003; Mayfield et al., 2021). Despite the recent attention to the SGD, in the western Mediterranean, these fluxes have been already calculated in some coastal areas (Garcia-Solsona et al., 2010; Mejías et al., 2012, Rodellas et al., 2012, 2017). The total SGD fluxes to the Mediterranean Sea ranges from 0.3 x 10^{12} to 4.8 x 10^{12} m^3/yr, i.e. up to 16 times larger than the riverine fluxes and it constitutes an important source of nutrients and trace elements (Basterretxea et al., 2010; Garcia-Solsona et al., 2010; Rodellas et al., 2015; Trezzi et al., 2016; Roy-Barman et al., 2019; Bejannin et al., 2020). In their study of karstic submarine groundwater discharge, Chen et al. (2020) estimated dissolved inorganic nitrogen (DIN) and dissolved inorganic phosphorous (DIP) fluxes to the Mediterranean Sea equivalent to 8 – 31% and 1 – 4% of total riverine inputs, respectively. Furthermore, in some regions of the Mediterranean Sea the importance of SGD has been demonstrated in the supply of dissolved trace metals when rivers cannot account for the observed enrichment (Moore et al., 1997; Shaw et al., 1998; Rodellas et al., 2014; Tovar-Sánchez et al., 2014; Trezzi et al., 2016; 2017). The Ba flux to the Mediterranean Sea can be controlled by climate patterns such as sea level changes (Shaw et al., 1998; Maciopinto and Liso, 2016; Veit and Conrad, 2016) but also by seasonal oscillations in groundwater chemistry related to the North Atlantic Oscillation (NAO) patterns (Llovel et al., 2011; Sweet and Zervas, 2011; Tremblay et al., 2011; Gonneea et al., 2013; Neves et al., 2019). The depth and location of the measurements also affect the value of the trace metal concentrations (Charette and Sholkovitz, 2002; Beck et al., 2007) and there is a high variability of the Ba/^{228}Ra ratio in groundwater. Given the likely importance of SGD in the Ba cycle, such uncertainties should be further investigated.

Hydrothermal vents and cold seeps

Some additional sources of dissolved Ba to deep waters have been largely ignored, the submarine hydrothermal fluxes and cold seeps. Hydrothermal vents and cold seeps expel chemical-rich fluids from the seafloor usually composed of natural gases and other hydrocarbons, being cold seeps less ephemeral and with fluxes at lower temperature. The actual amount of Ba derived from these systems is not well quantified in the global Ba budget due to difficulties in locating and measuring them. Still, it has been reported that cold seeps are common in continental margins and their contribution is as significant as hydrothermal vents, responsible for 2 – 13% of the global Ba input to the oceans (Elderfield et al., 1996; Von Dam et al., 1985; Edmond et al., 1979) with fluxes ranging 10-40 µmol/kg (Von Damm et al., 1985; Paytan and Kastner, 1996; Dickens et al., 2003) and similar for cold seeps (Torres et al., 1996, 2001, 2002; McQuay et al., 2008). The release of Ba from these systems may result in the authigenic precipitation of barite, which is mainly consumed in the formation of chimneys (Torres et al., 1996, 2002; Aquilina et al., 1997; Naehr et al., 2000; Greinert et al., 2002), and little Ba remains available for release to seawater. Nevertheless, barite microcrystals from friable cold seep structures can be exported and affect dissolved barium fluxes (Aloisi et al., 2004; McQuay et al., 2008). In the Mediterranean Sea, although some authors have overestimated the Ba fluxes from these systems (1.4 nmol/cm^2 yr. Carter et al., 2020), it has been demonstrated the influence of hydrothermalism releasing particulate Ba in some areas of the Aegean Sea, with a Ba flux from 0.11 to 1.45 µg/l (Megalovasilis, 2020). This is only an example among many others in the Mediterranean Sea (e.g., Varnavas and Cronan, 1991; Dando et al., 1999; Dekov and Savelli, 2004). As these fluxes are suspected to have a significant impact in the current Ba cycle in restricted marine basins (Torres et al., 2002) and may have affected the global ocean barium cycle in past geological times (Dickens et al., 2003; McQuay et al., 2008), it worthwhile to further investigate and quantify their Ba fluxes to the Mediterranean Sea.

Diffusion from the seafloor

Another Ba source to bottom waters is fluid diffusion from anoxic sediments where sulfate reduction has released barium to pore waters. This usually leads to barite reprecipitation fronts where reduced fluids interact with oxic pore waters or regular seawater at the water-sediment interface (Torres et al., 1996). It has been demonstrated that Ba increases with water depth in west of Sargaso Sea due to diffusion from the seafloor (Van Beek et al., 2007). Furthermore, high dissolved Ba concentrations in bottom waters in the Bay of Bengal also indicate fluxes from seabed, which means that Ba dissolution within the sediment and its subsequent migration in pore waters would suffice to provide dissolved Ba in stagnant bottom waters (Singh, et al., 2013). In the western Mediterranean Sea and the Sicily Strait, the Ba concentration in pore waters is four times that in the overlying seawater (Michard et al., 1974; Böttcher et al., 2003), promoting Ba fluxes from sediments. Indeed, Roy-Barman et al. (2019) used high resolution pore water data from other authors (Michard et al., 1974) and calculated a Ba concentration gradient of ~1020 nmol/kg/m and a diffusive flux of Ba from the sediment to seawater of 1.8 x 10^{-2} nmol/m^2/s, which is high compared with those from other oceanic sediments (Falkner et al., 1993; McManus et al., 1998). These authors give a Ba flux approximation of 4.75 x 10^8 mol/yr from sediments to the western Mediterranean basin, proposing that this could be enough to balance the Ba budget of the deep water masses. Meanwhile, in the eastern basin, the outcrop of Messinian evaporites can release dissolved Ba from the sediment directly to deep waters, so that Ba-rich bottom waters are associated with brines (De Lange et al., 1990; Jacquet et al., 2016). Although evaporites have been identified through seismic data in the western basin (Driussi et al., 2015), no current brines have been observed. Diffusion from the seafloor presents in general many uncertainties and it should be further investigated in order to assess more accurately the Ba budgets in the whole Mediterranean Sea.

Other Ba sources

It is a fact that human activities leave an ecological footprint on our environment, but the anthropogenic influence on marine Ba budgets has been overlooked, considered irrelevant on the geologic time scale. Yet given the peaking interest in climate change and the ecological impact of our developing industry, it would be advisable to take into account anthropogenic Ba fluxes in the current Mediterranean Ba balance. Ba-rich drilling fluids can reach estuarine sediments and be discharged to the ocean during oil well drilling campaigns, both onshore and offshore, releasing high amounts of particulate Ba (Boothe and Presley, 1989; Neff and Sauer, 1995). In fact, between 295 to 411 tons of barite are discharged to the ocean in conjunction with these activities (Roy-Barman et al., 2019), and this mineral may slowly dissolve under reducing conditions, enriching the neighbouring waters in dissolved Ba (Boothe and Presley, 1989). Proof of this is that in some coral strains from Thailand were found to have up to 18 µg/g of Ba in their skeletons as a result of well drilling (Howard and Brown, 1986). The intensified exploration after the 60s resulted in a higher variability of the Ba/Ca ratios in coral skeletons (Neff, 2002). Other marine organisms, such as bivalves, have likewise been affected by production platforms, showing higher Ba concentrations in their tissues than animals from areas without oil well influence (e.g., Boesch and Rabalais, 1989; Spangenberg, and Cherr, 1996). Offshore oil exploration has been minimal in the Mediterranean Sea, limited to some explorations by Italy in the Adriatic Sea during the 50's and in the Aegean Sea and the Gulf of Gabest (east coast of Tunisia) during the 70's-80's, drilled by Greece, Italy and Libya. Nevertheless, at present there are production platforms offshore of Spain, Malta and some North African countries, and there are plans to drill and install wells offshore Northern Greece during 2021 (Carpenter and Kostianoy, 2018). Turkey has also become interested in the Libyan sea exploration, having signed an agreement with the Tripoli Government. This means that human impact is on the rise in the Mediterranean, so that its influence in Ba cycling could be much more significant in the coming decades. Further research is needed to clarify the actual and eventual impact of this factor.

V.2. Results

The salinities and temperatures (Figure 2) of the studied water masses are ranging 37 to ~39 psu and 15 to ~18 °C for the surface waters (0-200 m), from the Alboran Sea to the Levantine Basin respectively. Regarding the LIW (200-600 m), it is characterized at ~39 psu and ~16 °C, which denotes a higher density than the overlying water mass. The deep waters in the eastern basin (>600 m) have similar salinity than the LIW but presents lower temperature (~14 °C), whereas the western Mediterranean shows a slightly lower salinity (~38 psu) and temperature (~13 °C). The average nutrient concentrations in the surface layer in site 1, located in the Alboran Sea, are 5.3 µM for

NO_3 and 0.3 µM for PO_4, while in the easternmost part of the Mediterranean (site 14) are 0.24 for NO_3 and 0.02 µM for PO_4 (Figure 4). As expected, for the LIW the nutrient concentrations increase with depth, 10.1 µM for NO_3 and 0.5 µM for PO_4 in site 1, and 3.4 for NO_3 and 0.1 µM for PO_4 in site 14. On the other hand, the average oxygen concentrations thought the whole basin for surface waters is of ~224 µM, for intermediate waters of ~182 µM and for deep waters ~189 µM. Note that in Figure 4 the plot do not shows O_2 for surface waters since there are seasonal changes with a strong influence in the values, reflecting the different sampling periods. Our Ba profiles show dissolved Ba concentrations ranging from 6.5 to 11.1 µg/l, which are roughly in line with the 5.6 µg/l to 10.3 µg/l (40.5 – 75.3 nmol/kg) given by Roy-Barman et al. (2019) for the whole Mediterranean. Our data show averaged Ba concentrations of 8.3 µg/l in the surface layer (0-200 m) at the Gibraltar entrance, coherent with the 8.5 µg/l (62 nmol/kg) of Roy-Barman et al. (2019), and 7.5 µg/l in the easternmost part of the basin for the same water mass, which supports the discrete productivity in the Mediterranean Sea and it is only slightly higher than values previously reported (45 nmol/kg – 53.9 nmol/kg, thus 6.3 – 7.4 µg/l; Jacquet et al., 2016; Roy-Barman et al., 2019; see Figure 6 of this chapter). In the intermediate water layer (200-600 m), the dissolved Ba concentration average is of 9.1 µg/l, which concurs with the previously reported data (Jaquet et al., 2016; Roy-Barman et al., 2019), and a lateral gradient of 0.6 µg/l is observed (avg. 8.9 µg/l in the eastern to 9.5 µg/l in the western basin; see Table 2, Figure 6), which can be attributed to progressive Ba accumulation during its pathway westward, that is, hydrodynamics. For the deep waters the Ba concentrations appear to be quite constant, with a dissolved Ba average of 10.3 µg/l, nearly the same as reported in previous research (70-75 nmol/kg, thus 9.6 µg/l; Jaquet et al., 2016; Roy-Barman et al., 2019). The lowest dissolved Ba value is of 6.4 µg/l and it is located in the Alboran sea (site 4), whereas the highest value is of 11.7 µg/l, in the Erathostenes seamount (site 14). At station 10, we observe a significant Ba increase from ~10 at 3104 dbar to 11µg/l at 3107 dbar, along to the typical salinity and temperatures of the deep water mass (38.7 psu and 13.9 °C).

Table 1. Dissolved Ba concentrations in the water column of the studied stations.

1	Depth	Ba	2	Depth	Ba	3	Depth	Ba	4	Depth	Ba	5	Depth	Ba	6	Depth	Ba	7	Depth	Ba
1	834	10.4	1	2769	9.4	1	2797	10.4	1	2793	10.4	1	2881	10.6	1	2563	10.8	1	2967	
2	828	11.0	2	2760	10.9	2	2700	11.0	2	2770	10.4	2	2509	10.1	2	2563	10.5	2	2967	
3	807	11.1	3	2723	10.1	3	2495	9.9	3	2722	10.6	3	1820	10.1	3	2557	10.6	3	2967	
4	706	10.8	4	2500	11.0	4	2036	11.1	4	2501	10.4	5	446	9.8	4	2538	10.3	4	2967	9.7
5	706	10.0	5	2001	10.8	5	1526	10.8	5	2000		6	354	9.7	5	2440	10.1	5	2967	10.3
6	605	10.8	6	1499	10.8	6	1018	10.7	6	1500	10.4	7	254	8.8	6	1933	10.6	6	2762	10.2
7	454	10.5	7	997	10.6	7	1018	10.2	7	1002	10.2	8	121	8.1	7	1599	10.6	7	2558	10.0
8	303	9.8	8	750	10.6	8	509	10.3	8	599	10.1	9	95	7.4	8	1262	10.5	8	2044	10.3
9	226	9.5	9	500	10.4	9	509	9.2	9	449	9.7	10	85	7.4	9	970	10.2	9	1536	10.2
10	177	9.4	10	350	9.2	10	257	9.1	10	303	9.2	11	80	7.9	10	658	10.0	10	1027	10.3
11	127	9.3	11	250	8.8	11	257	8.3	11	249	8.9	12	75	7.3	11	396	9.4	11	722	10.2
12	105	9.0	12	201	8.3	12	156	7.8	12	102		13	70	7.4	23	396	9.3	12	520	9.5
13	96	8.9	13	148	8.3	13	106	8.1	13	75	7.1	14	61	6.8	12	298	9.3	13	267	9.1
14	90	9.2	14	101	8.2	14	106	7.8	14	60	7.0	15	48	7.2	13	197	8.5	14	129	8.1
15	85	8.7	15	75	7.3	15	95	8.4	15	55	6.9	16	65		14	122	7.8	15	105	7.1
16	81	8.0	16	58	7.4	16	90	7.9	16	50	6.9	17	91		15	93	7.6	16	94	6.7
17	75	7.7	17	50	7.4	17	85	7.8	17	46	7.3	18	151		16	80	7.6	17	84	6.9
18	70	7.5	18	45	7.4	18	80	7.8	18	43	7.0	19	302		17	69	7.2	18	80	6.9
19	70	7.7	19	39	7.1	19	75	7.7	19	39	6.5	20	354		18	64	7.3	19	75	6.9
20	65	7.6	20	34	7.2	20	70	7.5	20	33	6.5	21	667		19	59	7.0	20	69	6.8
21	60	7.5	21	30	7.4	21	65	7.3	21	30	6.8	22	809		20	54	7.5	21	64	6.9
22	49	7.3	22	25	6.9	22	55	7.3	22	24	6.4				21	49		22	55	7.1
			23	21	7.0				23	20	6.5				22	39	6.8	23	44	6.9
			24	14	6.9				24	15										

8	Depth	Ba	9	Depth	Ba	10	Depth	Ba	11	Depth	Ba	12	Depth	Ba	13	Depth	Ba	14	Depth	Ba
1	2380	10.1	1	3390	11.1	1	3107	11.0	1	3542	10.4	1	655		1	1513		1	2028	10.8
2	2380	9.7	2	3390	10.8	2	3100	10.2	2	3500	10.5	2	655	9.1	2	1513	9.3	2	1998	10.7
3	2378	9.9	3	3350	10.9	3	3001	10.3	3	2746	10.6	3	645	9.1	3	1490	9.4	3	1973	11.1
4	2368		4	3300	10.9	4	2500	10.4	4	2000	9.8	4	635	9.1	4	1465	9.5	4	1923	11.4
5	2320	10.1	5	3200	10.9	5	2000	10.7	5	1750	10.5	5	615	9.0	5	1440	10.0	5	1822	11.7
6	2200	10.2	6	3100	10.1	6	1500	10.7	6	1450	10.7	6	600	9.2	6	1415	9.2	6	1517	11.3
7	1699	10.2	7	3000	10.9	7	1000	10.9	7	1100	10.8	7	550	9.0	7	1365	9.6	7	1263	11.5
8	1199	10.9	8	2500	10.8	9	450	9.6	8	900	10.5	8	500	8.6	8	1200	9.3	8	1110	11.4
9	698	9.8	9	1750	10.9	10	299	8.8	9	750	11.0	9	400	8.6	9	1100	9.3	9	1010	10.8
10	399	9.5	10	1250	10.1	11	174	8.5	10	350	8.6	10	300	8.0	10	1000	8.9	10	1010	
11	239	8.6	11	750	10.9	12	100	8.0	11	250	7.7	11	200	7.8	11	850	9.6	11	1010	
12	150	7.7	12	500	10.2	13	85	7.9	12	180	7.6	12	150	7.3	12	700	8.6	12	909	11.4
13	109	7.4	13	375	9.7	14	75	7.8	13	160	7.4	13	145	8.0	13	600	8.8	13	808	11.1
14	95	7.8	14	250	8.9	15	70	7.7	14	145	7.4	14	140	8.4	14	500	8.3	14	706	10.4
15	85	7.6	15	200	8.1	16	65	7.6	15	130	7.5	15	125	7.6	15	400	8.1	15	656	10.7
16	80	7.6	16	175	8.2	17	60	7.7	16	116	7.5	16	100	7.8	16	300	7.8	16	606	10.4
17	75	7.3	17	150	7.9	18	54	7.5	17	100	7.6	17	60	7.8	17	250	8.0	17	504	9.6
18	70	7.0	18	125	7.8	19	50	7.5	18	91	7.5	18	40	7.9	18	200	7.8	18	403	8.3
19	64	7.2	19	100	7.7	20	45	7.5	19	85	7.5	19	30	8.1	19	150	7.6	19	302	7.5
20	60	7.0	20	75	7.9	21	40	7.3	20	80	7.6	20	20	8.7	20	125	7.6	20	201	7.5
21	55	6.9	21	50	7.9	22	34	7.5	21	75	7.4	21	15	9.2	21	100	7.6	21	152	7.7
22	49	6.8	22	25	7.6	23	24	7.3	22	65	7.6	22	10	9.3	22	75	7.5	22	126	7.4
23	45	7.0				24	15	7.5	23	50	7.4	23	5	9.7	23	50	7.5	23	102	
						24	30	7.4							24	25	7.4	24	76	

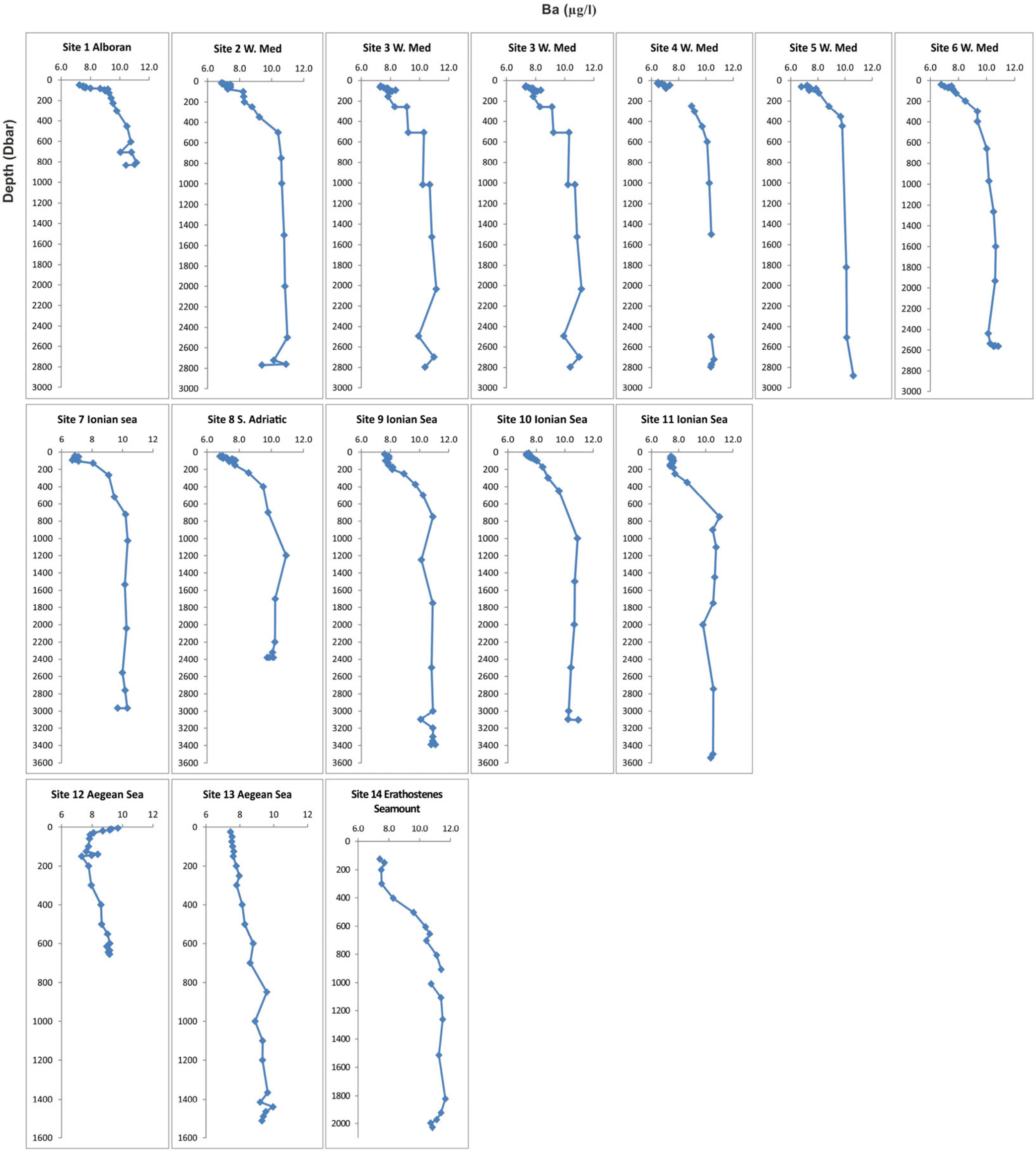

Fig. 5. Vertical profiles of dissolved Ba concentrations in the seawater column of the studied stations.

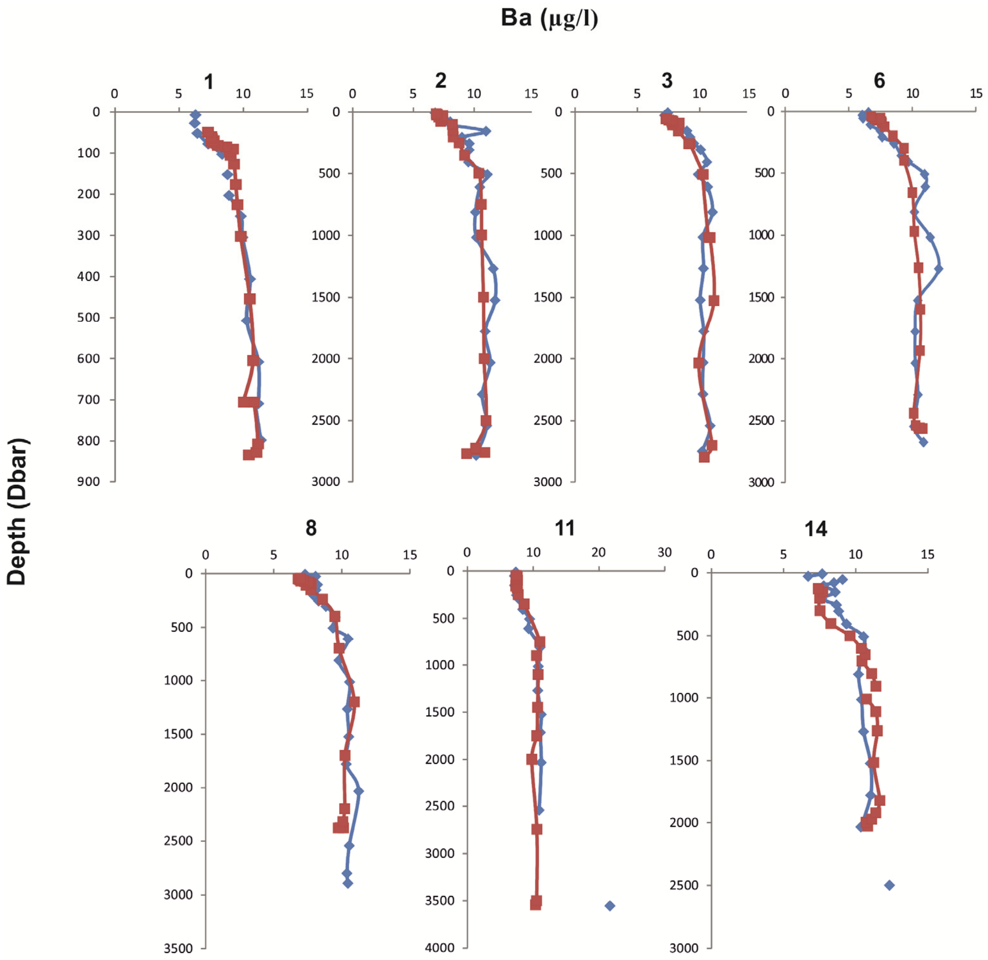

Fig. 6. Some Ba profiles obtained in this thesis, in red, compared with those from Jacquet et al. (2016), in blue.

V.3. Discussion

The physical and chemical attributes of the water masses forming the Mediterranean Sea overturning circulation are confirmed by our salinity and temperature profiles, from the Atlantic inflowing surface waters, relatively warm, to the denser and colder intermediate and deep waters (Figures 1 and 2). Our data also corroborates the Mediterranean oligrotrophy and the ultra-depletion of phosphate especially in the eastern basin (Figures 3 and 4). The derived nitrate/phosphate ratio (N:P=20-28:1) it is in line with the previously reported (N:P=20-27:1), very far from the world ocean´s typical Redfield ratio (N:P=16:1). The increasing nutrients concentration both for intermediate and deep water masses flowing from westward (Figure 4) concur with the data reported by other authors (Lazzari et al., 2016) and with the general biological processes and accumulation with aging of the deep water masses as in e.g., the North Atlantic – Pacific system. In Figure 4, it can be also deduced the good ventilation of the Mediterranean water masses in comparison to other oceans since the oxygen concentrations are relatively high and have a slight increase eastward, supporting the oxygen excess over biological demand due to deep-water formation inputs and low productivity, with the oxygen minimum located as expected. Regarding the Ba profiles provided in this thesis (Table 1, Figure 4 and 5), these corroborate the biological Ba-uptake in surface waters and the dissolved Ba increase with water depth due to barite partial dissolution, also in line with the previously reported (Bernat, 1972; Dehairs et al., 1987; Jacquet et al., 2016, Roy-Barman et al., 2019). Ba concentrations in surface waters support the discrete productivity in the Mediterranean Sea, which is further confirmed by the slight Ba increase westward in the intermediate layer, highlighting the modest age of this water mass. The high dissolved Ba accumulations in the deep water masses can be attributed to partial dissolution of barite because of pressure changes and the ultra-low saturation index of Ba (Jacquet et al., 2016). The data provided here also reflect the gradual increase in the dissolved Ba concentrations in the deep water flowing westward due to the accumulation after several Ba cycles with the aging of the deep water mass. Overall, there is a Ba increase from surface waters to 600 m and a relatively constant concentration below 600 m. This profile reflects the nutrient-type behavior of Ba, with Ba uptake by biologic processes in the upper and intermediate water layers and gradual release downward tied to organic matter decay and partial dissolution of barite (Figs. 6 and 7). In general, the high dissolved Ba concentrations are correlated with the AOU and the OML, thus bacterial activity and organic matter decay linked to marine barite precipitation. It may be contradictory, but barite precipitation needs of such Ba saturated microenvironments and at the same time the low Ba saturation in the Mediterranean Sea leads to dissolve any sinking barite crystal (Jacquet et al., 2016). The highest values of dissolved Ba were observed at station 14, coincident with the pelagic high of the Erathostenes Seamount, have been also reported by Jacquet et al. (2016) who attributed the high values to the local dissolution of settling barite particles related to organic debris. Despite of these assumptions, productivity in itself nor the traditional accounted sources (dust and rivers) cannot explain the Ba unbalance in the Mediterranean Sea. In station 10 (3107 dbar), we observe a significant Ba increase just above the bottom, which could be attributed to the influence from sediment fluxes or perhaps even to a brine located at roughly 3323 dbar in the Bannock Basin, which is close to this site (Luther et al., 1990; De Lange et al., 1990), since the dissolution of barite in brines and at the interface lead to enhanced concentrations of Ba (De Lange et al., 1990). The Ba provided by the brine could contribute to the increase at the bottom of station 10 by lateral transport, although the temperature and salinity profiles in this site do not reflect such an influence. The role of brines, cold seeps and diffusion from the seafloor should be further investigated and quantified in order to improve our knowledge of the Ba cycle and its budgets in the Mediterranean Sea. Figure 7 offers a general diagram of the marine Ba cycle for the Western Mediterranean Sea (Carter et al., 2020). These authors reported the terrestrial input including rivers and groundwater (6.14 nmol/cm^{2}yr) although these sources are likely being underestimated since most of them are under-explored and the same thing would occur for the hydrothermal sources. They also include dissolution in sediment in the flux for dissolution in the water column but they justify the most Ba fluxes mainly with the internal cycling. Calculating Ba fluxes accurately is no easy task, given the uncertainty of variables such as the fractional solubility of Ba, among others, traditionally considered the same as for Al (Weldeab et al., 2014). Over the last decades very different values have been proposed, ranging from 1.5% to 80% (Dehairs et al., 1987; Arraes-Mescoff et al., 2001), although the most broadly accepted values are 1.5-5% (Chester et al., 1993; Desboeufs et al., 1999; Measures et al., 2008; Hsu et al., 2010). In light of this data fluctuation, results may lean towards an overestimation or underestimation of the Ba fluxes. Other authors recently studying Ba budgets for the Western Mediterranean report a net flux of -7.0 x 10^8 ± 1.8 x 10^8 mol/yr at the Gibraltar strait (Roy-Barman et al., 2019), a negative Ba balance that reflects the need of additional Ba sources. These authors also give a flux of 0.6 x 10^8 mol/yr from dust deposition taking into account the highest Ba desorption value (80%), and the same flux from rivers (Rhone and Ebro) assuming a particulate Ba dissolution of 100% and neglecting the contribution of other rivers, meaning a possible underestimation of their Ba contribution. At any rate, inputs from Saharan dust and rivers appear to be insufficient for explaining the imbalance in the Mediterranean Ba budget, since their contributions are at least 10 times lower than the net Ba loss from the western basin. Sediment fluxes could balance the Ba budgets in the deep water masses: they are estimated to provide, in the western Mediterranean alone, a total Ba flux of 4.75 x 10^8 mol/yr (Roy-Barman et al., 2019). Another potential source of dissolved Ba would be Ba-rich brines from Messinian evaporites, identified both the eastern and the western basin (De Lange et al., 1990; Driussi et al., 2015; Jacquet et al., 2016), although their contribution to the Ba budget calls for quantification. Likewise, submarine groundwater discharge is an underexplored dissolved Ba source that could account for the Ba imbalance, with a flux of 3.6 ± 2.4 x 10^8 mol/yr just for the western basin (Roy-Barman et al., 2019). All things considered, some kind of diffusion would be the most plausible source behind the Ba imbalance in the entire basin, but it is necessary to quantify their contribution. Moreover, the internal Ba cycle is only partially understood; the role of bacteria —which is discussed in the next chapter— requires further investigation, beyond the qualitative approaches undertaken to date.

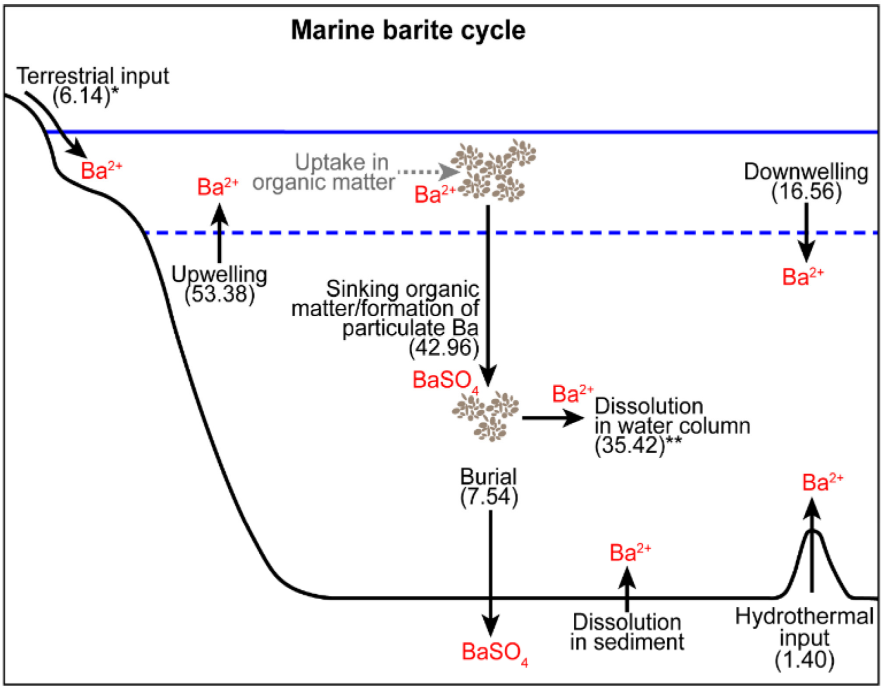

Fig. 7. Simplified diagram of the marine barite cycle in the Western Mediterranean Sea. Fluxes are shown with black arrows (in nmol/cm^{2}yr). Dissolved Ba is shown in red as Ba^{2+} and the particulate phase as $BaSO_4$. Note that Ba is also associated with other terrigenous and authigenic minerals not depicted in this figure. *Terrestrial input includes rivers and groundwater discharge. ** Dissolution in sediment is included in the flux for dissolution in the water column (from Carter et al., 2020).

V.4. Conclusions

High-resolution data on barium in the water column along a Mediterranean Sea transect, obtained during different oceanographic campaigns, have been analyzed in order to a better understanding of Ba distribution in this basin. In surface waters, the chemistry of the water column reflects the modest productivity of the Mediterranean Sea and a Ba gradient evidencing high Ba concentration in the deep layers throughout the basin when compared to those from the surface layer. The data of this thesis further support the Ba imbalance predicted by Dehairs et al. (1987) and recently corroborated by Roy-Barman et al. (2019), highlighting the underestimation of an external Ba source to the Mediterranean Sea since the contributions of Sahara dust and rivers are insufficient to explain the Ba deficit in this basin. Submarine groundwater discharge, brines from Messinian deposits, or some kind of diffusion from sediments could be suitable candidates for the missing Ba input in the balance and further investigations are needed on this topic. The complexity and difficulty of unveiling the actual Ba contribution from these sources underline the necessity to clarify Ba budgets and fluxes in the whole Mediterranean Sea in order to constrain the likely source. In this sense, we provide new data correlated with those from previous research, which will help to complete the Ba balance scheme in future investigations.

Chapter VI. Role of bacteria in marine barite precipitation: A case study using Mediterranean seawater

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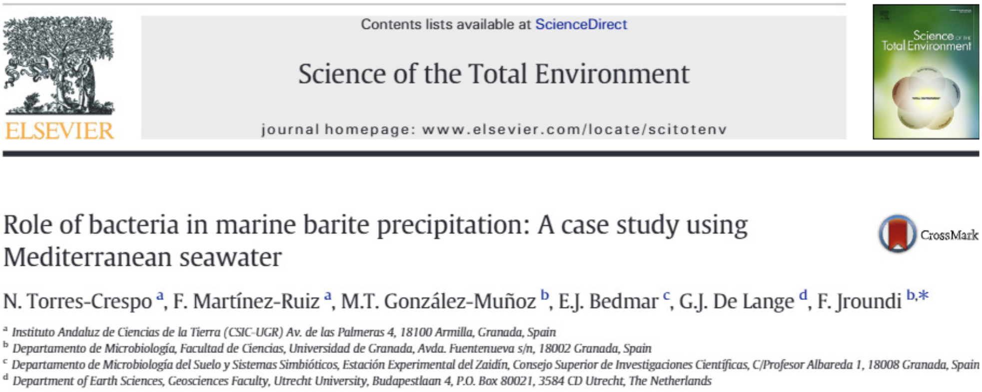

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Role of bacteria in marine barite precipitation: A case study using Mediterranean seawater

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VI.1. Abstract

Marine bacteria isolated from natural seawater were used to test their capacity to promote barite precipitation under laboratory conditions. Seawater samples were collected in the western and eastern Mediterranean at 250 m and 200 m depths, respectively, since marine barite formation is thought to occur in the upper water column. The results indicate that *Pseudoalteromonas* sp., *Idiomarina* sp. and *Alteromonas* sp. actually precipitate barite under experimental conditions. Barite precipitates show typical characteristics of microbial precipitation in terms of size, morphology and composition. Initially, a P-rich phase precipitates and subsequently evolves to barite crystals with low P contents. Under laboratory conditions barite formation correlates with extracellular polymeric substances (EPS) production. Barite precipitates are particularly abundant in cultures where EPS production is similarly abundant. Our results further support the idea that bacteria may provide appropriate microenvironments for mineral precipitation in the water column. Therefore, bacterial production in the past ocean should be considered when using Ba proxies for paleoproductivity reconstructions.

Keywords:

Marine barite, Bacteria, Water column, Microbial processes, Biomineralization.

VI.2. Introduction

Biogenic barium content in marine sediments has been traditionally considered as a reliable proxy for reconstructing past-ocean productivity (Paytan and Griffith, 2007 and references therein). At present, it usually occurs as discrete crystals of the mineral barite in ocean waters underlying high productivity regions. As barite subsequently accumulates in proportion to export production from the surface waters in marine sediments, Ba proxies (barite and algorithms based on Bio-Ba) have served to recognize productivity variations in the past. The settling of particulate Ba has been broadly demonstrated, as has its correlation with organic carbon fluxes and primary production (e.g., Dehairs et al., 1980, 1987, 1991, 1992, 2008; Bishop, 1988; Dymond et al., 1992; Planchon et al., 2013). Moreover, barite is very refractory under oxic conditions; thus, it is well preserved in oxic settings, and dissolution may occur only in anoxic sediments. Since the early discoveries of Ba enrichment in marine sediments (Goldberg and Arrhenius, 1958; Chow and Goldberg, 1960), a vast body of literature on Ba proxies has appeared (e.g., Van Santvoort et al., 1996; Griffith and Paytan, 2012 and references therein). However, the mechanisms behind barite precipitation in seawater that is undersaturated in barite (e.g., Monnin et al., 1999) still remain unknown. Barite-supersaturated microenvironments may result from the decay of organic matter (e.g., Dehairs et al., 1980, 1990; Bishop, 1988; Ganeshram et al., 2003) or the dissolution of acantharian celestite ($SrSO_4$) skeletons, enriched in barium (e.g., Bernstein et al., 1987; Bernstein and Byrne, 2004). Yet the dissolution of acantharians would only contribute up to 20% of the observed barite precipitation, as demonstrated in the Southern Ocean by Jacquet et al. (2007). There is likewise a lack of clear correlation between acantharian abundance and barite concentration in sediments or in the water column (Bertramand Cowen, 1997). To date a direct biologicmediation of barite precipitation by living organisms has not been demonstrated. In this regard, the bacterial precipitation of barite is a promising field to explore. Microbially induced precipitation of barite has been shown in diverse environments; González-Muñoz et al. (2003) were able to precipitate barite in culture experiments with a soil bacterium, and suggested that bacteria may have also played a significant role in barite precipitation in natural environments. Glamoclija et al. (2004), Sanchez-Moral et al. (2004) and Senko et al. (2004) reported barite precipitation in natural environments where microbes may have played a role either oxidizing sulfur compounds to generate sulfate or providing biofilms favoring biomineralization. Bonny and Jones (2007) suggested a re-evaluation of the capability of different microbial groups to mediate barite precipitation, which should be taken into account when using particulate marine barite as a paleoproductivity proxy. In a warm sulfur spring in Canada, Bonny and Jones (2008) reported barite crystals that nucleated on microbial cell surfaces and in microbial extracellular polymeric substances (EPS), suggesting that microbes are capable of adsorbing and bioaccumulating barium. Recently, González-Muñoz et al. (2012) demonstrated that marine bacteria promoted barite precipitation under laboratory conditions. These authors assayed marine bacteria for experimental work, using selected strains with proven biomineralization capability such as *Idiomarina* (González-Muñoz et al., 2008), as well as other strains of Gram-negative and Gram-positive bacteria. Other authors have shown that higher mesopelagic Ba in the Pacific coincides with greater bacterial activity, suggesting a potential relationship (Dehairs et al., 2008). Similarly, Jacquet et al. (2011) describe a correlation between mesopelagic Ba contents and bacterial activity in the Southern Ocean. Additional work is required to fill gaps in our knowledge of barite production in seawater. As it seems to be correlated with enhanced bacterial activity and is produced throughout the mesopelagic layer, natural samples from the Mediterranean at depths of 200 m and 250 m were selected to isolate bacteria for the present study. Our objective was to explore the potential role of marine bacteria living in the upper water column in barite precipitation, as well as the mechanisms involved in such precipitation.

VI.3. Material and methods

VI.3.1. Sampling

Water column samples were obtained from two Mediterranean stations (Fig. 1): the Algero-Balearic basin station RL33CT (37° 44.05′N 2° 12.43′ E) and the Ionian basin station RL81CT (34° 43.00′ N 22° 12.90′ E), from hereon referred to as stations WM (Western Mediterranean) and EM (Eastern Mediterranean) respectively. These samples were collected during the Ristretto e Lungo cruise M83/3 (Research vessel "Meteor") using a CTD (conductivity, temperature and depth) rosette with Niskin bottles of 12 l volume. One water sample was gathered at 250 m depth in the WM station, and another at 200 m depth in the EM station. Sampling depths were selected in view of indications of barite formation in the upper water column. The distribution of barite particles suggests that much of it would come from decaying organic matter aggregates, occurring mainly in the upper water column, where most organic matter is regenerated (e.g., Dehairs et al., 1980).

VI.3.2. Culture media

For marine bacteria isolation and cultivation, "Marine Broth" (MB, DIFCO laboratories, USA) culture medium was used in both liquid and solid forms (jellified with 2% purified agar-agar, DIFCO laboratories, USA). The barite production assay was conducted using solid culture media enrichedwith Ba (CMBa) (González-Muñoz et al., 2012) initially containing no free sulfate so as to avoid inorganic barite precipitation. As barite is thought to precipitate in microenvironments where the Ba concentration may be higher than in seawater, the media were enriched in Ba accordingly. If sulfate were available in such conditions, inorganic precipitation would occur; hence a sulfate free medium is required to test mediating biogenic precipitation. We used two types of CMBa that differed in NaCl concentrations: 1) CMBa1 composed of yeast extract 0.4%, $BaCl_2 \cdot 2H_2O$ 2 mM, NaCl 1.9%, purified agar 2%, pH 7; and 2) CMBa2 composed of yeast extract 0.4%, $BaCl_2 \cdot 2H_2O$ 2 mM, NaCl 3.5%, purified agar 2%, pH 7. As in previous studies of barite precipitation by bacteria (González-Muñoz et al., 2012), two different NaCl concentrationswere used: that of the "Marine Broth" medium as well as the concentration closest to seawater salinity, to further test any potential salinity effect on bacterial growth and precipitation.

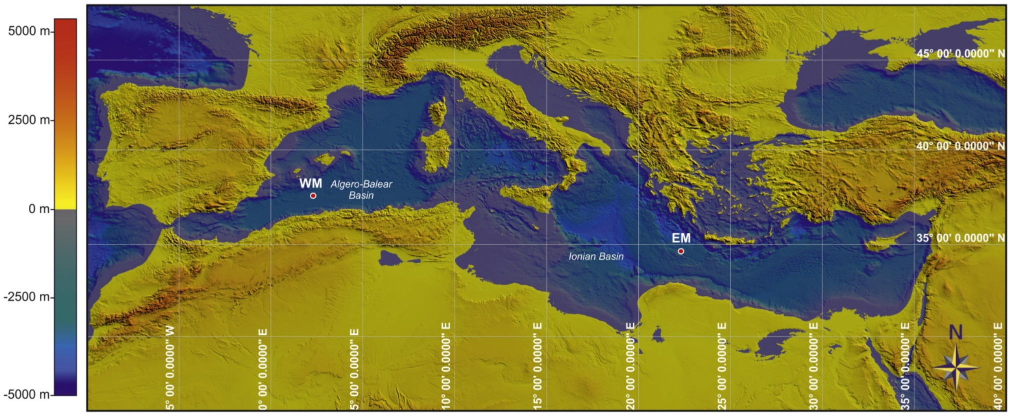

Fig. 1. Bathymetric map of the Mediterranean showing the position of the CTD stations from which the water samples analyzed in this study were recovered (WM in the Algero-Balearic basin, Western Mediterranean, and EM in the Ionian basin, Eastern Mediterranean, respectively). Source map: Amante and Eakins (2009).

VI.3.3. Isolation of bacteria

From each water sample, 100 ml was aseptically filtered using a filter with 0.45 μm pore size (Millipore) in a Kitasato system with a vacuum pump. The filter was recovered with tweezers and gently shaken in 10 ml of NaCl solution (3.5%). Afterwards, appropriate serial dilutions for both samples were performed and 100 μl aliquots of each dilution were inoculated and spread on MB solid medium and incubated at 28 °C; although this is an optimal growth temperature, incubation at a wide range of temperatures has been shown to produce similar results in terms of precipitation (González-Muñoz et al., 2012). Colonies were observed at two time intervals (6 and 20 days), and counted after the incubation to determine colony forming units (CFUs). The selection of colonies was based on visual characteristics such as color, shape and morphology. The purity of selected isolates was confirmed by repeated streaking on the same medium. Pure bacterial suspensions were stored in glycerol at −80 °C.

VI.3.4. Identification of bacteria

Genomic DNA was extracted from bacterial cells using the RealPure Genomic DNA Extraction kit (Durviz, Valencia, Spain), following the manufacturer's instructions. The quantity of DNA was determined employing a Nanodrop spectrophotometer (NanoDrop ND1000, Thermo Fisher Scientific). Additional quality analyses entailed electrophoresis of the extracted DNA (5 µl) on 0.7% agarose gel at 100 V for 40 min, staining with a Gel Red solution (70 µl/l; stock: 10,000x) for 20 min, and visualizing by means of an UVP documentation system (BioRad Transilluminator, Universal Hood; Mitsubishi P93D-printer). To obtain fingerprint patterns of all isolates and cluster them in different groups, repetitive extragenic palindromic-polymerase chain reactions (rep-PCR) were performed using primers REPIR-I and REP2-I (de Bruijn, 1992), as described in detail in Jroundi et al. (2010). PCR products of all isolates were electrophoresed on 1.5% agarose gel and the DNA fingerprints were compared by visual inspection of the banding patterns. The representative strains of each cluster were identified on the basis of their 16S rDNA amplification using the two opposing primers, fD1 and rD1 (Jroundi et al., 2010). PCR products were purified with the Qiagen PCR product purification system and directly sequenced using the same fD1/rD1 primers with an ABI PRISM 3130xI Genetic Analyzer. The sequences were compared with those from the GenBank using the BLASTN tool (Altschul et al., 1997) available at EMBL-EBI database (http://www.ebi.ac.uk) and the EzTaxon-e tool, an extension of the original EzTaxon database (Chun et al., 2007) available at EzBioCloud (http://eztaxon-e.ezbiocloud.net). Phylogenetic analyses were performed by means of the BioEdit computer program (package version 7.1.11; Hall, 2007), which uses the neighbor-joining algorithm, to be visualized with MEGA 4.0.2 (Tamura et al., 2007). The 16S rDNA sequences obtained in this study were deposited in the GenBank database. The accession numbers are shown on the phylogenetic tree (Fig. 2).

VI.3.5. Barite production assays

Representative strains of each rep-PCR group were inoculated in MB liquid culture medium (test tubes with 5 ml/tube) and incubated under shaking (200 rpm) for 24 h at 28 °C. Afterwards, cultures were centrifuged (15,871 xg for 5 min), washed 3 times with a NaCl solution (1.9%, the same concentration as in MB) and finally re-suspended in a 5 ml final volume of the NaCl solution. Drops of 20 µl were inoculated on CMBa1 and CMBa2 solid media and incubated at 28 °C for more than one month. Plates were periodically observed by optic microscopy (4x and 10x objectives) to detect the presence of crystals.

VI.3.6. Analysis of precipitates

– *X-ray diffraction* (XRD): Prior to XRD analyses, bacterial cells were killed by exposition to a germicide UV lamp for 2 h. Portions of the agar containing dead bacterial cells and precipitates were dried for 24–48 h at 37 °C. The mineralogical composition of the precipitates in these portions was determined using an X'Pert Pro diffractometer with an X'Celerator detector. As the sample size was small, a zero background sample holder was used. The diffractograms were interpreted using X-powder software (http://www.xpowder.com; Martín-Ramos, 2004).

– *High Resolution Scanning Electron Microscopy* (HRSEM): For morphological and compositional analyses of the precipitates by HRSEM, agar pieces with dead bacterial cells (treatment with UV for 2 h) were immersed in distilled water and melted in a microwave oven (600 W for 50 s). After centrifugation (30 s at 6261 xg), the precipitates were recovered and washed with distilled water to eliminate the remaining culture medium, salts and cellular debris. Precipitates were transferred to a clean glass plate, dried for 24 h at 37 °C, then prepared and sputtered with carbon for HRSEM observation. Morphological and chemical analyses of the precipitates were performed with an AURIGA model microscope (Carl Zeiss SMT) coupled with energy-dispersive X-ray microanalysis (EDX) and Electron Backscatter Diffraction (EBSD), Oxford Instruments.

VI.4. Results

VI.4.1. Bacterial isolation and identification

The counts of bacterial colonies gave an average bacterial load of (3.6 ± 1.2) x 10^5 CFU/ml in the EM water sample and of (7.19 ± 1.0) x 10^4 CFU/ml in the WM water sample. Different heterotrophic bacteria were isolated from the two water samples.

Rep-PCR grouped the 78 isolates (35 from EM water and 43 from WM water) into 32 REP patterns (Table 1). A phylogenetic tree derived from the nearly complete 16S rRNA gene sequences (Fig. 2) of a representative strain of each REP pattern showed the isolates to pertain to six genera, namely *Marinobacter*, *Alteromonas*, *Pseudoalteromonas*, *Idiomarina*, *Bacillus* and *Brevibacillus*. The 35 isolates recovered from the EM water sample (Ionian basin) mainly belonged to phyla Proteobacteria (62.8%) and Firmicutes (37.2%) (Table 2). Within the Gammaproteobacteria, 15 isolates (42.8%) were affiliated with the genus *Marinobacter* from the family Alteromonadaceae, this being the most abundant genus. The strains were clustered in five rep-PCR groups (IV, V, VI, VII and VIII), and

Marinobacter algicola DG893T was found to be the closest relative species, with similarities ranging from 94% to 99%. Within the same family, the genus *Alteromonas* was represented by seven isolates (20%), which clustered in three rep-PCR groups (I, II and III) showing similarity of over 98% with *Alteromonas stellipolaris* LMG 21861T (groups II and III) and more than 97% with *Alteromonas addita* R10SW13T (group I). More than 34% of the isolates were related to the genus *Bacillus* and they clustered in six rep-PCR groups (X, XI, XII, XIII, XIV and XV), showing similarities with *Bacillus anthracis* ATCC 14578T ranging from 99.85% to 100%. One additional isolate (rep-PCR group IX) was affiliated with the genus *Brevibacillus* from the family Peanibacillaceae (2.6%) and showed a similarity of more than 97% with *Brevibacillus formosus* NRRL NRS-863T. From the WM water sample (Algero-Balearic basin), 43 isolates were identified as members of the same two phyla found in the EM water sample. Within the Gammaproteobacteria, three families dominated: Idiomarinaceae (25.6%), Pseudoalteromonadaceae (20.9%) and Alteromonadaceae (12.9%). One isolate (rep-PCR group IV) affiliated with the genus *Alteromonas* showed more than 98% similarity with the closest relative species, *A. stellipolaris* LMG 21861T. Three rep-PCR groups (V, VI, and VII) were related to the genus *Marinobacter*, the closest relative species being *M. algicola* DG893T. Nine isolates (20.9%) belonged to the genus *Pseudoalteromonas* and clustered in three rep-PCR groups (I, II, and III) showing almost 99.8% similarity with the type strain *Pseudoalteromonas espejiana* NCIMB 2127T. Within the family Idiomarinaceae, the genus *Idiomarina* dominated with three different rep-PCR groups (VIII, IX, and X), and *Idiomarina abyssalis* NCIMB 2127T as the closest relative species, with similarities ranging from 87.7% to 99.3%. Seventeen isolates were highly related to the members of the Firmicutes, *Bacillus* (32.6%) and *Brevibacillus* (7%). The isolates belonging to the genus *Bacillus* clustered in six rep-PCR groups (XII, XIII, XIV, XV, XVI, and XVII), the closest relative species being *B. anthracis* ATCC 14578T with similarities ranging from 99.85% to 100%. One rep-PCR group (XI) was related to the genus *Brevibacillu*s, the closest relative species being *B. formosus* NRRL NRS-863T, having 97% similarity.

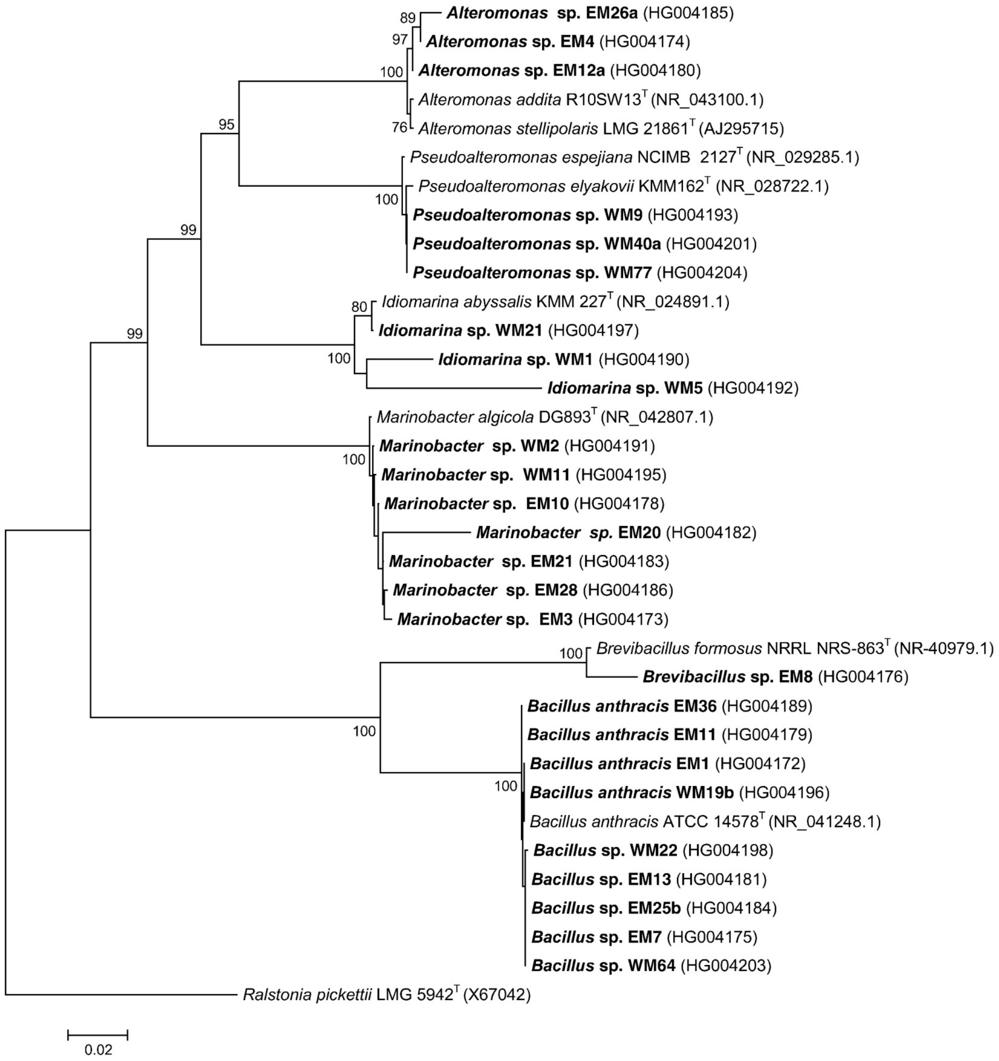

Fig. 2. Neighbor-joining phylogenetic tree based on nearly complete 16S rRNA gene sequences of the studied strains from Mediterranean seawater samples and phylogenetically related species within the genera *Pseudoalteromonas*, *Alteromonas*, *Idiomarina*, *Marinobacter*, *Brevibacillus* and *Bacillus.* The rep-PCR representative strains are in bold letters. Bootstrap values are indicated as percentages derived from 1000 replications. Values lower than 70 are not shown. Bar, 2 nucleotide substitutions per 100 nucleotides. The tree is rooted on *Ralstonia pickettii* LMG 5942^T.

VI.4.2. Barite production assays

No precipitates were observed under optic microscopy among the killed cell controls. In the bacterial colonies of *Pseudoalteromonas* sp., precipitates appeared after the 6th day of incubation (Table 3). They were easily recognized as a concentration of small dense points that were particularly abundant at 15 days and even detectable with the naked eye at one month. This strain showed conspicuous mucus on both solid and liquid culture media, and HRSEM observations also revealed grid-like structures most likely corresponding to EPS (Fig. 3). The isolate *Idiomarina* sp. was able to precipitate barite crystals but they were abundant only after one month of incubation. *Alteromonas* sp. also produced barite precipitates at the border of the colonies but after longer incubation and these precipitates were less abundant. On the other hand, *Brevibacillus* sp. and *Marinobacter* sp. showed low growth on any of the media used in this study (CMBa1 or CMBa2). All isolates related to *B. anthracis* species were eliminated for the sake of biological safety.

VI.4.3. Analysis of precipitates

XRD analysis confirmed that barite is the mineral phase precipitated in the bacterial cultures. Diffractograms also revealed halite as a consequence of desiccating the culture media (Fig. 4). HRSEM showed that barite aggregates were mostly spherical or ellipsoidal in shape, with a size ranging from 0.5 to 2 μm. In general the morphologies resembled previously reported barite that was microbially mediated in culture experiments mostly spherulitic, and occasionally dumbbell-shaped aggregates. As the size of the aggregates reflects the rate of growth, barite aggregates in the bacterial colonies of *Pseudoalteromonas* sp. were larger than those of *Idiomarina* sp. and *Alteromonas* sp. colonies. EDX analyses showed the composition of barium, sulfur and oxygen, although phosphorous was also abundant in some precipitates (Fig. 5). The compositions also generally agree with previous experimental results in terms of P enrichment in the initial phases of crystallization (González-Muñoz et al., 2012). Thus, larger spheroidal aggregates had more abundant Ba and S as compared to P. In contrast, smaller and less compact aggregates had relatively high P content with respect to S and Ba.

Table 1. Phylogenetic classification of marine strains identified in this study.

Strains[a]	Rep-PCR pattern	Closest relative species on basis of 16S rRNA gene	Accession numbers c the closest type strains	Similarity %	Family	Class	Phylum
Algero-Balearic basin water sample (WM)							
WM9, WM8b, WM19a	I	*Pseudoalteromonas espejiana*	NR_029285.1	99.78	Pseudoalteromonadaceae	Gammaproteobacteria	Proteobacteria
WM40a, WM57, WM40b, WM35	II	*Pseudoalteromonas espejiana*	NR_029285.1	99.78	Pseudoalteromonadaceae	Gammaproteobacteria	Proteobacteria
WM77, WM16	III	*Pseudoalteromonas espejiana*	NR_029285.1	99.78	Pseudoalteromonadaceae	Gammaproteobacteria	Proteobacteria
'WM32b	IV	*Alteromonas stellipolaris*	NR_025433.1	98.69	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
WM2	V	*Marinobacter algicola*	NR_042807.1	98.1	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
WM11, WM7, WM10	VI	*Marinobacter algicola*	NR_042807.1	99.11	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
[2]WM48	VII	*Marinobacter algicola*	NR_042807.1	98.57	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
WM1, WM46, WM49, WM56	VIII	*Idiomarina abyssalis*	NR_024891.1	95.74	Idiomarinaceae	Gammaproteobacteria	Proteobacteria
WM5, WM6a, WM14a, WM8a	IX	*Idiomarina abyssalis*	NR_024891.1	87.69	Idiomarinaceae	Gammaproteobacteria	Proteobacteria
WM21, WM30, WM17a	X	*Idiomarina abyssalis*	NR_024891.1	99.38	Idiomarinaceae	Gammaproteobacteria	Proteobacteria

[3]WM3, WM31a, WM31b	XI	*Brevibacillus formosus*	NR_40979.1	97.04	Paenibacillaceae	Bacilli	Firmicutes
[4]WM17b, WM27, WM6b	XII	*Bacillus anthracis*	NR_041248.1	99.92	Bacillaceae	Bacilli	Firmicutes
[5]WM13	XIII	*Bacillus anthracis*	NR_041248.1	99.92	Bacillaceae	Bacilli	Firmicutes
[6]WM14b, WM29	XIV	*Bacillus anthracis*	NR_041248.1	100	Bacillaceae	Bacilli	Firmicutes
WM19b, WM20, WM4a, WM4b	XV	*Bacillus anthracis*	NR_041248.1	99.92	Bacillaceae	Bacilli	Firmicutes
WM22, WM25, WM24	XVI	*Bacillus anthracis*	NR_041248.1	99.85	Bacillaceae	Bacilli	Firmicutes
WM64	XVII	*Bacillus anthracis*	NR_041248.1	99.85	Bacillaceae	Bacilli	Firmicutes
Ionian basin water sample (EM)							
EM26a, EM26b, EM27	I	*Alteromonas addita*	NR_043100.1	97.73	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
EM4, EM67	II	*Alteromonas stellipolaris*	NR_025433.1	98.69	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
EM12a, EM66b	III	*Alteromonas stellipolaris*	NR_025433.1	98.69	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
EM3, EM5	IV	*Marinobacter algicola*	NR_042807.1	98.77	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
EM10, EM9b	V	*Marinobacter algicola*	NR_042807.1	98.57	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
EM20, EM18, EM17a	VI	*Marinobacter algicola*	NR_042807.1	94.01	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
EM21, EM22	VII	*Marinobacter algicola*	NR_042807.1	99.04	Alteromonadaceae	Gammaproteobacteria	Proteobacteria

EM28, EM29, EM65, EM66a, EM31, EM44	VIII	*Marinobacter algicola*	NR_042807.1	98.70	Alteromonadaceae	Gammaproteobacteria	Proteobacteria
EM8	IX	*Brevibacillus formosus*	NR_40979.1	97.04	Paenibacillaceae	Bacilli	Firmicutes
EM1	X	*Bacillus anthracis*	NR_041248.1	100	Bacillaceae	Bacilli	Firmicutes
EM7	XI	*Bacillus anthracis*	NR_041248.1	99.85	Bacillaceae	Bacilli	Firmicutes
EM11	XII	*Bacillus anthracis*	NR_041248.1	99.92	Bacillaceae	Bacilli	Firmicutes
EM13, EM14	XIII	*Bacillus anthracis*	NR_041248.1	99.85	Bacillaceae	Bacilli	Firmicutes
EM25b, EM30, EM33, EM2	XIV	*Bacillus anthracis*	NR_041248.1	99.85	Bacillaceae	Bacilli	Firmicutes
EM36, EM48, EM32	XV	*Bacillus anthracis*	NR_041248.1	99.92	Bacillaceae	Bacilli	Firmicutes

[a] For a given strain, the letters WM and EM stand for Western Mediterranean and Eastern Mediterranean respectively, the names of the locations from which water samples were collected. Strains in bold were chosen as representative strains of each rep-PCR group. 1 to 6: The representative strains of these groups are EM4, EM10, EM8, EM36, EM11 and EM1, respectively.

Table 2. Number of isolates (N) related to the corresponding genera and their percentages in each water sample.

Genus	Algero-Balearic basin water sample (WM)		Ionian basin water sample (EM)	
	N	%	N	%
Alteromonas	1	2.3	7	20
Pseudoalteromonas	9	20.9	0	0
Idiomarina	11	25.6	0	0
Marinobacter	5	11.6	15	42.8
Brevibacillus	3	7	1	2.9
Bacillus	14	32.6	12	34.3
Total of 6 genera	43	100	35	100

Table 3. Bacterial growth on both CMBa culture media (CMBa1 and CMBa2) and barite production by the tested strains.

Strains	Growth		Barite production	
	CMBa1	CMBa2	CMBal	CMBa2
Alteromonas sp. EM4, EM12a and EM26a	++	++	P	P
Pseudoalteromonas sp. WM9, WM40a and WM77	++	++	P	P
Idiomarina sp. WM1, WM5 and WM21	++	++	P	P
Marinobacter sp. WM2, WM11, EM3, EM10, EM20, EM21 and EM28	+	+	–	–
Brevibacillus sp. EM8	+	+	–	–

+: Low growth; ++: Moderate growth; P: Crystal production; –: Unknown.

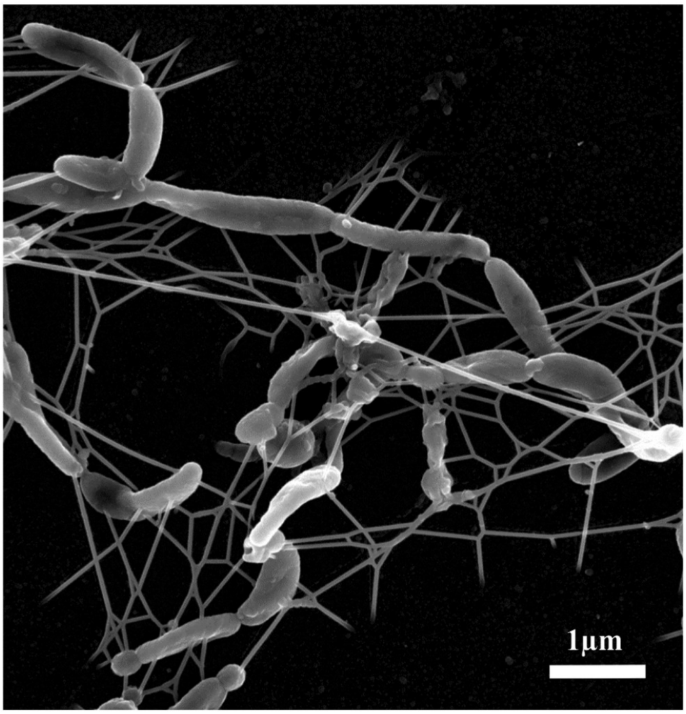

Fig. 3. Scanning electron micrographs (backscattered) showing cells of *Pseudoalteromonas* sp. strain WM40a that present abundant grid-like structures, most likely corresponding to EPS. Samples were prepared using the CO_2 critical point method.

VI.5. Discussion

The precipitation of barite under laboratory conditions in association with bacteria isolated from natural seawater represents a step forward towards demonstrating the significance of bacteria in the precipitation of barite in natural environments. Laboratory experiments (e.g., Bonny and Jones, 2008; González-Muñoz et al., 2003, 2012) and field observations (e.g., Bonny and Jones, 2007 and references therein) have revealed that the precipitation of barite can be microbially mediated. Furthermore, our experimental work showed that the sizes, morphologies, and presence of high P concentration at the initial stages of crystallization are consistent with microbial precipitation (Fig. 5). In fact, the formation of P-rich precursors is a common step in bacterial biomineralization. Nucleation of an amorphous phosphate phase has been previously reported for the microbial precipitation of barite (González-Muñoz et al., 2003, 2012), aragonite, apatite and iron oxides, both under laboratory conditions and in natural environments (Rivadeneyra et al., 2010 and references therein). As suggested by González-Muñoz et al. (2012), phosphoryl groups in structural polymers as EPS or cell membranes may act as sorbing constituents during the precipitation process, resulting in amorphous or poorly crystalline P-rich precursors. Our results show that in precipitates from *Idiomarina* sp. and *Alteromonas* sp. which appeared later than those from *Pseudoalteromonas* sp. and where smaller aggregates were observed, P is particularly abundant in comparison to S. This further suggests that crystals produced at initial stages have higher P contents. Moreover, marine barite is usually found in oceanic sinking aggregates in association with decaying organic matter (e.g., Bishop, 1988; Dymond et al., 1992; Dehairs et al., 2000), and in suchmicroenvironments a high bacterial activity is expected from organic matter degradation. A correlation between higher mesopelagic Ba and bacterial activity has been demonstrated in several regions from the Pacific and the Southern Oceans. It has been proposed that mesopelagic particulate biogenic Ba reflects bacterial degradation of organic matter, and is related to oxygen consumption and bacterial carbon respiration (Dehairs et al., 2008; Jacquet et al., 2011; Planchon et al., 2013). Thus, the precipitation of barite in association with bacteria isolated from natural seawater, at depths where particulate Ba is more abundant, further points to a connection between high bacterial activity and marine barite formation. In terms of the phylogenetic groups retrieved in this study, our results are not very different from those obtained in offshore oligotrophic waters from the Mediterranean Sea, and from the Pacific and Atlantic oceans. The rep-PCR fingerprint technique, used in this study, has enough sensitivity to accurately detect genotype microbes at the strain or subspecies levels. This is a valuable tool used to group, identify, track and examine diversity among environmentally important microorganisms (Versalovic et al., 1991, 1998; Sadowsky and Hur, 1998; Rademaker et al., 2004; Healy et al., 2005; Ishii et al., 2009). Bacteria with the same rep-PCR pattern belong to the same bacterial species or even the same strain (de Bruijn, 1992; Ishii and Sadowsky, 2009), while the same species with different fingerprint patterns indicate that the isolates belong to different strains from the same species. The bacteria isolated from the two areas of our study showed similar phylogenetic distributions and taxonomic resolutions: the communities were dominated by Gammaproteobacteria (60.4% in the western sample and 62.8% in the eastern sample), although members of Firmicutes (39.6% in the western sample and 37.2% in the eastern sample) were also abundant. These results confirm that these bacteria are common in seawater from different oceans (Sawabe et al., 2000; Van Trappen et al., 2004; Ivanova et al., 2005; López-López et al., 2005; Green et al., 2006; Keim et al., 2006; Ki et al., 2009; Porob et al., 2013; Lekunberri et al., 2014). The genus *Bacillus*, within the Firmicutes, is one of the most frequently isolated from marine waters (Eilers et al., 2000). The sequences found here matched very well with *B. anthracis*, though this species belongs to the *B. anthracis* group (*B. anthracis*, *Bacillus cereus*, *Bacillus thuringiensis* and *Bacillus toyonensis*), a taxonomic group that includes species/subspecies that are not distinguishable by the 16S rRNA sequence. The same observation was made by Ki et al. (2009), who isolated several marine *Bacillus* species from diverse marine environments but failed to separate, on the basis of 16S rRNA sequences, the species of *B. cereus* group in individuals. Accordingly, discerning the members of *B. anthracis* group found here would call for further studies. The genus *Brevibacillus* is related to the genus *Bacillus*, which is distributed widely throughout nature. The habitat of both genera overlaps, occurring in diverse environments such as rocks, dust, aquatic environments, and guts of various insects and animals (Nicholson, 2002). Gammaproteobacteria represent a large phylogenetic group of cosmopolitan species (Bowman et al., 1997; Pinhassi et al., 1997; Suzuki et al., 1997; Eilers et al., 2000). Our results are consistent with those of Lekunberri et al. (2014), who used culture-dependent methods to show that this group of bacteria dominated in the waters in the NW Mediterranean Sea (Blanes Bay Microbial Observatory, approximately 70 km north of Barcelona). Mullins et al. (1995) found that the Gammaproteobacteria subclass contains the largest number of sequences from cultured marine isolates. In our study, this is reflected by the high degree of similarity between our sequences and those of members of the genera *Alteromonas*, *Idiomarina*, *Marinobacter* and *Pseudoalteromonas*. The occurrence of these genera could be an effect of the peculiar depth conditions of the chlorophyll maximum (DCM), with a much higher abundance of phytoplankton and perhaps a higher availability of organic nutrients; or it could signal a change in physical conditions, e.g. water temperature and light intensity. The isolates representing *Marinobacter* sp. appear to branch at some distance from the other Gammaproteobacteria (Fig. 2) and are related to halotolerant bacteria. The genus *Idiomarina* has a widespread distribution in marine and hypersaline habitats (Martínez-Cánovas et al., 2004), and different species were isolated from the Pacific Ocean deep-sea (Ivanova et al., 2000), hydrothermal vents at a depth of 1296 m (Donachie et al., 2003), surface water of the central Baltic Sea (Brettar et al., 2003), hypersaline water in Spain (Martínez-Cánovas et al., 2004), and wetland and seashore sand in Korea (Choi and Cho, 2005; Kwon et al., 2006). Recently, several species of this genus have served to demonstrate the capability of marine bacteria to precipitate barite (González-Muñoz et al., 2012). The predominant occurrence of *Alteromonas* and *Pseudoalteromonas* spp. in marine waters all around the world had been suggested previously (Acinas et al., 1999; Eilers et al., 2000; Schäfer et al., 2000; Lekunberri et al., 2014). In particular, *Alteromonas* species were abundant in Mediterranean offshore samples, in the surface and down to 400 m (Acinas et al., 1999). This was supported by our rep-PCR DNA fingerprints and further identification on the basis of the 16S rDNA, as we recovered *Alteromonas* sp. from both eastern and western Mediterranean waters at respective depths of 250 m and 200 m. Other authors describe the significant presence of this marine organism in samples from the Atlantic (Klochko et al., 2012), and in many other tropical and temperate seas (García-Martínez et al., 2002). The genus *Pseudoalteromonas* is common to the marine environment, including seawater, algae, and marine invertebrates (Baumann et al., 1972, 1984; Baumann and Baumann, 1981; Gauthier and Breittmayer, 1992; Sawabe et al., 2000). It was described for the first time by Gauthier et al. (1995) as a heterotrophic, Gram-negative, aerobic, rod-shaped, and polarly flagellated marine bacteria. All pseudoalteromonads require Na+ ions for growth (Mikhailov et al., 2006), and could therefore be regarded as obligate marine microorganisms. The sequences found herein matched very well with *P. espejiana*, characterized by non-pigmented, translucent and highly mucoid colonies, all of which confirm the phenotypic characteristics of this microorganism described previously by a number of authors (Chan et al., 1978; Bowman, 1998). Some members of the genus *Pseudoalteromonas* are highly effective in biofilm formation (Saravanan et al., 2006; Huang et al., 2007; Iijima et al., 2009) and accordingly might be hosted by marine fauna and flora for defense, by means of biofouling (Lindquist and Fenical, 1991; Holmström et al., 1996, 2002; Dobretsov et al., 2006, 2007). Interestingly, several bacterial strains of the genus

Pseudoalteromonas, including *P. espejiana*, commonly produce EPS. Itwould appear that this characteristic provides a range of survival strategies for the cells. This EPS acts as protective barriers against antibiotics and against predation by protozoa (Costerton et al., 1987), reduces the diffusion of some substances to and from the cells, and functions as enhancers for nutrient and ion uptake (Geesey, 1982). EPS production would therefore play a key role in barite precipitation, as is the case for other mineral phases (Bontognali et al., 2014; Tourney and Ngwenya, 2014). Ba would bind to phosphate groups on EPS and bacterial cells, locally increasing its concentration (González-Muñoz et al., 2012). As a second step, the replacement of phosphate by sulfate occurs. Under experimental conditions, bacteria also oxidize sulfur compounds and generate the sulfate needed for barite precipitation. However, in seawater, where sulfate is available, the major role played by bacteria would be providing binding sites for cations in the EPS and cell membranes. It is also important to note that living organisms such as plankton contain a relatively large pool of labile Ba, which is readily released during decomposition, as demonstrated by Ganeshram et al. (2003), who also suggested that Ba enrichment rather than sulfate induced barite precipitation. Thus, in natural seawater where sulfate is available, bacteria could contribute to Ba enrichment and saturation in microenvironments where barite may precipitate. As the assayed bacterial strains are common and abundant in seawater, it is likely that bacteria promote Ba saturation in natural decomposing organic matter microenvironments. Barite formation may involve the Ba binding to phosphate groups on EPS and bacterial cells, the increasing of Ba concentrations at nucleation sites followed by the replacement of phosphate by seawater sulfate and barite crystal growth. It is known that organic matter degradation, particularly in sinking organic aggregates, is linked tomicrobial activity. Thus, bacterial production would significantly impact barite precipitation, and this would be a major process to take into account when using Ba proxies for paleoceanographic reconstructions.

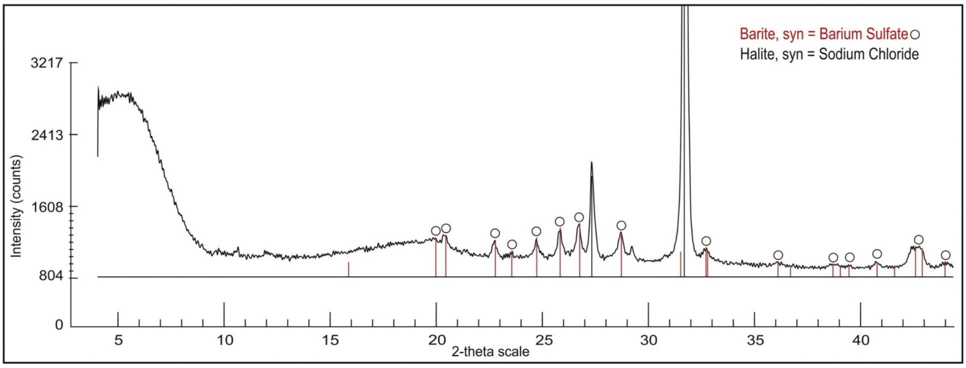

Fig. 4. X-ray diffraction diagram of the precipitates formed in culture experiments by *Pseudoalteromonas* sp. (sample WM40a); barite peaks are indicated with red bars. Abundant halite precipitates as a result of desiccating the medium.

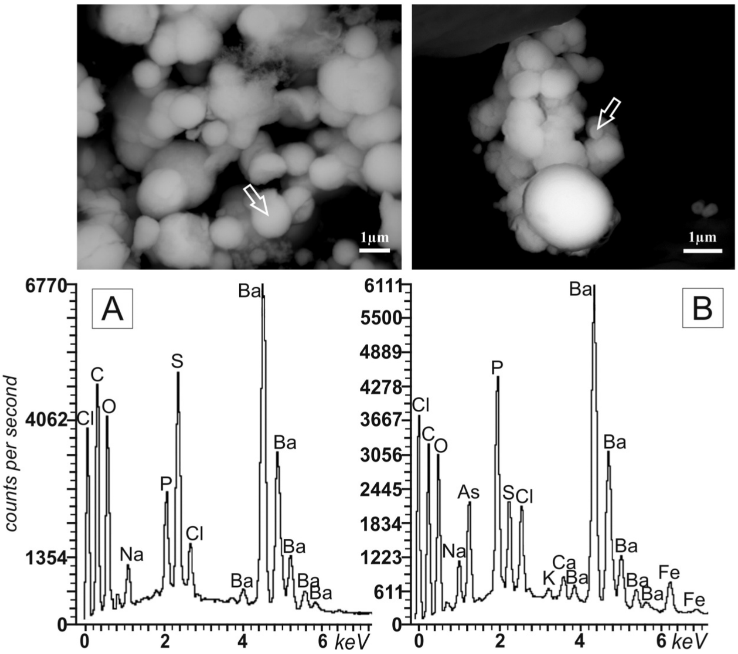

Fig. 5. Scanning electron micrographs (backscattered images) showing typical morphologies and sizes of the barite precipitates (*Pseudoalteromonas* sp.WM40a), and acquired energy-dispersive X-ray spectroscopy spectra: A) from one large spherical aggregate that shows relative abundance of sulfur in relation to phosphorous; B) from one small aggregate, probably corresponding to initial stages of crystallization, that shows a significantly higher phosphorous concentration (see arrows).

VI.6. Conclusions

Several bacterial strains isolated from natural Mediterranean sub-surface seawater, at 200 m and 250 m depths, promoted barite precipitation under laboratory conditions. Our results show that barite precipitates are particularly abundant in cultures where EPS production is also high. In fact, EPS production and bacterial growing ability under experimental conditions would appear to affect the rates of barite production. Therefore, the role of EPS in barite precipitation in the water column is an important issue for future research. Microbially induced precipitation of barite under laboratory conditions triggered by bacteria that are common in marine seawater suggests that bacterially induced precipitation may also occur in the oceans. Therefore, bacterial production in the past and the mechanisms involved in barite precipitation should be considered in paleoproductivity reconstructions.

VI.7. Acknowledgments

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Chapter VII. General discussion

Barium is ubiquitous in the water column in areas of high biological productivity where organic matter decomposition takes place. Although the biological mechanisms that retain barium in such microenvironments are not well understood, it is well known that the formation of marine barite occurs due to biological intermediation leading to Ba saturation. In the Mediterranean, the precipitation of marine barite in the water column and its subsequent deposition has caused the accumulation of this mineral in the underlying organic-rich sediments, the so-called sapropels. As previously mentioned, in this thesis sapropels were studied from Pliocene to Holocene ages, and a review of climate and productivity conditions leading to sapropel deposition over time was carried out. Ba distribution in the water column across a transect within the Mediterranean Sea has also been analyzed, and the mechanisms for marine barite formation have been investigated, in particular, focused on the role that marine bacteria play in barite precipitation within high productivity environments.

VII.1. Ba distribution in Mediterranean Sea sapropels: Climate and productivity.

As the Ba cycle is closely related to primary productivity, assessing the mechanisms controlling past productivity variations relies on the further understanding of Ba concentrations both in the water column and in sediments. Indeed, the importance of marine barite within sapropels as a proxy has been demonstrated in many paleoproductivity studies (e.g., Dehairs et al., 1996; Jilbert et al., 2010; Hennekam et al., 2014; Martínez-Ruiz et al., 2018). At present, the eastern Mediterranean basin shows high Ba concentrations in surface waters linked to lower Ba uptake, thus lower productivity than in the western basin. Still, the discrete productivity of the present Mediterranean Sea is maintained because of the density increase of the surface water masses eastward due to evaporation, and their sinking in the easternmost part; this leads to nutrient and oxygen distribution from surface waters towards the intermediate layer, just below, with the subsequent productivity improvement. At present, the intermediate layer travels westward, passing through the Sicily strait, and in the western basin becomes a confined water mass because the less dense Atlantic inflow generates strong stratification, which provokes poor water ventilation. In this way hydrodynamics affect the basin´s productivity, as is reflected in the Mediterranean sedimentary record. However, the present framework of biological productivity cannot be extrapolated to other time periods with higher marine productivity; for instance the most recent Holocene sapropel (S1) in the Alboran basin has 535 ppm of barium, while in the Levantine basin the concentrations are up to 1003 ppm for the same period (Chapter IV, Table 4). One exception is the Erathostenes Seamount (avg. 226 ppm), because its shallow nature does not allow much barite accumulation, yet it is in line with values observed for other seas such as the Arabian Sea (Schenau et al., 2001). Primary productivity depends on several factors, including the solar energy received, temperature, CO_2 concentrations, nutrient availability, and community interactions. All these factors are in turn linked to climate changes. The first sapropels were deposited roughly 15.4 My ago (Taylforth et al., 2014), when the Mediterranean basin became a semi-enclosed system and buoyancy changes began to affect water masses layering. Since then, sapropels have been deposited during glacial and also during interglacial periods. During the glacial periods, abrupt climatic oscillations occurred as a consequence of changes in the oceanic circulation, i.e. the warm cycles Dansgaadr-Oeschger during the last glaciation (110000-10000 years BP). The transitions between glacial and interglacial periods were fast, and there were also abrupt setbacks such as the Younger Dryas (13000-11600 years BP). The warm interglacial periods were more stable than the glacial periods, with some smaller scale variations. The Riss glaciation began 200000 years BP and coincides with the deposition of sapropels S8 (i-cycle 20), S7 (i-cycle 18) and S6 (i-cycle 16). Then, 145000 years ago marked the onset of an abrupt and short warming until 125000 years ago, coinciding with the deposition of sapropel S5 (i-cycle 12). The S5 was triggered by a productivity increase because of higher nutrient input by riverine, faster Atlantic inflowing waters together with induced upwelling by westerlies (Moreno et al., 2004). Around that point a gradual and slow cooling began, until 110000 years BP, a period during which cold summers were common. There was a stabilization of temperatures, and sapropels S4 (i-cycle 10) and S3 (i-cycle 8) were deposited. Afterward, 60000 years ago, the last glaciation of the Quaternary era (Würm) was initiated and finished 18000 years ago, when the characteristic warm period of the current Holocene began. Roughly 8000 years BP the last great ice age was almost finished, coinciding with the deposition of sapropel S1 (i-cycle 2), triggered by sea level rise that created a connection between the Black Sea and the Mediterranean Sea (Rohling et al., 2015). There are no differences between glacial and interglacial sapropels, because an important requirement for the formation of these sediments is the extra freshwater input into the Mediterranean basin, either by monsoon intensification or by sea level rise during the deglaciation, giving rise to similar conditions for sapropel deposition in terms of nutrients and oxygenation. Sapropels are commonly formed during precession minima (Hilgen et al., 1993, 1995; Lourens et al., 1996, 2001), when perihelion coincides with summer in the northern hemisphere, enhancing summer insolation and therefore the contrast between seasons. This intensifies the summer monsoon, although not all the precession minima developed sapropels. Regarding deglaciation, the buoyancy increased Mediterranean seawater layering, leading to the poor oxygen bottom conditions that are required for organic matter preservation. In short, regardless if sapropels deposited during glaciar or interglaciar periods, the factors influencing the carbon content in sediments are mainly productivity, oxygenation (thus buoyancy leading to water layering), physiography, marine currents and water mass residence time. Both productivity and preservation are the most significant factors, being more directly related to sapropel formation. During the Miocene and Pliocene, productivity and anoxic conditions seem to be high and constant (Menzel et al., 2003, Gallego-Torres et al., 2011; Vasiliev et al., 2019). On the basis of the global $\delta^{18}O$ record of benthic foraminifera (Lisiecki and Raymo, 2005), the Earth's climate has been strongly controlled by obliquity forcing (41 kyr) during the late Pliocene to early Pleistocene. The deep water circulation in the eastern basin was slower during the Pliocene and Early Pleistocene, with the subsequent restriction of water ventilation and therefore better preservation (Gallego-Torres et al., 2011), whereas productivity was somewhat more relevant than preservation during the Late Pleistocene and Holocene (De Lange et al., 2008; Rohling et al., 2015, Van Helmond et al., 2015, Zwiep et al., 2018). On the other hand, more saline water output through the Gibraltar Strait into the Atlantic Ocean might influence the global oceanic circulation strength, as proposed for the Bering Sea by other authors (Hu et al., 2010). All the productivity variations and ventilation changes are well reflected by the Ba record in the Mediterranean Sea. Although from Late Pleistocene to present productivity seems to be the main factor for sapropels deposition, there is a generalized carbon decreasing trend towards the present that could be attributed to decreasing productivity from past to present, supported by the Ba data presented here (Chapter IV: Figure 2, Tables 2, 3 and 4). As for the spatial Ba distribution, physiography can be evoked as an important conditioning factor; for Pleistocene and Holocene sapropels, the highest Ba concentrations are observed in the Mediterranean Ridge, regardless of the high sedimentation rates diluting Ba during the Late Pleistocene. This highlights the exceptional productivity during a period coincident with an insolation maximum enhancing the monsoon effect, leading to higher riverine nutrient input in this basin (Rohling et al., 2015). The high freshwater input also enhanced changes in buoyancy that produced stratification and oxygen depletion, aiding a good preservation of sapropels.

VII.2. Ba distribution in the water column

In the Mediterranean Sea, the dissolved Ba concentration increases from the inflowing Atlantic surface water (avg. 8.3 µg/l) to the intermediate (avg. 9.1 µg/l) and deep (avg. 10.3 µg/l) waters. This Ba export to the intermediate and deep Mediterranean water bodies occurs through convection during winter involving an intense Ba cycling with barite dissolution in deep waters (Jullion et al., 2017; Roy-Barman et al., 2019). The lateral Ba advection is also reflected in its concentration in each basin along the Mediterranean Sea. The data of this work indicate that in the western Mediterranean (stations 1 to 6) the dissolved Ba average in surface waters is 7.5 µg/l and increases eastward until an average of 7.8 µg/l in the eastern basin (stations 12 to 14), while the intermediate and deep waters are Ba enriched westward; from 8.5 µg/l (stations 12 to 14) to 9.5 µg/l (stations 1 to 6) in the mesopelagic layer and from 9.8 µg/l (stations 12 to 14) to 10.5 µg/l (stations 1 to 6) in the deep water masses. The obtained results are in agreement with data previously reported (Jacquet et al., 2015, 2020; Roy Barman et al 2019). Evaporation by itself cannot account for the increase of Ba from the Atlantic inflow to the deep waters (roughly 30% if we take into account the data of the water masses averages) because the salinity increase accounts only for 8% (Roy-Barman et al., 2019). In general, the vertical dissolved Ba gradient is higher than the lateral variations through the whole basin. This is due to the close link between the distribution of particulate and dissolved Ba in the water column and organic matter degradation (Jacquet et al., 2020). Although the current Mediterranean Sea is oligotrophic, Roy-Barman et al. (2019) has pointed out that the biological particulate Ba cycling in the Mediterranean Sea is more intense than previously estimated by other authors (Dehairs et al., 1987; Jacquet et al., 2016). The high dissolved Ba concentrations in deep waters can also be attributed, in part, to the residence time of seawater masses and the subsequent low water ventilation, increasing Ba content after several cycles. But the relatively modest residence time of the Mediterranean seawater masses compared to other oceans makes it necessary to invoke other Ba sources to explain the unbalance in the basin. To date, the most complete Ba budget scheme has been proposed for the western Mediterranean by Roy-Barman et al. (2019), who concluded that both Saharan dust and river inputs are not enough to explain the Ba distribution in the Mediterranean Sea, which could be explained by other processes previously ignored such as the submarine groundwater discharge or Ba diffusion from Messinian evaporites. The importance of submarine groundwater discharge as Ba source in coastal waters has been demonstrated over the world and it has been shown that in some cases the groundwater input of Ba appears to be much more important than the river input (e.g., Shaw et al., 1998). This is still an open question and further research is needed as well as more data from rivers since the Ba contribution could have also been underestimated. On the other hand, there is another factor that has been overlooked regarding the assessment of the Ba budgets in the current Mediterranean Sea, the anthropogenic contamination. Anthropogenic activity has introduced major amounts of heavy metals into the oceans (Fowler et al., 1987; Von et al., 1987; Bethoux et al., 1990). In most cases, the most significant contribution of anthropogenic metals to a marine coastal area is of terrestrial origin, such as mining and other industrial activities near rivers and estuaries (Chow et al., 1978; Morton and Blackmore, 2001; Carman et al., 2007; Karageorgis and Kaberi, 2005; El-Sorogy et al., 2016; Tornero and Hanke, 2016; Lu et al., 2018). Although the Ba input from land is only relevant in coastal areas (Singh et al., 2013), drilling fluids and produced waters associated with shale gas exploration represent important anthropogenic sources of Ba contamination (Harper, 2008; Kargbo et al., 2010; Payne et al., 2011). In the Eastern Mediterranean gas and oil explorations have been conducted for many decades (Kok and Merey, 2014), and therefore the present and future potential contamination can be important. Furthermore, most coastal areas of the Mediterranean were found to discharge untreated sewage to the sea during the seventies. The Ebro coast in Spain (Albaigés et al., 1984; Fowler, 1985), the French and the Italian Rivieras (Padan et al., 2016) and the Egypt coastal waters (Martin et al., 1989, Guieu et al., 1991, Dorten et al., 1991, Palanques and Diaz, 1994, Roussiez et al., 2006) are considered the most polluted areas in the Mediterranean Sea. The anthropogenic signal is notably evidenced in most of the Mediterranean near-shore sediments as well as in some deeper parts of the Mediterranean (Nolting and Helder, 1991, Fernex et al., 1992, 2001; Marin, 1998; Ferrand et al., 1999; Miralles et al., 2006; Palanques et al., 2008; Martin et al., 2009). Indeed, the anthropogenic impact on an industrialized coastal marine area in Sicily (Italy) has recorded a high anthropogenic disturbance of sediments by Ba and other contaminants (Romano et al., 2020). Furthermore, the maritime traffic increasing trend elevates the risk of pollution caused by shipping (Tournadre, 2014), whose most released substances during 1978 – 2013 in European waters included barium (Tornero and Hanke, 2016). A proof of the contaminants released to the seas has been recorded in North-Eastern Atlantic deep-sea fish (Mormede and Davies, 2003) as well as in fish larvae from coastal areas of the Black Sea (Niemiec et al., 2018). Given this fact, it would be appropriate to consider anthropogenic contamination or at least to further research its role in the Ba inputs to the Mediterranean Sea. On the whole, the study of the Ba distribution in the Mediterranean Sea has a global significance since its hydrodynamics has an impact on the global thermohaline circulation and this basin is considered a natural laboratory to further investigate processes occurring on a higher scale. Overall, the investigations of the Ba distribution in the water masses of the Mediterranean have been related to productivity reconstructions, since particulate barium records have been used for such reconstructions. The global ocean is generally undersaturated concerning barite (Monnin et al., 1999; Monnin and Cividini, 2006) and the Mediterranean is particularly undersaturated, thus Ba enrichment in marine sediments has served to support barite precipitation associated with organic matter decay in the mesopelagic zone as reported in the open ocean (Dehairs et al., 1980, 1997, 2000; Collier and Edmond, 1984; Bertram and Cowen, 1997; Bishop, 1988; Ganeshram et al., 2003).

VII.3. Bacterial role in barite precipitation

Investigating the bacterial role in barite precipitation is one of the main objectives of this thesis. This hypothesis was built on previous studies of bacterial barite production by the soil strain *Myxococcus xanthus 422* (González-Muñoz et al., 2003) as well as by diverse marine strains from the genus *Idiomarina* and the *Marinobacter hydrocarbonoclasticus* and *Planomicrobium okeanokoite* species (González-Muñoz et al., 2012). As this thesis focuses on the Mediterranean, the barite production assays were carried out with bacteria from seawater in an aim to demonstrate the capability of bacteria from natural environments to precipitate barite. Most of the isolated strains demonstrated this capacity, specifically the strains belonging to *Alteromonas*, *Pseudoalteromonas* and *Idiomarina* genera grew properly in the culture media MB and CMBa and produced abundant barite (this thesis, Chapter VI). In other cases (e.g., *Marinobacter* and *Brevibacillus* genera) we could not confirm their ability as they did not show significant growth in the tested media, at least in the specific conditions of our experiments; yet this does not make these bacteria exempt of such a capacity under other conditions. The obtained results are of great interest in that they evidence that all strains exhibiting good growth in the media MB and CMBa produce barite, highlighting the production capacity of this mineral by phylogenetically different bacteria. The experiments performed further support the results of González-Muñoz et al. (2003, 2012), evidencing that for barite precipitation, Ba initially binds to phosphate groups from the cell walls and EPS, and Ba phosphate eventually evolves to barium sulphate. The assays also showed that well developed barite crystals have a relatively higher S content than the initial amorphous phases, which are richer in P. This finding was further confirmed by Martínez-Ruiz et al. (2018, 2019, 2020) in both experimental and natural conditions. The substitution of the phosphate anions by sulphates —phosphates being an amorphous precursor phase— has likewise been demonstrated for example in carbonates (Rinaudo et al., 1994; Rivadeneyra et al., 2010). The PO_4^{2-} and SO_4^{2-} anions are tetrahedral, they have similar electronegativities, the same valency, and, both have similar sizes (sulphates being only slightly larger), so there is enough space in the barite lattice to allow for that substitution, making solid solution possible (Pinto et al., 2008; Alshemary et al., 2013). This substitution of the phosphate groups by sulphates occurs frequently in nature, even in abiotic processes

(Smirnov et al., 2017; Tran et al., 2017; Ruiz-Agudo et al., 2020). Another important aspect that our experiments demonstrate is the important role of EPS in barite precipitation. In the present thesis (Chapter VI) it has been demonstrated that higher EPS production is linked to enhanced barite precipitation in bacterial cultures. Different works (Obst et al., 2009; Warren et al., 2001; Bontognali et al., 2006, 2013; Krause et al., 2012; Zhu and Dittrich, 2016; Petrash et al., 2017; Al Disi et al., 2019) have demonstrated that EPS can enhance mineral precipitation of carbonates, since these substances act as a template forming nucleation sites, even in an undersaturated carbonate environment; furthermore, carbonates precipitating on the cell surface are dependent on the culture growth phase. In the specific case of the bacteria used in this thesis, the role of EPS production is similarly demonstrated, for instance by *Pseudomonas* (see Chapter VI, Figure 3). The role that bacteria and EPS production play in marine environments enhancing marine barite nucleation is also under investigation. Research of barite production under laboratory conditions simulating natural marine environments serves to highlight the role of biofilms in Ba accumulation and reveal the mechanism by which bacteria and other potential EPS-producing organisms provide for barite precipitation in the water column (Martínez-Ruiz et al., 2018, 2019, 2020). These authors have also described how Ba phosphate acts as a precursor for a later Ba-enriched phase that becomes more crystalline, evolving to barite crystals. In general, the role of EPS in concentrating diverse elements has been broadly demonstrated, for instance for bioremediation due to the capacity to remove toxic metals (Gupta and Diwan, 2016). Some authors underline the utility of certain bacteria in retaining lantanides, lead, silver and uranium by sorption (González-Muñoz et al., 1997; Merroun et al., 1998, 2001, 2003) and, more recently, it has been shown that sponge-associated bacteria can mineralize arsenic and barium on intracellular vesicles (Keren et al., 2017). Specifically, the EPS of *Marinobacter* sp., one of the species studied in this thesis, have proven to be sorbent of metals like lead and copper (Bhaskar et al., 2006). Large EPS are essential in the formation of biofilms (Shutterland, 2001; Mancuso Nichols et al., 2005) that in turn allow solutions to be enriched with cations, increasing local supersaturation and promoting precipitation. The results of this thesis further support the hypothesis that during high productivity periods, the organic matter decay and EPS production in microenvironments lead to Ba accumulation and barite precipitation. Regarding the surrounding environment some authors found that biofilms —whose main attaching structures are EPS— enrich the solution with cations, which affects the balance between nucleation and growth rates by increasing local supersaturation and promoting the formation of Sr-rich barite nuclei in a microenvironment. In fact, it has been shown that in even in a Ba undersaturated extreme environment, like Lake Superior in North America, it is possible to find authigenic barite (Horner et al., 2017), which highlights the necessity of a constrained framework for barite precipitation and the importance of the microenvironment. Biogenic processes mediating barite precipitation are further supported by Ba isotope fractionation (Horner et al., 2015; Cao et al., 2016; Bates et al., 2017; Bridgestock et al., 2018); a better assessment of Ba isotopes fractionation could provide interesting information about productivity and water mass mixing, meaning there is still an exciting and broad field to investigate regarding to the Ba cycle.

Chapter VIII. General conclusions

To further understand mechanisms involved in barite precipitation in the Mediterranean, this thesis has focused on three main aspects of the Ba biogeochemical cycle: i) its distribution in sediments including both spatial variability and fluctuations over time; ii) the dissolved Ba distribution at present to assess Ba sources and cycling, and iii) mechanisms of barite precipitation leading to the subsequent Ba enrichment in marine sediments.

In a relatively restricted basin as the Mediterranean Sea, with an exceptional Ba record preserved mirroring productivity fluctuations, the study of the spatial and temporal Ba distribution is crucial to further understand such fluctuations. Ba content in the Mediterranean sapropels has evidenced that these organic-rich sediments deposited during enhanced productivity periods associated with insolation maxima. Increasing precipitation and high freshwater input at this time also led to changes in buoyancy and to stratification and oxygen depletion in bottom waters, which in turn contributed to sapropel preservation. The physiography of the Mediterranean basin has also been an important factor controlling the spatial Ba distribution, which is demonstrated by the distribution of Pliocene to Holocene sapropels. Over time, productivity oscillations and barite precipitation have also controlled dissolved Ba. At present, the dissolved Ba concentration in the Mediterranean increases from the inflowing Atlantic surface water (avg. 8.3 µg/l) to the intermediate (avg. 9.1 µg/l) and deep (avg. 10.3 µg/l) waters. These differences between the water bodies are a consequence of strong convection during winter and an intense Ba cycling releasing Ba in deep waters. Although it has been recently proposed that the internal Ba cycle is more important than previously thought, the high Ba concentrations with depth can be attributed to the residence time of the water masses, thus the Ba content is increasing after several cycles. Nevertheless, the relatively low residence time of the Mediterranean water masses compared to other oceans suggests that other Ba sources may contribute to the Ba unbalance in the Mediterranean. The Ba budget scheme proposed for the western Mediterranean by diverse authors has revealed that Saharan dust and river inputs are not enough to explain the Ba distribution, and have highlighted other potential Ba sources, such as submarine groundwater discharge or

Ba diffusion from Messinian evaporites. The obtained data in this thesis also support the importance of these sources, though more information on the Ba input from rivers is required since it can be usually underestimated. The role of the anthropogenic Ba release to the Mediterranean also requires further investigation to constrain this source.

Regarding barite formation, despite oceans are generally undersaturated in Ba, marine barite precipitates in the water column. Although it is well known that barite precipitation takes place within Ba-rich microenvironments during organic matter decomposition, the precise mechanisms involved in the precipitation are not fully understood, therefore this thesis has contributed to further understand such precipitation mechanisms, in particular, the potential role played by bacteria. Barite production assays carried out with bacteria from natural samples have demonstrated the capability of bacteria from Mediterranean seawater to precipitate barium sulphate under experimental conditions. These results further support the hypothesis that bacteria and associated EPS within decaying organic matter promote Ba accumulation in microenvironments where barite precipitation occurs.

Within this context the main conclusions of this study are:

1. The analysis of the Ba distribution across the Mediterranean basins has revealed that in addition to primary productivity, other factors are also affecting the Ba content in marine sediments. In particular, water depth is an important one, as demonstrated by higher Ba accumulation rates in deeper sites. Moreover, high sedimentation rates also have a dilution effect in Ba accumulation, which should be taken into account for paleoproductivity estimates. Spatial variability in Ba accumulation is also affected by physiography and seawater circulation and in turn by redox conditions.

2. Regarding Ba accumulation fluctuations over time, TOC and Ba contents reflect productivity versus preservation. Both were apparently of equal importance during the Pliocene and Early Pleistocene, when restricted water ventilation enhanced sapropel preservation, while Late Pleistocene and Holocene sapropels suggest productivity was the main factor triggering organic matter accumulation.

3. The study of barite separated from sapropel layers evidence good preservation of barite despite reducing conditions may have been important during certain periods, which suggests pore waters have been saturated in sulphate, which is likely in the Mediterranean due to the Messinian deposits.

4. The analysis of Ba distribution in the water column shows typical vertical profiles of dissolved Ba, which reflects the biological role in barite precipitation in the upper layers, its partial dissolution in the intermediate layers, and the exporting of dissolved Ba to the deep water.

5. While in surface water the distribution of dissolved Ba is controlled by primary productivity, in the deep water masses hydrodynamics seems to enhance dissolved Ba content by accumulation after several cycles. Despite the low residence time of the Mediterranean Sea water masses compared with other oceans, the relatively high residence time of deep waters promotes, along with barite partial dissolution at depth, a Ba increase in the basin.

6. The high resolution dataset of dissolved Ba in the water column provided here show a very good correlation with other data previously published by other authors who also predicted a Ba fluxes imbalance in the Mediterranean Sea. External Ba sources have been largely underestimated, such as the submarine groundwater discharge, brines from Messinian deposits, or some kind of diffusion from sediments, which should be further investigated.

7. Regarding mechanism involved in barite precipitation, the results demonstrate that diverse bacterial strains, isolated from natural Mediterranean Sea surface water samples, are capable to promote barite production under laboratory conditions. Such production occurs in culture media of composition similar to seawater, which is nutrient-enriched. Because these strains belong to very different phylogenetic groups and they are present in diverse marine habitats, it is suggested that the participation of bacteria in barite precipitation in the seawater column it is not linked to specific strains, but rather it is a widespread process. This demonstrates that bacteria play an important role in barite production at present, which supports they have played also a significant role in the past.

8. As the strains more effective in the production of barite are those showing higher content of EPS, it can be inferred that the presence of bacterial EPS (which is common in their natural habitats) is of crucial importance in bacterial barite production. Its role would be to fix Ba ions to phosphate groups present in the EPS, reaching a Ba concentration higher than seawater. Therefore, it would make it possible to reach concentrations high enough to allow barite precipitation.

9. As demonstrated in initial experimental work using soil bacteria, the obtained experimental results also support that barite formation initiates with a barium phosphate phase, a common step in bacterial biomineralization, that evolves to barite through intermediate Ba phosphate and Ba sulphate phases.

10. Since the bacterial population's development is higher as higher is the quantity of available nutrients, and the metabolic pathways that bacteria can use are diverse, it can be deduced that in higher productivity periods - with higher nutrient input – the bacterial production of marine barite is consequently higher than in times of lower productivity.

In summary, the results of this thesis further support the connection between high bacterial activity and marine barite formation, and it can be concluded that the bacterial populations had a major impact in barite precipitation in the oceans and this would be a relevant process to take into account when using Ba proxies for paleoceanographic reconstructions.

Chapter IX. References

Acinas, S.G., Antón, J., Rodríguez-Valera, F., 1999. Diversity of free-living and attached bacteria in offshore Western Mediterranean waters as depicted by analysis of genes encoding 16S rDNA. Appl. Environ. Microbiol. 65, 514–522. http://dx.doi.org/10.1128/AEM.65.2.514-522.1999.

Adloff, F., Somot, S., Sevault, F., Jordà, G., Aznar, R., Déqué, M., Herrmann, M., Marcos, M., Dubois, D., Padorno, E., Alvarez-Fanjul, E., Gomis, D., 2015. Mediterranean Sea response to climate change in an ensemble of twenty first century scenarios. Clim. Dyn. 45**,** 2775–2802. https://doi.org/10.1007/s00382-015-2507-3.

Aloisi, G., Wallmann, K., Bollwerk, S.M., Derkachev, A., Bohrmann, G., Suess, E., 2004. The effect of dissolved barium on biogeochemical processes at cold seeps. Geochim. Cosmochim. Acta 68 (8), 1735-1748. https://doi.org/10.1016/j.gca.2003.10.010.

Altschul, S.F., Madden, T.L., Schäfer, A.A., Zhang, J., Zhang, Z., Miller,W., Lipman, D.J., 1997. Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. Nucleic Acids Res. 25, 3389–3402. http://dx.doi.org/10.1093/nar/25.17.3389.

Álvarez-Salgado, X.A., Gago, J., Míguez, B.M., Pérez, F.F., 2001. Net ecosystem production of dissolved organic carbon in a coastal upwelling system: the Ría de Vigo, Iberian margin of the North Atlantic. Limn Oceanogr 46. http://dx.doi.org/10.4319/lo.2001.46.1.0135.

Amante, C., Eakins, B.W., 2009. ETOPO1 1 arc-minute global reliefmodel: procedures, data sources and analysis. NOAA Technical Memorandum NESDIS NGDC-24. National Geophysical Data Center, NOAA http://dx.doi.org/10.7289/V5C8276M (Oct/2011).

Amies, J.D., Rohling, E.J., Grant, K.M., Rodríguez-Sanz, L., Marino, G., 2019. Quantification of African Monsoon Runoff During Last Interglacial Sapropel S5. Paleoceanogr. Paleoclimat. 34, 1487–1516. https://doi.org/10.1029/2019PA003652.

Anderson, T.F., 1951. Techniques for the preservation of three-dimensional structures in preparing specimens for electron microscopy. Trans. New York Acad. Sci. 13, 130–134.

Angelucci, M.G., Pinardi, N., Castellari, S., 1998. Air-sea fluxes from operational analyses fields: Intercomparison between ECMWF and NCEP analyses over the Mediterranean area. Phys. Chem. of the Earth 23 (5–6), 569-574. https://doi.org/10.1016/S0079-1946(98)00071-8.

Arnaboldi, M., Meyers, P.A., 2007. Trace element indicators of increased primary production and decreased water-column ventilation during deposition of latest Pliocene sapropels at five locations across the Mediterranean Sea. Palaeogeog. Palaeoclimat. Palaeoecol. 249 (3–4), 425-443. https://doi.org/10.1016/j.palaeo.2007.02.016.

Arnosti, C., 2011. Microbial extracellular enzymes and the marine carbon cycle. Ann. Rev. Mar. Sci. 3, 401-25.

https://doi.org/10.1146/annurev-marine-120709-142731.

Arraes-Mescoff, R., Roy-Barman, M., Coppola, L., Souhaut, M., Tachikawa, K., Jeandel, C., Sempéré, R., Yoro, C., 2001. The behavior of Al, Mn, Ba, Sr, REE and Th isotopes during in vitro degradation of large marine particles. Mar. Chemi. 73 (1), 1-19. https://doi.org/10.1016/S0304-4203(00)00065-7.

Astraldi, M., Gasparini, G.P., Vetrano, A., Vignudelli, S., 2002. Hydrographic characteristics and interannual variability of water masses in the central Mediterranean: a sensitivity test for long-term changes in the Mediterranean Sea. Deep Sea Res. Part I: Oceanogr. Res. Papers 49 (4), 661-680. https://doi.org/10.1016/S0967-0637(01)00059-0.

Aquilina, L., Dia, A.N., Boulègue, J., Bourgois, J., Fouillac, A.M., 1997. Massive barite deposits in the convergent margin off Peru: Implications for fluid circulation within subduction zones. Geochim. Cosmochim. Acta 61 (6), 1233-1245,. https://doi.org/10.1016/S0016-7037(96)00402-4.

Avila, A., Alarcón, M., Queralt, I., 1998. The chemical composition of dust transported in red rains—its contribution to the biogeochemical cycle of a holm oak forest in Catalonia (Spain). Atmosph. Environ. 32 (2), 179-191. https://doi.org/10.1016/S1352-2310(97)00286-0.

Awasthi, A., Li, J., 2016. Integrated bioleaching of copper metal from waste printed circuit board—a comprehensive review of approaches and challenges. Environ. Sci. and Pollution Res. 23, 21141–21156. http://dx.doi.org/10.1007/s11356-016-7529-9.

Bacon, M.P., Anderson, R.F., 1982. Distribution of Tho-rium Isotopes between dissolved and particulate forms inthe deep sea. J. Geophys. Res.-Oceans. 87, 2045–2056. https://doi.org/10.1029/JC087iC03p02045.

Bahr, A., Kaboth, S., Jiménez-Espejo, F.J., Sierro, F.J., Voelker, A.H.L., Lourens, L., Röhl, U., Reichart, G.J., Escutia, C., Hernández-Molina, F.J., Pross, J., Friedrich, O., 2015. Persistent monsoonal forcing of Mediterranean Outflow Water dynamics during the late Pleistocene. Geology 43 (11), 951–954. https://doi.org/10.1130/G37013.1.

Bale, N.J., Hennekam, R., Hopmans, E.C., Dorhout, D., Reichart, G., Van der Meer, M., Villareal, T.A., Sinninghe Damsté, J.S., Schouten, S., 2019. Biomarker evidence for nitrogen-fixing cyanobacterial blooms in a brackish surface layer in the Nile River plume during sapropel deposition. Geology 47 (11), 1088–1092. doi: https://doi.org/10.1130/G46682.1.

Basterretxea, G., Tovar-Sanchez, A., Beck, A.J., Masqué, P., Bokuniewicz, H.J., Coffey, R., Duarte, C.M., Garcia-Orellana, J., Garcia-Solsona, E., Martinez-Ribes, L., Vaquer-Sunyer, R., 2010. Submarine Groundwater Discharge to the Coastal Environment of a Mediterranean Island (Majorca, Spain): Ecosystem and Biogeochemical Significance. Ecosystems 13, 629–643. https://doi.org/10.1007/s10021-010-9334-5.

Bates, S.L., Hendry, K.R., Pryer, H.V., Kinsley, C.W., Pyle, K.M., Woodward, E.M.S., Horner, T.J., 2017. Barium isotopes reveal role of ocean circulation on barium cycling in the Atlantic. Geochim. Cosmochim. Acta 204, 286–299. https://doi.org/10.1016/j.gca.2017.01.043.

Baumann, P., Baumann, L., 1981. The marine Gram-negative eubacteria; genera *Photobacterium, Beneckea, Alteromonas,* *Pseudomonas* and *Alcaligenes*. In: The Prokaryotes. A Handbook on Habitats, Isolation and Identification of Bacteria. Springer, Berlin, 1302–1330.

Baumann, L., Baumann, P., Mandel, M., Allen, R.D., 1972. Taxonomy of aerobic marine eubacteria. J. Bacteriol. 110, 402–429. http://dx.doi.org/10.1128/JB.110.1.402-429.1972.

Baumann, P., Gauthier, M.J., Baumann, L., 1984. Genus *Alteromonas*. In: Bergey's Manual of Systematic Bacteriology. Williams and Wilkins Co, Baltimore, Maryland, 243–354.

Bazylinski, D.A., Frankel, R.B., Konhauser, K.O., 2007. Modes of Biomineralization of Magnetite by Microbes. Geomicrobiol. J. 24 (6), 465-475. . http://dx.doi.org/10.1080/01490450701572259.

Beck, A., Charette, M., Cochran, J. K., Gonneea, M., Peucker-Ehrenbrink, B., 2013. Dissolved strontium behavior in the subterranean estuary and implications for the Sr mass balance and isotope budget of the global ocean. Geochim. Cosmochim. Acta 117 (33–52). http://dx.doi.org/10.1016/j.gca.2013.03.021.

Beck, A.J., Tsukamoto, Y., Tovar-Sanchez, A., Huerta-Diaz, M., Bokuniewicz, H.J., Sañudo-Wilhelmy, S.A., 2007. Importance of geochemical transformations in determining submarine groundwater discharge-derived trace metal and nutrient fluxes. Applied Geochem. 22 (2), 477-490. https://doi.org/10.1016/j.apgeochem.2006.10.005.

Bejannin, S., Tamborski, J., Van Beek, P., Souhaut, M., Stieglitz, T., Radakovitch, O., Claude, Christelle, Conan, P., Pujo-Pay, M., Crispi, O., et al., 2020. Nutrient fluxes associated wth submarine groundwater discharge from karstic coastal aquifers (CôteBleue, French Mediterranean Coastline). Front. Environ. Sci. 7. http://dx.doi.org/10.3389/fenvs.2019.00205.

Bejannin, S., Van Beek, P., Stieglitz, T., Souhaut, M., Tamborski, J., 2017. Combining airborne thermal infrared images and radium isotopes to study submarine groundwater discharge along the French Mediterranean coastline. J. Hydrology: Regional Studies 13, 72-90. https://doi.org/10.1016/j.ejrh.2017.08.001.

Béranger, K., Testor, P., Crépon, M., 2009. Modelling water mass formation in the gulf of lions (Mediterranean Sea). CIESM Workshop Monographs. 38, 91-100.

Bernat, M., Church, T., Allegre, C.J., 1972. Barium and strontium concentrations in Pacific and Mediterranean sea water profiles by direct isotope dilution mass spectrometry. Earth Planet. Sci. Lett. 16 (1), 75-80. https://doi.org/10.1016/0012-821X(72)90238-5.

Bernstein, R.E., Betzer, P.R., Feely, R.A., Byrne, R.H., Lamb, M.F., Michaels, A.F., 1987. Acantharian fluxes and strontium to chlorinity ratios in the North Pacific Ocean. Science 237, 1490–1494. http://dx.doi.org/10.1126/science.237.4821.1490.

Bernstein, R.E., Byrne, R.H., 2004. Acantharians and marine barite. Mar. Chem. 86, 45–50. http://dx.doi.org/10.1016/j.marchem.2003.12.003.

Bernstein, R.E., Byrne, R.H., Betzer, P.R., Greco, A.M., 1992. Morphologies and transformations of celestite in seawater: The role of acantharians in strontium and barium geochemistry. Geochim. Cosmochim. Acta 56(8), 3273-3279. https://doi.org/10.1016/0016-7037(92)90304-2.

Bernstein, R.E., Byrne, R.H., Schijf, J., 1998. Acantharians: a missing link in the oceanic biogeochemistry of barium. Deep Sea Res. Part I: Oceanogr. Res. Papers 45 (2–3), 491-505. https://doi.org/10.1016/S0967-0637(97)00095-2.

Bertram, M.A., Cowen, J.P., 1997. Morphological and compositional evidence for biotic precipitation of marine barite. J. Mar. Res. 55, 577–593. http://dx.doi.org/10.1357/0022240973224292.

Beşiktepe, S.T., Sur, H.I., Özsoy, E., Abdul, M., Latif, M.A., Oğuz, T., Ünlüata, U., 1994. The circulation and hydrography of the Marmara Sea. Progr. Ocean. 34 (4), 285-334. https://doi.org/10.1016/0079-6611(94)90018-3.

Bethoux, J.P., 1979. Budgets of the Mediterranean Sea - their dependance on the local climate and on the characteristics of the atlantic waters. Oceanol. Acta, 2 (2), 157-163.

Bethoux, J.P., Copin-Montégut, G., 1984. Effects de la dynamique méditerranéenne sur le devenir des apports anthropogéniques. J. Tième (Ed.), Etud. Pollutions, C.I.E.S.M, Lucerne, 165-171.

Bethoux, J.P., Gentili, B., Morin, P., Nicolas, N., Pierre, C., Ruiz-Pino, D., 1999. The Mediterranean Sea: a miniature ocean for climatic and environmental studies and a key for the climatic functioning of the North Atlantic. Progr. Oceanogr. 44 (1–3), 131-146. https://doi.org/10.1016/S0079-6611(99)00023-3.

Bethoux, J.P., Morin, P., Ruiz-Pino, D.P., 2002. Temporal trends in nutrient ratios: chemical evidence of Mediterranean ecosystem changes driven by human activity. Deep Sea Res. Part II: Topical Studies in Oceanogr. 49 (11), 2007-2016. https://doi.org/10.1016/S0967-0645(02)00024-3.

Betts, J.N., Holland, H.D., 1991. The oxygen content of ocean bottom waters, the burial efficiency of organic carbon, and the regulation of atmospheric oxygen. Global Planet. Change 5 (1–2), 5–18. https://doi.org/10.1016/0921-8181(91)90123-E.

Beusen, A.H.W., Slomp, C.P., Bouwman, A.F., 2013. Global land-ocean linkage: direct inputs of nitrogen to coastal waters via submarine groundwater discharge. Environ. Res. Lett. 8, 6.

Bianchi, D., Zavatarelli, M., Pinardi, N., Capozzi, R., Capotondi, L., Corselli, C., Masina, S., 2006. Simulations of ecosystem response during the sapropel S1 deposition event. Palaeogeogr. Palaeoclimatol. Palaeoecol. 235 (1–3), 265-287. https://doi.org/10.1016/j.palaeo.2005.09.032.

Bishop, J.K.B., 1988. The barite-opal-organic carbon association in oceanic particulate matter. Nature 331, 341–343. http://dx.doi.org/10.1038/332341a0.

Boesch, D.F., Rabalais, N.N., 1989. Environmental impact of produced water discharges in coastal Louisiana. Report to the Louisiana Division of the Mid-Continent Oil and Gas Association, Baton Rouge, Louisiana.

Bonnet, S., Guieu, C., 2006. Atmospheric forcing on the annual iron cycle in the western Mediterranean Sea: A 1-year survey. J. Geophys. Res. 111, C09010. http://dx.doi.org/10.1029/2005JC003213.

Bonny, S., Jones, B., 2007. Diatom-mediated barite precipitation in microbial mats calcifying at Stinking Springs, a warm sulphur spring system in Northwestern Utah, USA. Sediment. Geol. 194, 223–244.

http://dx.doi.org/10.1016/j.sedgeo.2006.06.007.

Bonny, S.M., Jones, B., 2008. Experimental precipitation of barite (BaSO4) among streamers of sulfur-oxidizing bacteria. J. Sediment. Res. 78 (5), 357–365. http://dx.doi.org/10.2110/jsr.2008.038.

Bontognali, T.R.R., McKenzie, J.A., Warthmann, R.J., Vasconcelos, C., 2014. Microbially influenced formation of Mg-calcite and Ca-dolomite in the presence of exopolymeric substances produced by sulphate-reducing bacteria. Terra Nova 26 (1), 72–77. http://dx.doi.org/10.1111/ter.12072.

Boothe, P.N., Presley, B.J., 1989. Trends in sediment trace element concentrations around six petroleum drilling platforms in the northwestern Gulf of Mexico. In: Drilling Wastes, Elsevier Applied Science, London, 3-21.

Bopp, L., Resplandy, L., Orr, JC., Doney, S.C., Dunne, J.P., Gehlen, M., Halloran, P., Heinze, C., Ilyina, T., Séférian, R., Tjiputra, J., Vichi, M., 2013. Multiple stressors of ocean ecosystems in the 21st century: projections with CMIP5 models. Biogeosci. 10, 6225–6245. https://doi.org/10.5194/bg-10-6225-2013.

Böttcher, M.E., Rinna, J., Warning, B., Wehausen, R., Howell, M.W., Schnetger, B., Stein, R., Brumsack, H.J., Rullkötter, J., 2003. Geochemistry of sediments from the connection between the western and the eastern Mediterranean Sea (Strait of Sicily, ODP Site 963). Palaeogeogr. Palaeoclimatol. Palaeoecol. 190, 165-194. https://doi.org/10.1016/S0031-0182(02)00604-1.

Boukthir, M., Barnier, B., 2000. Seasonal and inter-annual variations in the surface freshwater flux in the Mediterranean Sea from the ECMWF re-analysis project. J. Mar. Syst. 24 (3–4), 343-354. https://doi.org/10.1016/S0924-7963(99)00094-9.

Bouloubassi, I., Guehenneux, G., Rullkötter, J., 1998. Biological marker significance of organic matter origin in sapropels from the Mediteranean Ridge, site 9691. Proceedings of the Ocean Drilling Program, Scientific Results, Vol. 160. http://dx.doi.org/10.2973/odp.proc.sr.160.003.1998.

Bowman, J.P., 1998. *Pseudoalteromonas prydzensis* sp. nov., a psychrotrophic, halotolerant bacterium from Antarctic sea ice. Int. J. Syst. Bacteriol. 48, 1037–1041. http://dx.doi.org/10.1099/00207713-48-3-1037.

Bowman, J.P., McCammon, S., Brown, M.V., Nichols, D.S., McMeekin, T.A., 1997. Diversity and association of psychrophilic bacteria in Antartic sea ice. Appl. Environ. Microbiol. 63, 3068–3078. http://dx.doi.org/10.1128/AEM.63.8.3068-3078.1997.

Boyd, P.W., Claustre, H., Levy, M., Siegel, D.A., Weber, T., 2019. Multi-Faceted Particle Pumps Drive Carbon Sequestration in the Ocean. Nature 568, 327–35. http://dx.doi.org/10.5194/bg-14-177-2017.

Boyd, P., Jickells, T., Law, C., Blain, S., Boyle, E., Buesseler, K., Coale, K., Cullen, J., De Baar, H. J. W., Follows, M., Harvey, M., Lancelot, C., Levasseur, M., Pollard, R., Rivkin, R., Sarmiento, J., Schoemann, V., Smetacek, V., Takeda, S., Tsuda, A., Turner, S., Watson, A., 2007. A synthesis of mesoscale iron-enrichment experiments 1993-2005: key findings and implications for ocean biogeochemistry. Science 315, 612-617.

Boyer, T.P., Baranova, O.K., Coleman, C., Garcia, H.E., Grodsky, A., Locarnini, R.A., Mishonov, A.V., O'Brien, T.D., Paver, C.R., Reagan, J.R., Seidov, D., Smolyar, I.V., Weathers, K., Zweng, M.M., 2018. World

Ocean Database 2018 (in preparation). MEDAR Group, 2002.

Bowie, A.R., Whitworth, D.J., Achterberg, E.P., Mantoura, R.F.C., Worsfold, P.J., 2002. Biogeochemistry of Fe and other trace elements (Al, Co, Ni) in the upper Atlantic Ocean. Deep Sea Res. Part I: Oceanogr. Res. Papers 49 (4), 605-636. https://doi.org/10.1016/S0967-0637(01)00061-9.

Brettar, I., Christen, R., Höfle, M.G., 2003. *Idiomarina baltica* sp. nov., a marine bacterium with a high optimum growth temperature isolated from surface water of the central Baltic Sea. Int. J. Syst. Evol. Microbiol. 53, 407–413. http://dx.doi.org/10.1099/ijs.0.02399-0.

Bridgestock, L., Hsieh, Y., Porcelli, D., Homoky, W.B., Bryan, A., Henderson, G.M., 2018. Controls on the barium isotope compositions of marine sediments. Earth and Planet. Sci. Let. 481, 101–110. https://doi.org/10.1016/j.epsl.2017.10.019.

Bristow, L.A., Mohr, W., Ahmerkamp, S., Kuypers, M.M.M., 2017. Nutrients that limit growth in the ocean. Current Biol. 27(11), 474-478. https://doi.org/10.1016/j.cub.2017.03.030.

Broecker, W.S., Goddard, J., Sarmiento, J.L., 1976. The distribution of 226Ra in the Atlantic Ocean. Earth Planet. Sci. Lett.32 (2), 220-235. https://doi.org/10.1016/0012-821X(76)90063-7.

Broecker, W., Peng, T., Engh, R., 1980. Modeling the Carbon System. Radiocarbon, 22 (3), 565-598. https://doi.org/10.1017/S0033822200009966.

Bruland, K., Lohan, M., 2003. Controls of Trace Metals in Seawater. Treat. on Geochem. 6. https://doi.org/10.1016/B0-08-043751-6/06105-3.

Bryden, H.L., Kinder, T.H., 1991. Steady two-layer exchange through the Strait of Gibraltar. Deep Sea Res. Part A. Oceanogr. Res. Papers 38 (1), S445-S463. https://doi.org/10.1016/S0198-0149(12)80020-3.

Cacho, I., Grimalt, J.O., Canals, M., 2002. Response of the Western Mediterranean Sea to rapid climatic variability during the last 50,000 years: a molecular biomarker approach. J. Mar. Syst. 33–34, 253–272. https://doi.org/10.1016/S0924-7963(02)00061-1.

Calleja, M.L., Al-Otaibi, N., Morán, X.A.G., 2019. Dissolved organic carbon contribution to oxygen respiration in the central Red Sea. Sci. Rep. 9, 469. https://doi.org/10.1038/s41598-019-40753-w.

Calvert, S.E., 1983. Geochemistry of Pleistocene sapropels and associated sediments from the Eastern Mediterranean. Oceanol. Acta 6, 255–267.

Calvert, S.E., Pedersen, T.F., 1992. Organic carbon accumulation and preservation in marine sediments: How important is anoxia? In: Productivity, Accumulation and Preservation of Organic Matter in Recent and Ancient Sediments, Columbia Univ. Press, New York, 231–263.

Candelario, R., Monreal, M.A., Nicolaevich, S., 2008. Efectos de los vórtices en sistemas acuáticos y su relación con la química, biología y geología. Interciencia 33, 741–746.

Cane, T., Rohling, E.J., Kemp, A.E.S., Cooke, S., Pearce, R.B., 2002. High-resolution stratigraphic framework for Mediterranean sapropel S5: defining temporal relationships between records of Eemian climate variability. Palaeogeogr. Palaeoclimatol. Palaeoecol. 183 (1–2), 87–101.

https://doi.org/10.1016/S0031-0182(01)00461-8.

Canfield, D.E., 1989. Sulfate reduction and oxic respiration in marine sediments: implications for organic carbon preservation in euxinic environments. Deep Sea Res. Part A. Oceanogr. Res. Papers 36 (1), 121-138. https://doi.org/10.1016/0198-0149(89)90022-8.

Cao, Z., Siebert, C., Hathorne, E.C., Dai, M., Frank, M., 2016. Constraining the oceanic barium cycle with stable barium isotopes. Earth and Planet. Sci. Let. 434, 1–9. https://doi.org/10.1016/j.epsl.2015.11.017.

Capotondi, L., Girone, A., Lirer, F., Bergami, C., Verducci, M., Vallefuoco, M., Afferri, A., Ferraro, L., Pelosi, N., De Lange, G.J., 2016. Central Mediterranean Mid-Pleistocene paleoclimatic variability and its association with global climate. Palaeogeogr. Palaeoclimatol. Palaeoecol. 442, 72–-83. https://doi.org/10.1016/j.palaeo.2015.11.009.

Capozzi, R., Negri, A., 2009. Role of sea-level forced sedimentary processes on the distribution of organic carbon-rich marine sediments: A review of the Late Quaternary sapropels in the Mediterranean Sea. Palaeogeogr. Palaeoclimatol. Palaeoecol. 273 (3–4), 249–257. https://doi.org/10.1016/j.palaeo.2008.05.009.

Carpenter, A., Kostianoy, A., 2018. Oil Pollution in the Mediterranean Sea: Part I The International Context: The International Context. https://doi.org/10.1007/978-3-030-12236-2.

Carter, S.C.; Paytan, A.; Griffith, E.M., 2020. Toward an Improved Understanding of the Marine Barium Cycle and the Application of Marine Barite as a Paleoproductivity Proxy. Minerals 10, 421. https://doi.org/10.3390/min10050421.

Carvalho, M.C., Schulz, K.G., Eyre, B.D., 2017. Respiration of new and old carbon in the surface ocean: Implications for estimates of global oceanic gross primary productivity. Global Biogeochem. Cycles 31, 975– 984. https://doi.org/10.1002/2016GB005583.

Casford, J.S.L., Rohling, E.J., Abu-Zied, R.H., Fontanier, C., Jorissen, F.J., Leng, M.J., Schmiedl, G., Thomson, J., 2003. A dynamic concept for eastern Mediterranean circulation and oxygenation during sapropel formation. Palaeogeogr. Palaeoclimatol. Palaeoecol. 190, 103-119. https://doi.org/10.1016/S0031-0182(02)00601-6.

Castellari, S., Pinardi, N., Leaman, K., 1998. A model study of air–sea interactions in the Mediterranean Sea. J. Mar. Syst. 18 (1–3), 89-114. https://doi.org/10.1016/S0924-7963(98)90007-0.

Castradori, D., 1993. Calcareous nannofossils and the origin of eastern Mediterranean sapropels. Paleoceanogr. 8 (4), 459– 471. https://doi.org/10.1029/93PA00756.

Cavan, E., Henson, S., Belcher, A., Sanders, R., 2017. Role of zooplankton in determining the efficiency of the biological carbon pump. Biogeosci. 14, 177–186. http://dx.doi.org/10.5194/bg-14-177-2017.

Chan, K.Y., Baumann, L., Garza,M.M., Baumann, P., 1978. Two new species of *Alteromonas*: *Alteromonas espeijiana* and *Alteromonas undina*. Int. J. Syst. Bacteriol. 28, 217–222. http://dx.doi.org/10.1099/00207713-28-2-217.

Chan, L.H., Edmond, J.M., Stallard, R.F., Broecker, W.S., Chung, Y.C., Weiss, R.F., Ku, T.L., 1976. Radium and barium at GEOSECS stations in the Atlantic and Pacific. Earth Planet. Sci. Lett. 32, 258-267.

Charette, M.A., Morris, P.J., Henderson, P.B., Moore, W.S., 2015. Radium isotope distributions during the US GEOTRACES

North Atlantic cruises. Mar. Chem. 177 (1), 184-195. https://doi.org/10.1016/j.marchem.2015.01.001.

Charette, M.A., Sholkovitz, E.R., 2002. Oxidative precipitation of groundwater-derived ferrous iron in the subterranean estuary of a coastal bay, Geophys. Res. Lett. 29 (10). https://doi.org/10.1029/2001GL014512.

Chen, X., Cukrov, N., Santos, I.R., Rodellas, V., Cukrov, N., Du, J., 2020. Karstic submarine groundwater discharge into the Mediterranean: Radon-based nutrient fluxes in an anchialine cave and a basin-wide upscaling. Geochim. Cosmochim. Acta 268, 467-484. https://doi.org/10.1016/j.gca.2019.08.019.

Chen, S., Yang, Y., Liu, C., Dong, F., Liu, B., 2015. Column bioleaching copper and its kinetics of waste printed circuit boards (WPCBs) by *Acidithiobacillus ferrooxidans.* Chemosph. 141,162–168. https://doi.org/10.1016/j.chemosphere.2015.06.082.

Chester, R., Nimmo, M., Corcoran, P.A., 1997. Rain water-aerosol trace metal relationships at cap ferrat: A coastal site in the Western Mediterranean. Mar.e Chem. 58 (3–4), 293-312. https://doi.org/10.1016/S0304-4203(97)00056-X.

Chisholm, S.W., 2000. Stirring Times in the Southern Ocean. Nature 407, 685–86. https://doi.org/10.1038/35037696.

Cho, B., Azam, F., 1988. Major role of bacteria in biogeochemical fluxes in the ocean's interior. Nature 332, 441–443. https://doi.org/10.1038/332441a0.

Cho, H.M., Kim, G., Kwon, E., Moosdorf, N., Garcia-Orellana, J., Santos, I.R., 2018. Radium tracing nutrient inputs through submarine groundwater discharge in the global ocean. Sci. Rep. 8, 2439. https://doi.org/10.1038/s41598-018-20806-2.

Choi, D.H., Cho, B.C., 2005. *Idiomarina seosinensis* sp nov., isolated from hypersaline water of a solar saltern in Korea. Int. J. Syst. Evol. Microbiol. 55, 379–383. http://dx.doi.org/10.1099/ijs.0.63365-0.

Chou, L., Wollast, R., 1997. Biogeochemical behavior and mass balance of dissolved aluminum in the western Mediterranean Sea. Deep Sea Res. Part II: Topical Studies in Oceanogr. 44 (3–4), 741-768. https://doi.org/10.1016/S0967-0645(96)00092-6.

Chow, T.J., Goldberg, E.D., 1960. On the marine geochemistry of barium. Geochim. Cosmochim. Acta 20, 192–198. http://dx.doi.org/10.1016/0016-7037(60)90073-9.

Chung, A.P., Sousa, T., Pereira, A., Morais, P.V., 2014. Microorganisms – Tools for Bioremediation of Uranium Contaminated Environments. Procedia Earth and Planet. Sci. 8, 53-58. https://doi.org/10.1016/j.proeps.2014.05.012.

Chun, J., Lee, J.H., Jung, Y., Kim, M., Kim, S., Kwon Kim, B., Lim, Y.W., 2007. EzTaxon: a web-based tool for the identification of prokaryotes based on 16S ribosomal RNA gene sequences. Int. J. Syst. Evol. Microbiol. 57, 2259–2261. http://dx.doi.org/10.1099/ijs.0.64915-0.

Church, T.M., Wolgemuth, K., 1972. Marine barite saturation. Earth and Planet. Sci. Let.15 (1), 35–44. https://doi.org/10.1016/0012-821X(72)90026-X.

Cita, M.B., Vergnaud-Grazzini, C., Robert, C., Chamley, H., Ciaranfi, N., d'Onofrio, S., 1977. Paleoclimatic record of a long deep sea core from the eastern Mediterranean. Quat. Res.

8 (2), 205-235. https://doi.org/10.1016/0033-5894(77)90046-1.

Clarke, R.A., Coote, A.R., 1988. The formation of Labrador Sea water. Part III: The evolution of oxygen and nutrient concentration. J. Phys. Oceanogr. 18 , 469–480. https://doi.org/10.1175/1520-0485(1988)018<0469:TFOLSW>2.0.CO;2.

Cociasu, A., Dorogan, L., Humborg, C., Popa, L., 1996. Long-term ecological changes in Romanian Coastal Waters ofthe Black Sea. Mar. Poll. Bull. 32(l), 32-38. https://doi.org/10.1016/0025-326X(95)00106-W.

Cociasu, A., Popa, L., 2002. Long-term evolution of the nutrient concentration on the north-western shelf of the Black Sea. Estuarine, Coastal and Shelf Science.

Codispoti, L.A., Friederich, G.E., Murray, J.W., Sakamoto, C.M., 1991. Chemical variability in the Black Sea: implications of continuous vertical profiles that penetrated the oxic/anoxic interface. Deep Sea Res. Part A. Oceanogr. Res. Papers 38 (2), S691-S710. https://doi.org/10.1016/S0198-0149(10)80004-4.

Coleman, M.L., Curtis, D.C., Irwin, H., 1979. Burial rate a key to source and reservoir potential. World Oil 188, 83-92.

Collier, R., Edmond, J., 1984. The trace element geochemistry of marine biogenic particulate matter. Prog. Ocean. 13, 113-199. 10.1016/0079-6611(84)90008-9

Collins, J.R., Edwards, B.R., Thamatrakoln, K., Ossolinski, J.E., DiTullio, G.R., Bidle, K.D., Doney, S. C., Van Mooy, B.A.S., 2015. The multiple fates of sinking particles in the North Atlantic Ocean. Global Biogeoch. Cycles 29, 1471– 1494. https://doi.org/10.1002/2014GB005037.

Comas, M.C., Zahn, R., Klaus, A., ODP Leg 61 Scientific Party, 1996. Proc. ODP. Init. Repts. 161, College Station, TX (Ocean Drilling Program). https://doi.org/10.2973/odp.proc.ir.161.1996.

Conte, M., Giuffrida, A., Tedesco, S., 1989. The Mediterranean Oscil-lation: Impact on Precipitation and Hydrology in Italy. Conferenceon Climate and Water. Publicat. Acad. of Finland: Helsinki.

Coppola, L., Legendre, L., Lefevre, D., Prieur, L., Taillandier, V., Riquier, E.D., 2018. Seasonal and inter-annual variations of dissolved oxygen in the northwestern Mediterranean Sea (DYFAMED site). Progr. Oceanogr 162, 187-201, https://doi.org/10.1016/j.pocean.2018.03.001.

Corselli, C., Principato, M. S., Maffioli, P., and Crudeli, D., 2002. Changes in planktonic assemblages during sapropel S5 deposition: Evidence from Urania Basin area, eastern Mediterranean. Paleoceanogr. 17(3). http://dx.doi.org/10.1029/2000PA000536.

Cortina, A., Filippelli, G., Ochoa, D., Sierro, F.J., Flores, J.A., Grimalt, J.O., 2018. Climate-driven changes in sedimentation rate influence phosphorus burial along continental margins of the northwestern Mediterranean. Palaeogeogr. Palaeoclimatol. Palaeoecol. 500, 106-116. https://doi.org/10.1016/j.palaeo.2018.03.010.

Costa, K.M., Jacobel, A.W., McManus, J.F., Anderson, R.F., Winckler, G., Thiagarajan, N., 2017. Productivity patterns in the equatorial Pacific over the last 30,000 years. Global Biogeochem. 31, 850–865. http://dx.doi.org/10.1002/2016GB005579.

Costerton, J.W., Cheng, K.J., Geesey, G.G., Ladd, T.I.N.J.C., Dasgupta, M., Marrie, T.J., 1987. Bacterial biofilms in nature and disease. Annu. Rev. Microbiol. 41, 435–464. http://dx.doi.org/10.1146/annurev.mi.41.100187.002251.

Cramp, A. O'sullivan, G., 1999. Neogene sapropels in the Mediterranean: a review. Mar. Geol. 153, 11–28. http://dx.doi.org/10.1016/S0025-3227(98)00092-9.

Criado-Aldeanueva, F., Soto-Navarro, J , 2020. Climatic Indices over the Mediterranean Sea: A Review. Applied Sci., 10(17), 5790. https://doi.org/10.3390/app10175790.

Crispi, G., Mosetti, R., Solidoro, C., Crise, A., 2001. Nutrients cycling in Mediterranean basins: the role of the biological pump in the trophic regime. Ecol. Model. 138 (1–3), 101-114. https://doi.org/10.1016/S0304-3800(00)00396-3.

Cutter, G., Casciotti, K., Croot, P.r, Geibert, W., Heimbürger, L., Lohan, M., Planquette, H., Van de Flierdt, T., 2017. Sampling and Sample-handling Protocols for GEOTRACES Cruises. Version 3, August 2017. Toulouse, France, GEOTRACES International Project Office, 139 pp. and appendices. http://dx.doi.org/10.25607/OBP-2.

Dando, P.R., Stüben, D., Varnavas, S.P., 1999. Hydrothermalism in the mediterranean sea. Prog. Oceanogr. 44, 333–367. http://dx.doi.org/10.1016/S0079-6611(99)00032-4

Das, A., Krishnaswami, S., 2006. Barium in Deccan Basalt Rivers: Its Abundance, Relative Mobility and Flux. Aquat. Geochem. 12, 221–238. https://doi.org/10.1007/s10498-005-5856-4.

De Baar, H.J.W., Boyd, P.W., Coale, K.H., Landry M.R., Tsuda, A., Assmy, P., Bakker, D.C.E., Bozec, Y., Barber, R.T., Brzezinski, M.A., Buesseler, K.O., Boyé, M., Croot, P.L., Gervais, F., Gorbunov, M.Y., Harrison, P.J., Hiscock, W.T., Laan, P., Lancelot, C., Law, C.S., Levasseur, M., Marchetti, A., Millero, F.J., Nishioka, J., Nojiri, Y., Van Oijen, T., Riebesell, U., Rijkenberg, M.J.A., Saito, H., Takeda, H., Takeda, S., Timmermans, K.R., Veldhuis, M.J.W., Waite, A.M., Wong, C.S., 2005. Synthesis of iron fertilization experiments: From the Iron Age in the Age of Enlightenment. J. Geophys. Res. 110, C09S16. https://doi.org/10.1029/2004JC002601.

De Bruijn, F.J., 1992. Use of repetitive sequences and the polymerase chain reaction to fingerprint the genomes of Rhizobium meliloti isolates and other soil bacteria. Appl. Environ. Microbiol. 58, 2180–2187. https://doi.org/10.1016/S0723-2020(11)80018-4.

Dehairs, F., Baeyens, W., Goeyens, L., 1992. Accumulation of suspended barite at mesopelagic depths and export production in the Southern Ocean. Science 258 (5086), 1332–1335. http://dx.doi.org/10.1126/science.258.5086.1332.

Dehairs, F., Chesselet, R., Jedwab, J., 1980. Discrete suspended particles of barite and the barium cycle in the open ocean. Earth Planet. Sci. Lett. 49, 529–550. http://dx.doi.org/10.1016/0012-821X(80)90094-1.

Dehairs, F., Fagel, N., Antia, A.N., Peinert, R., Elskens, M., Goeyens, L., 2000. Export production in the Bay of Biscay as estimated from barium-barite in settlingmaterial: a comparison with new production. Deep-Sea Res. I 47, 583–601. http://dx.doi.org/10.1016/S0967-0637(99)00072-2.

Dehairs, F., Jacquet, S., Savoye, N., Van Mooy, B.A.S., Buesseler, K.O., Bishop, J.K.B., Lamborg, C.H., Elskens, M., Baeyens,W., Boyde, P.W., Casciotti, K.L.,Monnin, C., 2008. Barium in twilight zone suspended matter as a potential proxy for particulate organic carbon remineralization: results for the North Pacific. Deep-Sea Res. II 55 (14–15), 1673–1683.

http://dx.doi.org/10.1016/j.dsr2.2008.04.020.4.

Dehairs, F., Lambert, C.E., Chesselet, R., Risler, N., 1987. The biological production of marine suspended barite and the barium cycle in the Western Mediterranean Sea. Biogeochem. 4, 119–139.

Dehairs, F., Shopova, D., Ober, S., Goyens, L., 1997. Particulate barium stocks and oxygen consumption in the Southern Ocean mesopelagic water column during spring and early summer: relationship with export production. Deep-Sea Res. II 44 (1–2), 497–516. http://dx.doi.org/10.1016/S0967-0645(96)00072-0.

Dehairs, F., Stroobants, N., Goeyens, L., 1991. Suspended barite as a tracer of biological activity in the Southern Ocean. Mar. Chem. 35, 399–410. http://dx.doi.org/10.1016/S0304-4203(09)90032-9.

Dekov, V.M., Savelli, C., 2004. Hydrothermal activity in the SE Tyrrhenian Sea: an overview of 30 years of research. Mar. Geol. 204 (1–2), 161-185. https://doi.org/10.1016/S0025-3227(03)00355-4.

De Lange, G.J., Catalano, G., Klinkhammer, G.P., Luther, G.W., 1990. The interface between oxic seawater and the anoxic Bannock brine; its sharpness and the consequences for the redox-related cycling of Mn and Ba. Mar. Chem. 31 (1–3), 205–217. https://doi.org/10.1016/0304-4203(90)90039-F.

De Lange, G.J., Middelburg, J.J., Van der Weijden, C.H., Catalano, G., Luther, G.W., Hydes, D.J., Woittiez, J.R.W., Klinkhammer, G.P., 1990. Composition of anoxic hypersaline brines in the Tyro and Bannock Basins, eastern Mediterranean. Mar. Chem. 31 (1–3), 63-88. https://doi.org/10.1016/0304-4203(90)90031-7.

De Lange, G., Thomson, J., Reitz, A. Slomp, C.P., Principato, M.S., Erba, E., Corselli, C., 2008. Synchronous basin-wide formation and redox-controlled preservation of a Mediterranean sapropel. Nat. Geosci. 1, 606–610. https://doi.org/10.1038/ngeo283.

De Noblet, N., Bracannot, P., Joussaume, S., Masson, V., 1996. Sensitivity of simulated Asian and African summer monsoons to orbitally induced variations in insolation 126, 115 and 6 kBP. Clim. Dyn. 12, 589–603. https://doi.org/10.1007/BF00216268.

Desboeufs, K.V., Losno, R., Vimeux, F., Cholbi, S., 1999. The pH-dependent dissolution of wind-transported Saharan dust. J. Geophys. Res. 104 (D17), 21287– 21299. https://doi.org/10.1029/1999JD900236.

Dickens, G.R., Fewless, T., Thomas, E., Bralower, T.J., 2003. Excess barite accumulation during the Paleocene-Eocene Thermal Maximum: Massive input of dissolved barium from seafloor gas hydrate reservoirs. In Causes and Consequences of Globally Warm Climates in the Early Paleogene; Geological Society of America: Boulder, CO,USA, 369, 11–23.

Dickson, A.G., 1994. Determination of dissolved oxygen in sea water by Winkler titration. WOCE Hydrographic Program, Woods Hole, MA, 11.

Diester-Haass, L., Billups, K., Lear, C., 2018. Productivity changes across the mid-Pleistocene climate transition. Earth-Sci. Rev. 179, 372–391. https://doi.org/10.1016/j.earscirev.2018.02.016.

Diester-Haass, L., Faul, K., 2019. Paleoproductivity Reconstructions for the Paleogene Southern Ocean: A Direct Comparison of Geochemical and Micropaleontological Proxies. Paleoceanogr. Paleoclimat. 34, 79–97. https://doi.org/10.1029/2018PA003384.

Dobretsov, S., Dahms, H.-W., Qian, P.Y., 2006. Inhibition of biofouling by marine microorganisms and their metabolites. Biofouling 22, 43–54. http://dx.doi.org/10.1080/08927010500504784.

Dobretsov, S., Xiong, H.R., Xu, Y., Levin, L.A., Qian, P.Y., 2007. Novel antifoulants: inhibition of larval attachment by proteases. Mar. Biotechnol. 9, 388–397. http://dx.doi.org/10.1007/s10126-007-7091-z.

Dolenec, T., Faganeli, J., Pirc, S., 1998. Major, minor and trace elements in surficial sediments from the open Adriatic Sea: a regional geochemical study. Geol. Croat. 51, 59-73.

Donachie, S.P., Hou, S., Gregory, T.S., Malahoff, A., Alam, M., 2003. *Idiomarina loihiensis* sp. nov., a halophilic γ-Proteobacterium from the Lō'ihi submarine volcano, Hawai'i. Int. J. Syst. Evol. Microbiol. 53, 1873–1879. http://dx.doi.org/10.1099/ijs.0.02701-0.

Driussi, O., Maillard, A., Ochoa, D., Lofi, J., Chanier, F., Gaullier, V., Briais, A., Sage, F., Sierro, F., Garcia, M., 2015. Messinian Salinity Crisis deposits widespread over the Balearic Promontory: Insights from new high-resolution seismic data. Mar. and Petrol. Geol. 66 (1), 41-54. https://doi.org/10.1016/j.marpetgeo.2014.09.008.

Duce, R.A., Tindale, N.W., 1991. Atmospheric transport of iron and its deposition in the ocean. Limnol. Oceanogr. 36, http://dx.doi.org/10.4319/lo.1991.36.8.1715.

Dulac, F., Moulin, C., Lambert, C.E., Guillard, F., Poitou, J., Guelle, W., Quetel, C.R., Schneider, X., Ezat, U., 1996. Quantitative Remote Sensing of African Dust Transport to the Mediterranean. In: The Impact of Desert Dust Across the Mediterranean. Environmental Science and Technology Library 11. Springer, Dordrecht. https://doi.org/10.1007/978-94-017-3354-0_3.

Durrieu de Madron, the MerMex Group, 2011. Marine ecosystems' responses to climatic and anthropogenic forcings in the Mediterranean. Progr. Oceanogr. 91 (2), 97-166. https://doi.org/10.1016/j.pocean.2011.02.003.

Dymond, J., Collier, R., 1996. Particulate barium fluxes and their relationships to biological productivity. Deep Sea Res. Part II: Topical Studies in Oceanogr. 43(4–6), 1283–1308. https://doi.org/10.1016/0967-0645(96)00011-2.

Dymond, J., Suess, E., Lyle, M., 1992. Barium in deep-sea sediments: a geochemical proxy for paleoproductivity. Paleocean. 7, 163–181. http://dx.doi.org/10.1029/92PA00181.

Eagle, M., Paytan, A., Arrigo, K.R., Van Dijken, G., Murray, R.W., 2003. A comparison between excess barium and barite as indicators of carbon export: Paleoceanogr. 18, 21.1–21.13. https://doi.org/10.1029/2002PA000793.

Edmond, J.M., Measures, C., McDuff, R.E., Chan, L.H., Collier, R., Grant, B., Gordon, L.I., Corliss, J.B., 1979. Ridge crest hydrothermal activity and the balances of the majorand minor elements in the ocean—Galapagos data. Earth Planet. Sci. Lett. 46, 1–18.

Ehrmann, W., Schmiedl, G., Seidel, M., Krüger, S., Schulz, H., 2016. A distal 140 kyr sediment record of Nile discharge and East African monsoon variability. Clim. Past, 12, 713–727. https://doi.org/10.5194/cp-12-713-2016.

Eilers, H., Pernthaler, J., Glöckner, F.O., Amann, R., 2000. Culturability an in situ abundance of pelagic bacteria from the North Sea. Appl.

Environ. Microbiol. 66, 3044–3051. http://dx.doi.org/10.1128/AEM. 66.7.3044-3051.2000.

El-Bouraie, M.M., El-Barbary, A.A., Yehia, M.M., Motawea, E.A., 2010. Heavy metal concentrations in surface river water and bed sediments at Nile Delta in Egypt. Suo. 61 (1), 1-12.

Elderfield, H., Schultz, A., 1996. Mid-ocean ridge hydrothermal fluxes and the chemical composition of the ocean. Annu. Rev. Earth Planet. Sci. 24, 191–224. http://dx.doi.org/10.1146/annurev.earth.24.1.191.

Emeis, K.C., Robertson, A.H.F., Richter, C., et al., 1996. Proc. ODP, Init. Repts., 160: College Station, TX (Ocean Drilling Program). doi:10.2973/odp.proc.ir.160.1996.

Emeis, K., Sakamoto, T., Wehausen, R., Brumsack, H., 2000. The sapropel record of the eastern Mediterranean Sea — results of Ocean Drilling Program Leg 160. Palaeogeogr. Palaeoclimatol. Palaeoecol. 158 (3–4), 371–395. https://doi.org/10.1016/S0031-0182(00)00059-6.

Emeis, K.C., Schulz, H., Struck, U., Rossignol-Strick, M., Erlenkeuser, H., Howell, M.W., Kroon, D., Mackensen, A., Ishizuka, S., Oba, T., Sakamoto, T., Koizumi, I., 2003. Sapropel cycles in the Mediterranean Sea. PANGAEA, https://doi.org/10.1594/PANGAEA.842312, Supplement to: Emeis, K.C. et al. (2003): Eastern Mediterranean surface water temperatures and d18O composition during deposition of sapropels in the late Quaternary. Paleoceanogr. 18 (1), 1005. https://doi.org/10.1029/2000PA000617.

Emelianov, M., Font, J., Turiel, A., Millot, C., Solé, J., Poulain, P.-M., Julià, A., and Vitrià, M.-R., 2006. Transformation of Levantine Intermediate Water tracked by MEDARGO floats in the Western Mediterranean. Ocean Sci. 2, 281–290. https://doi.org/10.5194/os-2-281-2006.

Emerson, S., Hedges, J., 2006. Sediment diagenesis and benthic fluxes. In: The Oceans and Marine Geochemistry, Treatise on Geochemistry: Elsevier 6, 293-319.

Eppley, R., Peterson, B., 1979. Particulate organic matter flux and planktonic new production in the deep ocean. Nature 282, 677–680. https://doi.org/10.1038/282677a0.

Fagel, N., Dehairs, F., André, L., Bareille, G., Monnin, C., 2002. Ba distribution in surface Southern Ocean sediments and export production estimates. Paleoceanogr. 17 (2). https://doi.org/10.1029/2000PA000552, 2002.

Falkner, K.K., Klinkhammer, G.P., Bowers, T.S., Todd, J.F., Lewis, B.L., Landing, W.M., Edmond, J.M., 1993. The behavior of barium in anoxic marine waters. Geochim. Cosmochim. Acta 57, 537–554. https://doi.org/10.1016/0016-7037(93)90366-5.

Falkner, K.K., O'Neill, D.J., Todd, J.F., Moore, W.S., Edmond, J.M., 1991. Depletion of Ba and Ra-226 in Black Sea surface waters over the past thirty years. Nature, 350, 491-494. https://doi.org/10.1038/350491a0.

Falkowski, R., Scholes, R.J., Boyle, E., Canadell, J., Canfield, D., Elser, J., Gruber, N., Hibbard, K., Högberg, P., Linder, S., Mackenzie, F.T., Moore, B., Pedersen, T., Rosenthal, Y., Sieitzinger, S., Smetacek, V., Steffen, W., 2000. The Global Carbon Cycle: A Test of Our Knowledge of Earth as a System. Science 290(5490), 291–296. https://doi.org/10.1126/science.290.5490.291.

Fichaut M., Garcia M.J., Giorgetti A., Iona A., Kuznetsov A., Rixen M., Medar Group, 2003. MEDAR/MEDATLAS 2002: A Mediterranean and Black Sea database for operational oceanography Elsevier Oceanography Series. Building the European Capacity in Operational Oceanography, Proceedings of the Third International Conference on EuroGOOS Athens, Greece 3–6 December 2002. Elsevier Oceanogr Series 69. Edited by H. Dahlin, N.C. Flemming, K. Nittis and S.E. Petersson ISBN: 978-0-444-51550-6. 645–648. https://archimer.ifremer.fr/doc/00351/46177/.

Filippidi, A., De Lange, G.J., 2019. Eastern Mediterranean Deep Water Formation During Sapropel S1: A Reconstruction Using Geochemical Records Along a Bathymetric Transect in the Adriatic Outflow Region. Paleoceanogr Paleoclim 34, 409–429. https://doi.org/10.1029/2018PA003459.

Fowle, D.A., Fein, J.B., 2001. Quantifying the effects of *Bacillus subtilis* cell walls on the precipitation of copper hydroxide from aqueous solution. Geomicrobiol. J, 18, 77–91. https://doi.org/10.1080/01490450151079815.

Fox, G.E., Wisotzkey, J.D., Jurtshuk, P., 1992. How close is close: 16S RNA sequence identity may not be sufficient to guarantee species identity. Int. J. Syst. Bacteriol. 42, 166–170. http://dx.doi.org/10.1099/00207713-42-1-166.

Francois, R., Honjo, S., Manganini, S. J., Ravizza, G. E., 1995. Biogenic barium fluxes to the deep sea: Implications for paleoproductivity reconstruction. Global Biogeochem. Cycles 9(2), 289– 303. http://dx.doi.org/10.1029/95GB00021.

Fresnel, J., Galle, P., Gayral, P., 1979. Resultats de la microanalyse des cristaux vacuolaires chez deux Chromophytes unicellulaires marines: *Exanthemachrysis gayraliae, Pavlova* sp, (*Pryrnnesiophycees, Pavlovacees*). CR.Acad. Sci. Paris 288D, 823–825.

Frigola, J., Moreno, A., Cacho, I., Canals, M., Sierro, F.J., Flores, J.A., Grimalt, J.O., 2008. Evidence of abrupt changes in Western Mediterranean Deep Water circulation during the last 50kyr: A high-resolution marine record from the Balearic Sea. Quat. Internat. 181 (1), 88-104. https://doi.org/10.1016/j.quaint.2007.06.016.

Gadd, G.M., 2009. Biosorption: Critical review of scientific rationale, environmental importance and significance for pollution treatment. J. Chem. Tech. and Biotec. 84 (1), 13–28. https://doi.org/10.1002/jctb.1999.

Gaillardet, J., Viers, J., Dupré, B., 2003. 5.09 - Trace Elements in River Waters. In: Treatise on Geochemistry, Pergamon, 225-272. https://doi.org/10.1016/B0-08-043751-6/05165-3.

Gallego-Torres, D., Martínez-Ruiz, F., De Lange, G.J., Jimenez-Espejo, F.J., Ortega-Huertas, M., 2010. Trace-elemental derived paleoceanographic and paleoclimatic conditions for Pleistocene Eastern Mediterranean sapropels. Palaeogeogr. Palaeoclimatol. Palaeoecol. 293 (1–2), 76–89. https://doi.org/10.1016/j.palaeo.2010.05.001.

Gallego-Torres, D., Martinez-Ruiz, F., Meyers, P. A., Paytan, A., Jimenez-Espejo, F. J., and Ortega-Huertas, M., 2011. Productivity patterns and N-fixation associated with Pliocene-Holocene sapropels: paleoceanographic and paleoecological significance. Biogeosci 8, 415–431. https://doi.org/10.5194/bg-8-415-2011.

Gallego-Torres, D., Martínez-Ruiz, P., Paytan, A., Jiménez-Espejo, F.J., Ortega-Huertas, M., 2007. Pliocene–Holocene evolution of depositional conditions in the eastern

Mediterranean: Role of anoxia vs. productivity at time of sapropel deposition. Palaeogeogr. Palaeoclimatol. Palaeoecol. 246 (2–4), 424-439. https://doi.org/10.1016/j.palaeo.2006.10.008.

Ganeshram, R.S., Francois, R., Commeau, J., Brown-Leger, S.L., 2003. An experimental investigation of barite formation in seawater. Geochim. Cosmochim. Acta 67, 2599–2605. http://dx.doi.org/10.1016/s0016-7037(03)00164-9.

García-Martínez, J., Acinas, S.G., Rodríguez-Valera, F., 2002. Prevalence and microdiversity of *Alteromonas macleodii*-like microorganisms in different oceanic regions. Environ. Microbiol. 4, 42–50. http://dx.doi.org/10.1046/j.1462-2920.2002.00255.x.

Garcia-Solsona, E., Garcia-Orellana, J., Masqué, P., Garcés, E., Radakovitch, O., Mayer, A., Estradé, S., Basterretxea, G., 2010. An assessment of karstic submarine groundwater and associated nutrient discharge to a Mediterranean coastal area (Balearic Islands, Spain) using radium isotopes. Biogeochemistry 97, 211–229. https://doi.org/10.1007/s10533-009-9368-y.

Garcia-Solsona, E., Jeandel, C., 2020. Balancing Rare Earth Element distributions in the Northwestern Mediterranean Sea. Chem. Geol. 532, 119372. https://doi.org/10.1016/j.chemgeo.2019.119372.

Gauthier, M.J., Breittmayer, V.A., 1992. The genera *Alteromonas* and *Marinomonas*. In: The Prokaryotes vol. 3. Springer, New York, 3046–3070.

Gauthier, G., Gauthier, M., Christen, R., 1995. Phylogenetic analysis of the genera *Alteromonas, Shewanella* and *Moritella* using genes coding for small-subunit rRNA sequences and division of the genus *Alteromonas* into two genera, *Alteromonas* (emended) and *Pseudoalteromonas* gen. nov., and proposal of twelve new species combinations. Int. J. Syst. Bacteriol. 45, 755–761. http://dx.doi.org/10.1099/00207713-45-4-755.

Geesey, G.G., 1982. Microbial exopolymers: ecological and economic considerations. ASM News 48, 9–14.

Geuer, J., 2015. Quantification and molecular characterisation of dissolved organic sulphur at the benthopelagic interface in the Weddell Sea. Master thesis, University of Applied Sciences Bremerhaven.

Gilman, C., Garrett, C., 1994. Heat flux parameterizations for the Mediterranean Sea: The role of atmospheric aerosols and constraints from the water budget. J. Geophys. Res. 99, 5119–513494. https://doi.org/10.1029/93JC03069

Gingele, F.X., Zabel, M., Kasten, S., Bonn, W.J., Nürnberg, C.C., 1999. Biogenic Barium as a Proxy for Paleoproductivity: Methods and Limitations of Application. In: Use of Proxies in Paleoceanography. Springer, Berlin, Heidelberg.

Glamoclija, M., Garrel, L., Berthon, J., López-García, P., 2004. Biosignatures and bacterial diversity in hydrothermal deposits of Solfatara Crater, Italy. Geomicrobiol. J. 21 (8), 529–541. http://dx.doi.org/10.1080/01490450490888235.

Goldberg, E.D., 1954. Marine Geochemistry 1. Chemical scavengers of the sea. J. Geol. 62, 249-265.

Goldberg, E.D., Arrhenius, G., 1958. Chemistry of pelagic sediments. Geochim. Cosmochim. Acta 13, 153–212. http://dx.doi.org/10.1016/0016-7037(58)90046-2.

Gomis, D., Tsimplis, M.N., Martín-Míguez, B., Ratsimandresy, A.W., García-Lafuente, J., Josey, S.A., 2006. Mediterranean Sea level and barotropic flow through the Strait of Gibraltar for the period 1958–2001 and reconstructed since 1659. J. Geophys. Res. 111, C11005. http://dx.doi.org/10.1029/2005JC003186

Gonneea, M.E., Mulligan, A.E., Charette, M.A., 2013. Climate-driven sea level anomalies modulate coastal groundwater dynamics and discharge. Geophys. Res. Lett., 40, 2701–2706, http://dx.doi.org/10.1002/grl.50192.

González-Benítez, N., García-Corral, L.S., Morán, X.A.G., Middelburg, J.J., Pizay, M.D., Gattuso, J.P., 2019. Drivers of Microbial Carbon Fluxes Variability in Two Oligotrophic Mediterranean Coastal Systems. Sci. Rep. 9, 17669. https://doi.org/10.1038/s41598-019-53650-z.

González-Muñoz, M.T., de Linares, C., Martínez-Ruiz, F., Morcillo, F., Martín-Ramos, D., Arias, J.M., 2008. Ca–Mg kutnahorite and struvite production by *Idiomarina* strains at modern seawater salinities. Chemosphere 72 (3), 465–472. http://dx.doi.org/10.1016/j.chemosphere.2008.02.010.

González-Muñoz, M.T., Fernández-Luque, B., Martínez-Ruiz, F., Ben Chekroun, K., Arias, J.M., Rodriguez-Gallego, M., Martínez-Cañamero, M., de Linares, C., Paytan, A., 2003. Precipitation of barite by Myxococcus xanthus: possible implications for the biogeochemical cycle of barium. Appl. Environ. Microbiol. 69 (9), 5722–5725. http://dx.doi.org/10.1128/aem.69.9.5722-5725.2003.

González-Muñoz, M.T., Martínez-Ruiz, F., Morcillo, F., Martín-Ramos, J.D., Paytan, A., 2012. Precipitation of barite by marine bacteria: a possible mechanism for marine barite formation. Geol. 40, 675–678. http://dx.doi.org/10.1130/G33006.1.

González-Muñoz, M.T., Merroun, M.L., Ben Omar, N., Arias, J.M., 1997. Biosorption of uranium by *Myxococcus xanthus.* Int. Biodeter. Biodegr. 40, 107–114. https://doi.org/10.1016/S0964-8305(97)00041-3.

González-Muñoz, M.T., Rodriguez-Navarro, C., Martínez-Ruiz, F., Arias, J.M., Merroun, M.L., Rodriguez-Gallego, M., 2010. Bacterial biomineralization: new insights from *Myxococcus*-induced mineral precipitation. Geol. Soc., London, Special Publications 336, 31–50. https://doi.org/10.1144/SP336.3.

Gooday, A.J., Nott J.A., 1982. IntracelIular barite crystals in two xenopbyophores, *Aschemonella ramuliformis* and *Galatheammina* sp. (protozoa: Rhizopoda) with comments on the taxonomy of *A. ramuliformis*. J. Mar. Biol. Assoc. U.K. 62, 595-605.

Grant, K.M., Grimm, R., Mikolajewicz, U., Marino, G., Ziegler, M., Rohling, E.J., 2016. The timing of Mediterranean sapropel deposition relative to insolation, sea-level and African monsoon changes. Quat. Sci. Rev. 140, 125–141. https://doi.org/10.1016/j.quascirev.2016.03.026.

Grasshoff, K., Ehrhardt, M., Kremling, K., 1983. Determination of alkalinity and total carbonate. Methods of Seawater Analysis, Verlag Chemie, Weinheim 99-123.

Grazzini, C., 1975. ^{18}O Changes in foraminifera carbonates during the last 10 years in the Mediterranean Sea. Science 190 (4211), 272-274. http://www.jstor.org/stable/1740061.

Green, D.H., Bowman, J.P., Smith, E.A., Gutierrez, T., Bolch, C.J., 2006. *Marinobacter algicola* sp. nov., isolated from laboratory cultures of paralytic shellfish toxinproducing dinoflagellates. Int. J. Syst. Evol. Microbiol. 56 (Pt 3), 523–527. http://dx.doi.org/10.1099/ijs.0.63447-0.

Grimm, R., Maier-Reimer, E., Mikolajewicz, U. Schmiedl, G., Müller-Navarra, K., Adloff, F., Grant, K.M., Ziegler, M., Lourens, L.J., Emeis, K.C., 2015. Late glacial initiation of Holocene eastern Mediterranean sapropel formation. Nat. Commun. 6, 7099. https://doi.org/10.1038/ncomms8099.

Griffith, E.M., Paytan, A., 2012. Barite in the ocean-occurrence, geochemistry and palaeoceanographic applications. Sediment. 59, 1817–1835. http://dx.doi.org/10.1111/j.1365-3091.2012.01327.x.

Griffith, E.M., Paytan, A., Wortmann, U.G., Eisenhauer, A., Scher, H.D., 2018. Combining metal and nonmetal isotopic measurements in barite to identify mode of formation. Chem. Geol. 500, 148–158. https://doi.org/10.1016/j.chemgeo.2018.09.031.

Grissom, K.R., 2015. The Distribution of Dissolved Barium Along the U.S. Geotraces North Atlantic Transect: Impact of Biogeochemical Processes, Circulation and Phase Association. Master's Theses, 118. https://aquila.usm.edu/masters_theses/118.

Guay, C.K.H., McLaughlin, F.A., Yamamoto-Kawai, M., 2009. Differentiating fluvial components of upper Canada Basin waters on the basis of measurements of dissolved barium combined with other physical and chemical tracers. J. Geophys. Res., 114, C00A09. https://doi.org/10.1029/2008JC005099.

Guerzoni, S., Chester, R., 1996. The Impact of Desert Dust Across the Mediterranean. Kluwer Acad. Publisher, Dordrecht, The Netherlands.

Guerzoni, S., Chester, R., Dulac, F., Herut, B., Loÿe-Pilot, M.D., Measures, C., Migon, C., Molinaroli, E., Moulin, C., Rossini, P., Saydam, C., Soudine, A., Ziveri, P., 1999. The role of atmospheric deposition in the biogeochemistry of the Mediterranean Sea. Progr. in Oceanogr. 44 (1–3), 147-190. https://doi.org/10.1016/S0079-6611(99)00024-5.

Guerzoni, S., Molinaroli, E., 2005. Input of Various Chemicals Transported by Saharan Dust and Depositing atthe Sea Surface in the Mediterranean Sea. In: The Mediterranean Sea. Handbook of Environmental Chemistry, vol 5K. Springer, Berlin, Heidelberg. https://doi.org/10.1007/b107149.

Guibaud, G., Bordas, F., Saaid, A., D'abzac, P., Van Hullebusch, E., 2008. Effect of pH on cadmium and lead binding by extracellular polymeric substances (EPS) extracted from environmental bacterial strains. Colloids and Surfaces B: Biointerfaces 63 (1), 48-54. https://doi.org/10.1016/j.colsurfb.2007.11.002.

Guieu, C., Chester, R., Dulac, F., Herut, B., Loÿe-Pilot, M.D., Measures, C., Migon, C., Molinaroli, E., Moulin, C., Rossini, P., Saydam, C., Soudine, A., Ziveri, P., 1999. The role of atmospheric deposition in the biogeochemistry of the Mediterranean Sea. Progr. Oceanogr. 44 (1–3), 147-190.https://doi.org/10.1016/S0079-6611(99)00024-5.

Guieu, C., Chester, R., Nimmo, M., Martin, J.M., Guerzoni, S., Nicolas, E., Mateu, J., Keyse, S., 1997. Atmospheric input of dissolved and particulate metals to the northwestern Mediterranean. Deep Sea Res. Part II: Topical Studies in Oceanogr. 44 (3–4), 655-674. https://doi.org/10.1016/S0967-0645(97)88508-6.

Guieu, C., Loÿe-Pilot, M.D., Benyahya, L., Dufour, A., 2010. Spatial variability of atmospheric fluxes of metals (Al, Fe, Cd, Zn and Pb) and phosphorus over the whole Mediterranean from a one-year monitoring experiment: Biogeochemical implications. Mar. Chem. 120 (1–4), 164-178. https://doi.org/10.1016/j.marchem.2009.02.004.

Guieu, C., Roy-Barman, M., Leblond, N., Jeandel, C., Souhaut, M., Le Cann, B., Dufour, A., Bournot, C., 2005. Vertical particle flux in the northeast Atlantic Ocean (POMME experiment). J. Geoph. Res. 110, C07S18. http://dx.doi.org/ 10.1029/2004JC002672.

Greinert, J., Bollwerk, S.M., Derkachev, A., Bohrmann, G., Suess, E., 2002. Massive barite deposits and carbonate mineralization in the Derugin Basin, Sea of Okhotsk: precipitation processes at cold seep sites. Earth and Planet. Sci. Lett. 203 (1), 165-180. https://doi.org/10.1016/S0012-821X(02)00830-0.

Halim, Y., 1991. The impact of human alterations of the hydrological cycle on ocean margins. In: Ocean Margin Processes in Global Change. John Wiley and Sons, 301–27.

Hall, T.A., 2007. BioEdit v7.1.11 2007 [online]. http://www.mbio.ncsu.edu/BioEdit/page2.html (Website last modified on January 29, 2013 (accessed on January 31, 2013). Available at). Healy, M., Huong, J., Bittner, T., Lising, M., Frye, S., Raza, S., Schrock, R., Manry, J., Renwick, A., Nieto, R., Woods, C., Versalovic, J., Lupski, J.R., 2005. Microbial DNA typing by automated repetitive sequence-based PCR. J. Clin. Microbiol. 43, 199–207. http://dx.doi.org/10.1128/JCM.43.1.199-207.2005.

Hamza, W., 2005. The Nile Estuary. In: Estuaries. The Handbook of Environmental Chemistry, vol 5H. Springer, Berlin, Heidelberg. https://doi.org/10.1007/698_5_025.

Han, Q., 2010. Crustal Tracers in the Atmosphere and Ocean: Relating their Concentrations, Fluxes, and Ages. Ph.D. thesis University of California, Irvine.

Han, Q., Moore, J. K., Zender, C., Measures, C., Hydes, D., 2008. Constraining oceanic dust deposition using surface ocean dissolved Al. Global Biogeochem. Cycles 22, GB2003. http://dx.doi.org/10.1029/2007GB002975.

Hansel, l.D., Carlson, C., 2015. Biogeochemistry of Marine Dissolved Organic Matter (Second Edition). Acad. Press, 65–126. https://doi.org/10.1016/B978-0-12-405940-5.00003-0.

Hansell, D.A., 2013. Recalcitrant dissolved organic carbon fractions. Annu. Rev. Mar. Scib. 5, 421–445. http://dx.doi.org/10.1128/JCM.43.1.199-207.2005.

Hartnett, H., Keil, R., Hedges, J., Devol., A.H., 1998. Influence of oxygen exposure time on organic carbon preservation in continental margin sediments. Nature 391, 572–575. https://doi.org/10.1038/35351.

Harzallah, A., Cadet, D.L., Crepon, M., 1993. Possible forcing effects of net evaporation, atmospheric pressure, and transients on water transports in the Mediterranean Sea. J. Geophys. Res. 98 (C7), 12341– 12350. https://doi.org/10.1029/93JC00376.

Healy, M., Huong, J., Bittner, T., Lising, M., Frye, S., Raza, S., Schrock, R., Manry, J., Renwick, A., Nieto, R., Woods, C., Versalovic, J., Lupski, J.R., 2005. Microbial DNA typing by automated repetitive sequence-based PCR. J. Clin. Microbiol. 43, 199–207. http://dx.doi.org/10.1128/JCM.43.1.199-207.2005.

Heath, R.T., Moore, T.C., Dauphin, J.L., 1977. Organic carbon in deep-sea sediments. In: The Fate of Fossil Fuel CO_2 in the Oceans, Plenum, New York, 605-625.

Hedges, J.I., Keil, R.G., 1995. Sedimentary organic matter preservation: an assessment and speculative synthesis. Mar. Chem. 49(2–3), 81–115. https://doi.org/10.1016/0304-4203(95)00008-F.

Heimbürger, L.E., Christophe, M., Losno, R., Miquel, J.C., Thibodeau, B., Stabholz, M., Dufour, A., Leblond, N., 2014. Vertical export flux of metals in the Mediterranean Sea. Deep Sea Res. Part I: Oceanogr. Res. Papers 87, 14-23. https://doi.org/10.1016/j.dsr.2014.02.001.

Hemleben, C., Becker, T., Bellas, S., Benningsen, G., Casford, J., Cagatay, N., Emeis, K.C., Engelen, B., Ertan, T., Fontanier, C., Friedrich, O., Frydas, D., Giunta, S., Hoffelner, H., Jorissen, F.J., Kahl, K., Kaszemeik, K., Lykousis, V., Meier, S., Nickel, J. Overman, J. Pross, T. Reichel, C. Robert, E. Rohling, W. Ruschmeier, M. Sakinc, G., Schiebel, R., Schmiedl, G., Schubert,K., Schulz, H., Tischnak, J., Truscheit, T., 2003. Ostatlantik-Mittelmeer-Schwarzes Meer Part 3 Cruise No. 51, Leg 3 14 November–10 December 2001, Valetta – Istanbul: METEOR-Berichte 03–1. Institut für Meereskunde der Universität Hamburg.

Hennekam, R., Jilbert, T., Schnetger, B., De Lange, G. J., 2014. Solar forcing of Nile discharge and sapropel S1 formation in the early to middle Holocene eastern Mediterranean. Paleoceanogr. 29, 343– 356. https://doi.org/10.1002/2013PA002553.

Herut, B., Krom, M., 1996. Atmospheric Input of Nutrients and Dust to the SE Mediterranean. In: The Impact of Desert Dust Across the Mediterranean. Environ. Sci. and Tech. Library 11. Springer, Dordrecht. https://doi.org/10.1007/978-94-017-3354-0_35.

Hieke, W., Hemleben, C., Linke, P., Türkay, M., Weikert, Mittelmeer, H., 1999. 1997/98, Cruise No 40: 28. October 1997–10. February 1998.: Meteor-Berichte 99–2. Institut für Meereskunde der Universität Hamburg.

Higgins, M.B., Robinson, R.S., Carter, S.J., Pearson, A., 2010. Evidence from chlorin nitrogen isotopes for alternating nutrient regimes in the Eastern Mediterranean Sea. Earth Planet. Sci. Lett. 290 (1–2), 102-107. https://doi.org/10.1016/j.epsl.2009.12.009.

Hilgen, F.J., 1991. Astronomical calibration of Gauss to Matuyama sapropels in the Mediterranean and implication for the Geomagnetic Polarity Time Scale. Earth Planet. Sci. Let.104 (2–4), 226–244. https://doi.org/10.1016/0012-821X(91)90206-W.

Hilgen, F.J., Abdul Aziz, H., Krijgsman, W., Raffi, I., Turco, E., 2003. Integrated stratigraphy and astronomical tuning of the Serravallian and lower Tortonian at Monte dei Corvi (Middle-Upper Miocene, northern Italy). Palaeogeogr. Palaeoclimatol. Palaeoecol. 199, 229–264. https://doi.org/10.1016/S0031-0182(03)00505-4.

Hilgen, F.J., Krijgsman, W., Langereis, C.G., Lourens, L.J., Santarelli, A., Zachariasse, W.J., 1995. Extending the astronomical (polarity) time scale into the Miocene. Earth Planet. Sci. Let. 136 (3–4), 495–510. https://doi.org/10.1016/0012-821X(95)00207-S.

Hilgen, F.J., Lourens, L.J., Berger, A., Loutre, M.F., 1993. Evaluation of the astronomically calibrated time scale for the Late Pliocene and Earliest Pleistocene. Paleocean. 8 (5), 549– 565. https://doi.org/10.1029/93PA01248.

Holmström, C., Egan, S., Franks, A., McCloy, S., Kjelleberg, S., 2002. Antifouling activities expressed by marine surface associated *Pseudoalteromonas* species. FEMS Microbiol. Ecol. 41, 47–58. http://dx.doi.org/10.1111/j.1574-6941.2002.tb00965.x.

Holmström, C., James, S., Egan, S., Kjelleberg, S., 1996. Inhibition of common fouling organisms by marine bacterial isolates with special reference to the role of pigmented bacteria. Biofouling 10, 251–259. http://dx.doi.org/10.1080/08927019609386284.

Hoppema, M., Dehairs, F., Navez, J., Monnin, C., Jeandel, C., Fahrbach, E., de Baar, H.J.W., 2010. Distribution of barium in the Weddell Gyre: Impact of circulation and biogeochemical processes. Mar. Chem. 122 (1–4), 118–129. https://doi.org/10.1016/j.marchem.2010.07.005.

Horner, T.J., Kinsley, C.W., Nielsen, S.G., 2015. Barium-isotopic fractionation in seawater mediated by barite cycling and oceanic circulation. Earth and Planet. Sci.Let.430, 511–522. https://doi.org/10.1016/j.epsl.2015.07.027.

Horner, T.J., Pryer, H.V., Nielsen, SG.. 2017. Crockford PW, Gauglitz JM, Wing BA and Ricketts RD. Pelagic barite precipitation at micromolar ambient sulfate. Nat. Commun. 8, 1342. https://doi.org/10.1038/s41467-017-01229-5.

Howard, L.S., Brown, B.E., 1987. Metals in Pocillopora damicornis exposed to tin smelter effluent. Mar. Pollut. Bullet. (8), 451-454. https://doi.org/10.1016/0025-326X(87)90623-0.

Hsieh, Y.T., Henderson, G.M., 2017. Barium stable isotopes in the global ocean: Tracer of Ba inputs and utilization. Earth Planet. Sci. Lett. 473, 269-278. https://doi.org/10.1016/j.epsl.2017.06.024.

Hsu, S.C., Wong, G.T.F., Gong, G.C., Shiah, F.K., Huang, Y.T., Kao, S.J., Tsai, F., Lung, S.C.C., Lin, F.J., Lin, I.I., Hung, C.C., Tseng, C.M., 2010. Sources, solubility, and dry deposition of aerosol trace elements over the East China Sea. Mar. Chem. 120 (1–4), 116-127. https://doi.org/10.1016/j.marchem.2008.10.003.

Huang, Y.L., Dobretsov, S., Xiong, H., Qian, P.Y., 2007. Effect of biofilm formation by *Pseudoalteromonas spongiae* on induction of larval settlement of the polychaete *Hydroides elegans*. Appl. Environ. Microbiol. 73, 6284–6288. http://dx.doi.org/10.1128/AEM.00578-07.

Huertas, I.E., Ríos, A.F., García-Lafuente, J., Navarro, G., Makaoui, A., Sánchez-Román, A., Rodriguez-Galvez, S., Orbi, A., Ruíz, J., Pérez, F.F., 2012. Atlantic forcing of the Mediterranean oligotrophy. Global Biogeochem. Cycles 26, GB2022. http://dx.doi.org/10.1029/2011GB004167.

Hurrell, J.W., 1995. Decadal trends in the North Atlantic Oscillation, regional temperatures and precipitation. Sci. 269, 676-679. http://dx.doi.org/10.1126/science.269.5224.676.

Hurrell, J.W., Deser, C., 2010. North Atlantic climate variability: The role of the North Atlantic Oscillation. J. Mar. Syst. 79 (3–4), 231-244. https://doi.org/10.1016/j.jmarsys.2009.11.002.

Hydes, D.J., De Lange, G.J., De Baar, H.J.W., 1988. Dissolved aluminium in the Mediterranean. Geochim. Cosmochim. Acta 52 (8), 2107-2114. https://doi.org/10.1016/0016-7037(88)90190-1.

Ibach, L.E.J., 1982. Relationship between sedimentation rate and total organic carbon content in ancient marine sediments. AAPG Bull. 66, 170-188. https://doi.org/10.1306/03B59A5D-16D1-11D7-8645000102C1865D.

Ignatiades, L., Gotsis-Skretas, O.l., Pagou, K., Krasakopoulou, E., 2009. Diversification of phytoplankton community structure and related parameters along a large east-west transect of the Mediterranean Sea. Journ. Plank. Res. 31, 441–448. https://doi.org/10.1093/plankt/fbn124.

Iijima, S., Washio, K., Okahara, R., Morikawa, M., 2009. Biofilm formation and proteolytic activities of *Pseudoalteromonas* bacteria that were isolated from fish farm sediments. Microb. Biotechnol. 2, 361–369. http://dx.doi.org/10.1111/j.1751-7915.2009.00097.x.

Ishii, M., Egen, J.G., Klauschen, F., Meier-Schellersheim, M., Saeki, Y., Vacher, J., Proia, R.L., Germain, R.N., 2009. Sphingosine-1-phosphate mobilizes osteoclast precursors and regulates bone homeostasis. Nature 458, 524–528. http://dx.doi.org/10.1038/nature07713.

Ishii, S., Sadowsky, M.J., 2009. Applications of the rep-PCR DNA fingerprinting technique to study microbial diversity, ecology and evolution. Environ. Microbiol. 11, 733–740. http://dx.doi.org/10.1111/j.1462-2920.2008.01856.x.

Ivanova, E.P., Bowman, J.P., Lysenko, A.M., Zhukova, N.V., Gorshkova, N.M., Sergee, A.F., Mikhailov, V.V., 2005. *Alteromonas addita* sp. nov. Int. J. Syst. Evol. Microbiol. 55 (3), 1065–1068. http://dx.doi.org/10.1099/ijs.0.63521-0.

Ivanova, E.P., Romanenko, L.A., Chun, J., Matte, M.H., Matte, G.R., Mikhailov, V.V., Svetashev, V.I., Huq, A., Maugel, T., Colwell, V., 2000. *Idiomarina* gen. nov., comprising novel indigenous deep-sea bacteria from the Pacific Ocean, including descriptions of two species, *Idiomarina abyssalis* sp. nov. and *Idiomarina zobellii* sp. nov. Int. J. Syst. Evol. Microbiol. 50, 901–907. http://dx.doi.org/10.1099/00207713-50-2-901.

Jacquet, S.H.M., Dehairs, F., Cardinal, D., Navez, J., Delille, B., 2005. Barium distribution across the Southern Ocean frontal system in the Crozet–Kerguelen Basin. Mar. Chem. 95(3–4), 149–162. https://doi.org/10.1016/j.marchem.2004.09.002.

Jacquet, S.H.M., Dehairs, F., Dumont, I., Becquevort, S., Cavagna, A.J., Cardinal, D., 2011. Twilight zone carbon remineralization in the Polar front Zone and Subantarctic Zone south of Tasmania. Deep-Sea Res. II 58, 2222–2234. http://dx.doi.org/10.1016/j.dsr2.2011.05.029.

Jacquet, S.H.M., Dehairs, F., Lefèvre, D., Cavagna, A.J., Planchon, F., Christaki, U., Monin, L., André, L., Closset, I., Cardinal, D., 2015. Early spring mesopelagic carbon remineralization and transfer efficiency in the naturally iron-fertilized Kerguelen area. Biogeosci. 12, 1713–1731. https://doi.org/10.5194/bg-12-1713-2015.

Jacquet, S.H.M., Dehairs, F., Rintoul, S., 2004. A high resolution transect of dissolved barium in the Southern Ocean. Geophys. Res. Lett. 31, L14301. http://dx.doi.org/10.1029/2004GL020016.

Jacquet, S.H.M., Dehairs, F., Savoye, N., Obernosterer, I., Christaki, U., Monnin, C., Cardinal, D., 2008. Mesopelagic organic carbon remineralization in the Kerguelen Plateau region tracked by biogenic particulate Ba. Deep Sea Res. Part II: Topical Studies in Oceanogr. 55 (5–7), 868–879. https://doi.org/10.1016/j.dsr2.2007.12.038.

Jacquet, S.H.M., Henjes, J., Dehairs, F., Worobiec, A., Savoye, N., Cardinal, D., 2007. Particulate Ba-barite and acantharians in the Southern Ocean during the European Iron Fertilization Experiment (EIFEX). J. Geophys. Res. 112 (G4). http://dx.doi.org/10.1029/2006JG000394

Jacquet, S.H.M., Monnin, C., Riou, V., Jullion, L., Tanhua, T., 2016. A high resolution and quasi-zonal transect of dissolved Ba in the

Mediterranean Sea. Mar. Chem. 178(1–7), 0304-4203. https://doi.org/10.1016/j.marchem.2015.12.001.

Jacquet, S.H.M., Tamburini, C., Garel, M., Dufour, A., Van Vambeke, F., Le Moigne, F. A. C., Bhairy, N., Guasco, S., 2020. Particulate biogenic barium tracer of mesopelagic carbon remineralization in the Mediterranean Sea (PEACETIME project), Biogeosci. Discuss. https://doi.org/10.5194/bg-2020-271.

Jeandel, C., Dupré, B., Lebaron, G., Monnin, C., Minster, J.F., 1996. Longitudinal distributions of dissolved barium, silica and alkalinity in the western and southern Indian Ocean. Deep Sea Res Part I: Oceanogr Res Papers 43(1), 1–31. https://doi.org/10.1016/0967-0637(95)00098-4.

Jeandel, C., Oelkers, E.H., 2015. The influence of terrigenous particulate material dissolution on ocean chemistry and global element cycles. Chem. Geol. 395, 50-66. https://doi.org/10.1016/j.chemgeo.2014.12.001.

Jeandel, C., Tachikawa, K., Bory, A., Dehairs, F., 2000. Biogenic barium in suspended and trapped material as a tracer of export production in the tropical NE Atlantic (EUMELI sites). Mar. Chem. 71 (1–2), 125-142. https://doi.org/10.1016/S0304-4203(00)00045-1.

Jickells, T., 1995. Atmospheric inputs of metals and nutrients to the oceans: their magnitude and effects. Mar. Chem. 48 (3–4), 199-214. https://doi.org/10.1016/0304-4203(95)92784-P.

Jickells, T. D., Buitenhuis, E., Altieri, K., Baker, A.R., Capone, D., Duce, R.A., Dentener, F., Fennel, K., Kanakidou, M., LaRoche, J., Lee, K., Liss, P., Middelburg, J.J., Moore, J.K., Okin, G., Oschlies, A., Sarin, M., Seitzinger, S., Sharples, J., Singh, A., Suntharalingam, P., Uematsu, M., Zamora, L.M., 2017. A reevaluation of the magnitude and impacts of anthropogenic atmospheric nitrogen inputs on the ocean. Global Biogeochem. Cycles 31, 289– 305. http://dx.doi.org/10.1002/2016GB005586.

Jiménez-Espejo, F.J., Martínez-Ruiz, F., Finlayson, C., Paytan, A., Sakamoto, T., Ortega-Huertas, M., Finlayson, G., Iijima, K., Gallego-Torres, D., Fa, D., 2007. Climate forcing and Neanderthal extinction in Southern Iberia: insights from a multiproxy marine record. Quat. Sci. Rev. 26 (7–8), 836-852. https://doi.org/10.1016/j.quascirev.2006.12.013.

Jiménez-Espejo, F.J., Martínez-Ruiz, F., Sakamoto, T., Iijima, K., Harada, N., 2006. Paleoenvironmental changes in the Baleraric basin since the last glacial: linking western and eastern climate responses in the Mediterranean. Palaeogeogr. Palaeoclimatol. Palaeoecol. https://doi.org/10.1016/j.palaeo.2006.10.005.

Josey, S.A., Marsh, R., 2005. Surface freshwater flux variability and recent freshening of the North Atlantic in the eastern subpolar gyre. J. Geophys. Res. 110, C05008. https://doi.org/10.1029/2004JC002521.

Josey, S.A., Somot, S., Tsimplis, M., 2011.. Impacts of atmospheric modes of variability on Mediterranean Sea surface heat exchange. J. Geophys. Res. 116, C02032. https://doi.org/10.1029/2010JC006685.

Jroundi, F., Fernández-Vivas, A., Rodriguez-Navarro, C., Bedmar, E.J., González-Muñoz, M.T., 2010. Bioconservation of deteriorated monumental calcarenite stone and identification of bacteria with carbonatogenic activity. Microb. Ecol. 60 (1), 39–54. http://dx.doi.org/10.1007/s00248-010-9665-y.

Karditsa, A., Poulos, S.E., Botsou, F., Alexakis, D., Stamatakis, M., 2014. Investigation of major and trace elements distribution pattern and pollution status of the surficial sediments of a microtidal inner shelf influenced by a transboundary river. The case of the Alexandroupolis Gulf (northeastern Aegean Sea, Greece). J. Geochem. Explor.146, 105-118. https://doi.org/10.1016/j.gexplo.2014.08.004.

Karnaska, Y., Maderich, V., 2008. Modelling of seasonal exchange flows through the Dardanelles Strait. Estuar. Coast. Shelf Sci. 79 (3), 449-458. http://dx.doi.org/10.1016/j.ecss.2008.04.019.

Keim, P., Mock, M., Young, J., Koehler, T.M., 2006. The international *Bacillus anthracis, B. cereus, and B. thuringiensis* conference, "*Bacillus-ACT05*". J. Bacteriol. 188 (10), 3433–3441. http://dx.doi.org/10.1128/JB.188.10.3433-3441.2006.

Khatiwala, S., Primeau, F., Holzer, M., 2012. Ventilation of the deep ocean constrained with tracer observations and implications for radiocarbon estimates of ideal mean age. Earth and Planet. Sci. Lett. 325–326, 116-125. https://doi.org/10.1016/j.epsl.2012.01.038.

Ki, J.S., Zhang, W., Qian, P.Y., 2009. Discovery of marine *Bacillus* species by 16S rRNA and rpoB comparisons and their usefulness for species identification. J. Microbiol. Methods 77 (1), 48–57. http://dx.doi.org/10.1016/j.mimet.2009.01.003.

Kidd, R.B., Cita, M.B., Ryan, W.B.F., 1978. Stratigraphy of eastern Mediterranean sapropel sequences recovered during Leg 42A and their paleoenvironmental significance. Init. Rep. DSDP 42A, 421–443.

Kikuchi, T., Tanaka, S., 2012. Biological removal and recovery of toxic heavy metals in water environment. Crit. Rev. Environ. Sci. Technol. 42, 1007-1057. http://dx.doi.org/10.1016/j.jece.2013.12.019.

Kinder, T. H., Parrilla, G., 1987. Yes, some of the Mediterranean outflow does come from great depth. J. Geophys. Res. 92 (C3), 2901–2906. http://dx.doi.org/10.1029/JC092iC03p02901.

Klochko, V., Zelena, L.B., Voychuk, S.I., Andriy, A.N., Ostapchuk, N., 2012. Peculiarities of *Alteromonas macleodii* strains reflects their deep/surface habitation rather than geographical distribution. J. Gen. Appl. Microbiol. 58, 129–135. http://dx.doi.org/10.2323/jgam.58.129.

Konovalov, S.K. Murray, J.W., 2001. Variations in the chemistry of the Black Sea on a time scale of decades (1960-1995). J. Mar. Sys. 31,217-243.

Kotthoff, U., Pross J., Müller U.C., Peyron O., Schmiedl G., Schulz H., Bordon A., 2008. Timing and characteristics of terrestrial vegetation change in the NE Mediterranean region associated with the formation of marine Sapropel S1: A land-sea correlation. Quat. Sci. Rev. 27, 832–845.

Kress, N., Herut, B., 2001. Spatial and seasonal evolution of dissolved oxygen and nutrients in the Southern Levantine Basin (Eastern Mediterranean Sea): chemical characterization of the water masses and inferences on the N:P ratios. Deep Sea Res. Part I: Oceanogr.c Res. Papers 48 (11), 2347-2372. https://doi.org/10.1016/S0967-0637(01)00022-X.

Krichak, S., Alpert, P., 2005. Signatures of the NAO in the atmospheric circulation during wet winter months over the Mediterranean region. Theor. Appl. Climatol. 82, 27–39.

https://doi.org/10.1007/s00704-004-0119-7.

Krijgsman, W., Hilgen, F.J., Langereis, C.G., Santarelli, A., Zachariasse, W.J., 1995. Late Miocene magnetostratigraphy, biostratigraphy and cyclostratigraphy in the Mediterranean. Earth Planet. Sci. Let. 136 (3–4), 475-494. https://doi.org/10.1016/0012-821X(95)00206-R.

Krom, M.D., Brenner, S., Kress, N., Neori, A., Gordon, L.I., 1992. Nutrient dynamics and new production in a warm-core eddy from the Eastern Mediterranean Sea. Deep-Sea Res. 39, 467-480. https://doi.org/10.1016/0198-0149(92)90083-6.

Krom, M.D., Brenner, S., Israilov, L., Krumgalz, B., 1991. Dissolved nutrients, preformed nutrients and calculated elemental ratios in the south-east Mediterranean Sea. Oceanol. Acta 14, 189-194.

Krom, M.D., Emeis, K.C., Van Cappellen, P., 2010. Why is the Eastern Mediterranean phosphorus limited?. Progr. Oceanogr. 85 (3–4), 236-244. https://doi.org/10.1016/j.pocean.2010.03.003.

Krom, M.D., Groom, S., Zohary, T., 2003. The Eastern Mediterranean. In: The Biogeochemistry of Marine Systems, Blackwell Publishing, Oxford, 91-122.

Krom, M.D., Shi, Z., Stockdale, A., Berman-Frank, I., Giannakourou, A., Herut, B., Lagaria, A., Papageorgiou, N., Pitta, P,, Psarra, S., Rahav, E., Scoullos, M., Stathopoulou, E., Tsiola, A., Tsagaraki, T.M., 2016. Response of the Eastern Mediterranean Microbial Ecosystem to Dust and Dust Affected by Acid Processing in the Atmosphere . Front. Mar. Sci. 3, 133.

Kujawinski, E., 2011. The Impact of Microbial Metabolism on Marine Dissolved Organic Matter. An. Rev. Mar. Sci. 3, 567–99. http://dx.doi.org/10.1146/annurev-marine-120308-081003.

Kwon, E.Y., Kim, G., Primeau, F., Moore, W.S., Cho, H.M., DeVries, T., Sarmiento, J.L., Charette, M.A., Cho, Y.K., 2014. Global estimate of submarine groundwater discharge based on an observationally constrained radium isotope model. Geophys. Res. Lett. 41, 8438– 8444, http://dx.doi.org/10.1002/2014GL061574.

Kwon, S.W., Kim, B.Y., Weon, H.Y., Baek, Y.K., Koo, B.S., Go, S.J., 2006. *Idiomarina homiensis* sp. nov., isolated from seashore sand in Korea. Int. J. Syst. Evol. Microbiol. 56, 2229–2233. http://dx.doi.org/10.1099/ijs.0.64283-0.

Lascaratos, A., 1993. Estimation of deep and intermediate water mass formation rates in the Mediterranean Sea. Deep-Sea Res. II 40, 1327-1332. http://dx.doi.org/10.1016/0967-0645(93)90072-U.

Lascaratos, A., Nittis, K., 1998. A high resolution 3-D numerical study of intermediate water formation in the Levantine Sea. J. Geophys. Res. 103, 18497-18511. https://doi.org/10.1029/98JC01196.

Lascaratos, A., Roether, W., Nittis, K., Klein, B., 1999. Recent changes in deep water formation and spreading in the eastern Mediterranean Sea: a review. Progr. Oceanogr. 44 (1–3), 5-36. https://doi.org/10.1016/S0079-6611(99)00019-1.

Lazzari, P., Solidoro, C., Salon, S., Bolzon, G., 2016. Spatial variability of phosphate and nitrate in the Mediterranean Sea: A modeling approach. Deep Sea Res. Part I:

Oceanogr. Res. Papers 108, 39-52. https://doi.org/10.1016/j.dsr.2015.12.006.

Leaman, K.D., Schott, F.A., 1991. Hydrographic Structure of the Convection Regime in the Gulf of Lions: Winter 1987. Journal of Physical Oceanography 21 (4), 575-598.

Lekunberri, I., Gasol, J.M., Acinas, S.G., Gómez-Consarnau, L., Crespo, B.G., Casamayor, E.O., Massana, R., Pedrós-Alió, C., Pinhassi, J., 2014. The phylogenetic and ecological context of cultured and whole genome-sequenced planktonic bacteria from the coastal NW Mediterranean Sea. Syst. Appl. Microbiol. 37, 216–228. http://dx.doi.org/10.1016/j.syapm.2013.11.005.

Lemaitre, N., Planquette, H., Planchon, F., Sarthou, G., Jacquet, S., García-Ibáñez, M. I., Gourain, A., Cheize, M., Monin, L., André, L., Laha, P., Terryn, H., Dehairs, F., 2018. Particulate barium tracing of significant mesopelagic carbon remineralisation in the North Atlantic. Biogeosci. 15, 2289–2307. https://doi.org/10.5194/bg-15-2289-2018.

Leoni, L., Sartori, F., Damiani, V., Ferreti, O., Viel, M., 1991. Trace element distributions in surficial sediments of the northern Tyrrhenian Sea: Contribution to heavy-metal pollution assessment. Environ. Geol. Water Sci. 17, 103–116. https://doi.org/10.1007/BF01701566.

Le Roy, E., Sanial, V., Charette, M.A., Van Beek, P., Lacan, F., Jacquet, S.H.M., Henderson, P.B., Souhaut, M., García-Ibáñez, M.I., Jeandel, C., Pérez, F.F., Sarthou, G., 2018. The ^{226}Ra–Ba relationship in the North Atlantic during GEOTRACES-GA01. Biogeosci. 15, 3027–3048. https://doi.org/10.5194/bg-15-3027-2018.

Levin, L., 1994. Paleoecology and ecology of xenophyophores. Palaios 9, 32–41. https://doi.org/10.2307/3515076.

Li, Y.H., Chan, L.H., 1979. Desorption of Ba and226Ra from river-borne sediments in the Hudson estuary. Earth Planet. Sci. Lett. 43 (3), 343-350. https://doi.org/10.1016/0012-821X(79)90089-X.

Li, P., Tanhua, T., 2020. Recent changes in deep ventilation of the Mediterranean Sea, evidence from long-term transient tracer observations. Front. Mar. Sci. 7, 594. doi: 10.3389/fmars.2020.00594.

Liguori, B., Almeida M.G., De Rezende, C., 2016. Barium and its Importance as an Indicator of (Paleo)Productivity. An. Acad. Bras. Cenc. 88 (4), 2093–2103. https://doi.org/10.1590/0001-3765201620140592.

Lindquist, N., Fenical,W., 1991. New tambjamine class alkaloids from the marine ascidian *Atapozoa* sp. and its nudibranch predators—origins of the tambjamines in *Atapozoa*. Experientia 47, 504–508.

Lionello, P., 2012. The Climate of the Mediterranean Region: From the Past to the Future. Elsevier, 592. https://doi.org/10.1016/C2011-0-06210-5.

Llovel, W., Meyssignac, B., Cazenave, A., 2011. Steric sea level variations over 2004-2010 as a function of region and depth: Inference on the mass component variability in the North Atlantic Ocean. Geophys. Res. Lett. 38, https://doi.org/10.1029/2011gl047411.

López-López, A., Bartual, S.G., Stal, L., Onyshchenko, O., Rodríguez-Valera, F., 2005. Genetic analysis of housekeeping genes reveals a deep-sea ecotype of *Alteromonas macleodii* in the Mediterranean Sea. Environ. Microbiol. 7 (5), 649–659. http://dx.doi.org/10.1111/j.1462-2920.2005.00733.x.

Lourens, L.J., Hilgen, F.J., Gudjonsson, L., Zachariasse, W.J., 1992. Late Pliocene to early Pleistocene astronomically forced sea surface productivity and temperature variations in the Mediterranean. Mar. Micropal.19 (1–2), 49–78. https://doi.org/10.1016/0377-8398(92)90021-B.

Lourens, L.J., Hilgen, F.J., Zachariasse, W.J., Van Hoof, A.A.M., Antonarakou, A., Vergnaud-Grazzini, C., 1996. Evaluation of the Plio-Pleistocene astronomical timescale. Paleocean. 11, 391–413. http://dx.doi.org/10.1029/96PA01125.

Lourens, L., Wehausen, R., Brumsack, H., 2001. Geological constraints on tidal dissipation and dynamical ellipticity of the Earth over the past three million years. Nature 409, 1029–1033. https://doi.org/10.1038/35059062.

Lowenstamm, H.A., 1981. Minerals formed by microorganisms. Science 211, 1126-1131.

Ludwig, W., Dumont, E., Meybeck, M., Heussner, S., 2009. River discharges of water and nutrients to the Mediterranean and Black Sea: Major drivers for ecosystem changes during past and future decades?. Progr. Oceanogr. 80 (3–4), 199-217. https://doi.org/10.1016/j.pocean.2009.02.001.

Luijendijk, E., Gleeson, T., Moosdorf, N., 2020. Fresh groundwater discharge insignificant for the world’s oceans but important for coastal ecosystems. Nat. Commun. 11 (1–12). https://doi.org/10.1038/s41467-020-15064-8.

Luther, G.W., Catalano, G-., De Lange, G.J., Woittiez, J.R.W., 1990. Reduced sulfur in the hypersaline anoxic basins of the Mediterranean Sea. Mar. Chem. 31 (1–3), 137-152. https://doi.org/10.1016/0304-4203(90)90035-B.

Luyten, J.R., Pedlosky, J., Stommel, H., 1983. The ventilated thermocline. J. Phys. Oceanogr. 13, 292-309. https://doi.org/10.1175/1520-0485(1983)013<0292:TVT>2.0.CO;2.

Macías, D., Garcia-Gorriz, E., Stips, A., 2015. Productivity changes in the Mediterranean Sea for the twenty-first century in response to changes in the regional atmospheric forcing. Front. Mar. Sci. 2. https://doi.org/10.3389/fmars.2015.00079.

Mackey, K., Buck, K., Casey, J., Cid, A., Lomas, M., Sohrin, Y., Paytan, A., 2012. Phytoplankton responses to atmospheric metal deposition in the coastal and open-ocean Sargasso Sea. Frontiers in Microbiology 3, 359. https://doi.org/10.3389/fmicb.2012.00359.

Mahowald, N.M., Baker, A.R., Bergametti, G., Brooks, N., Duce, R.A., Jickells, T.D., Kubilay, N., Prospero, J.M., Tegen, I., 2005. Atmospheric global dust cycle and iron inputs to the ocean. Global Biogeochem. Cycles 19, GB4025. https://doi.org/10.1029/2004GB002402.

Maier, C., Weinbauer, M., Gattuso, J.P., 2019. Fate of Mediterranean Scleractinian Cold-Water Corals as a Result of Global Climate Change. A Synthesis.

Mann S., 1983. Mineralization in biological systems. In: Inorganic Elements in Biochemistry. Structure and Bonding 54. Springer, Berlin, Heidelberg. https://doi.org/10.1007/BFb0111320.

Margat, J., 1992. L'eau des Méditerranéens: situation et perspective. Tech. Rep. 6, U. N. Environ. Programme Mediter. Action Plan, Athens, Greece.

Maring, H.B., Duce, R.A., 1987. The impact of atmospheric aerosols on trace metal chemistry in open ocean surface sea- water, 1. Aluminitun. Earth Planet. Sci. Lett. 84, 381-392. https://doi.org/10.1016/0012-821X(87)90003-3.

Marino, G., Rohling, E.J., Rijpstra, W.I.C., Sangiorgi, F., Schouten, S., Damsté, J.S.S., 2007. Aegean Sea as driver of hydrographic and ecological changes in the eastern Mediterranean. Geol. 35 (8), 675–678. https://doi.org/10.1130/G23831A.1.

Mariotti, A., Struglia, M.V., Zeng, N., Lau, K.M., 2002. The Hydrological Cycle in the Mediterranean Region and Implications for the Water Budget of the Mediterranean Sea. Journal of Climate, 15 (13), 1674-1690.

Mariotti, A., Pan, Y., Zeng, N., Alessandri, A., 2015. Long-term climate change in the Mediterranean region in the midst of decadal variability. Clim. Dyn. 44 (5-6), 1437-1456. https://doi.org/10.1007/s00382-015-2487-3.

Mariotti, A., Zeng, N., Yoon, J.H., Artale, V., Navarra, A., Alpert, P., Li, L.Z.X., 2008. Mediterranean water cycle changes: Transition to drier 21st century conditions in observations and CMIP3 simulations. Environ. Res. Lett. 3 , 044001. https://doi.org/10.1088/1748-9326/3/4/044001.

Martin, J.M., Elbaz-Poulichet, F., Guieu, C., Loÿe-Pilot, M.D., Han, G., 1989. River versus atmospheric input of material to the mediterranean sea: an overview. Mar. Chem. 28 (1–3), 159-182, https://doi.org/10.1016/0304-4203(89)90193-X.

Martínez-Cánovas, M.J., Béjar, V., Martínez-Checa, F., Páez, R., Quesada, E., 2004. *Idiomarina fontislapidosi* sp. nov. and *Idiomarina ramblicola* sp. nov., isolated from inland hypersaline habitats in Spain. Int. J. Syst. Evol. Microbiol. 54, 1793–1797. http://dx.doi.org/10.1099/ijs.0.63172-0.

Martín-Ramos, J.D., 2004. XPowder, a software package for powder X-ray diffraction analysis. Legal Deposit GR 1001/04 84-609-1497-6 (http://www.xpowder.com (accessed June 11, 2014).

Martínez-Ruiz, F., Jroundi, F., Paytan, A., Guerra-Tschuschke, I., Abad, M., González-Muñoz, M.T., 2018. Barium bioaccumulation by bacterial biofilms and implications for Ba cycling and use of Ba proxies. Nat .Commun. 9 (1619). http://dx.doi.org/10.1038/s41467-018-04069-z.

Martínez-Ruiz, F., Kastner, M., Gallego-Torres, D., Rodrigo-Gámiz, M., Nieto-Moreno, V., Ortega-Huertas, M., 2015. Paleoclimate and paleoceanography over the past 20,000 yr in the Mediterranean Sea Basins as indicated by sediment elemental proxies. Q. Sci. Rev. 107, 25–46. https://doi.org/10.1016/j.quascirev.2014.09.018.

Martínez-Ruiz, F., Kastner, M., Paytan, A., Ortega-Huertas, M., Bernasconi, S. M., 2000. Geochemical evidence for enhanced productivity during S1 sapropel deposition in the eastern Mediterranean. Paleoceanog. 15 (2), 200– 209. http://dx.doi.org/10.1029/1999PA000419.

Martínez-Ruiz, F., Paytan, A., González-Donoso, J.M., Linares, D., Bernasconi, S.M., Jiménez-Espejo, F.J., Kastner, M., 2003. Geochemical and mineralogical evidence for sapropel deposition in eastern and western Mediterranean basins: paleoclimate implications and paleoceanographic reconstruction. EGS - AGU - EUG Joint Assembly, Abstracts from the meeting held in Nice, France, 6 - 11 April 2003, abstract id. 11786.

Martínez-Ruiz, F., Paytan, A., González-Muñoz, M.T., Jroundi, F., Abad, M.M., Lam, P.J., Bishop, J.K.B., Horner, T.J., Morton, P.L., Kastner, M., 2019. Barite formation in the ocean: Origin of amorphous and crystalline precipitates. Chem. Geol. 511, 441–451.

https://doi.org/10.1016/j.chemgeo.2018.09.011.

Martínez-Ruiz, F., Paytan, A., González-Muñoz, M.T., Jroundi, F., Abad, M.M., Lam, P.J., Horner, T.J., Kastner, M., 2020. Barite precipitation on suspended organic matter in the mesopelagic zone. Frontiers in Earth Sciences 8, 567714. https://doi.org/10.3389/feart.2020.567714.

Marty, J.C., Chiavérini, J., 2002. Seasonal and interannual variations in phytoplankton production at DYFAMED time-series station, northwestern Mediterranean Sea. Deep-Sea Rese. Part II 49, 2017-2030. https://doi.org/110.1016/S0967-0645(02)00025-5.

Marullo, S., Santoleri, R., Malanotte-Rizzoli, P., Bergamasco, A., 1999. The sea surface temperature field in the Eastern Mediterranean from advanced very high resolution radiometer (AVHRR) data: Part I. Seasonal variability. J. Mar. Syst. 20 (1–4), 63-81. https://doi.org/10.1016/S0924-7963(98)00071-2.

Masciopinto, C., Liso, I.S., 2016. Assessment of the impact of sea-level rise due to climate change on coastal groundwater discharge. STOTEM 569–570, 672-680. https://doi.org/10.1016/j.scitotenv.2016.06.183.

Masqué, P., Fabres, J., Canals, M., Sanchez-Cabeza, J.A, Sanchez-Vidal, A., Cacho, I., Calafat, A.M., Bruach, J.M., 2003. Accumulation rates of major constituents of hemipelagic sediments in the deep Alboran Sea: a centennial perspective of sedimentary dynamics. Mar. Geol. 193 (3–4), 207–233. https://doi.org/10.1016/S0025-3227(02)00593-5.

Mata, J.A., 2006. Caracterización de los exopolisacáridos producidos por microorganismos halófilos pertenecientes a los géneros *Halomonas, Alteromonas, Idiomarina, Palleronia* y *Salipiger*. Tesis doctoral, Universidad de Granada. http://hdl.handle.net/10481/973.

Mayer, L.M., 1995. Sedimentary organic matter preservation: an assessment and speculative synthesis—a comment. Mar. Chem. 49 (2–3), 123-126. https://doi.org/10.1016/0304-4203(95)00011-F.

Mayfield, K.K., Eisenhauer, A., Santiago Ramos, D.P., Higgins, J.A., Horner, T.J., Auro, M., Magna, T., Moodsdorf, N., Charette, M.A., Gonneea, M.E., Brady, C.E., Komar, N., Peucker-Ehrenbrink, B., Paytan, A., 2021. Groundwater discharge impacts marine isotope budgets of Li, Mg, Ca, Sr, and Ba. Nat. Commun. 12, 148. https://doi.org/10.1038/s41467-020-20248-3.

McConnell, D., 1973. Apatite: Its crystal chemistry, mineralogy, utilization, and geologic and biologic occurrences. Appl. Min. 5. Vienna and New York (Springer-Verlag). Mineralogical Magazine*,* 39 (305), 617–618. https://doi.org/10.1180/minmag.1974.039.305.22.

McCoy, F.W., 1974. Late Quaternary Sedimentation in the Eastern Mediterranean Sea. Doctoral dissertation, Harvard University, Cambridge, Mass.

McManus, J., Berelson, W.M., Hammond, D.E., Klinkhammer, G.P. , 1999. Barium Cycling in the North Pacific: Implications for the Utility of Ba as a Paleoproductivity and Paleoalkalinity Proxy. Paleoceanogr. 14(1), 53– 61. https://doi.org/10.1029/1998PA900007.

McManus, J., Berelson, W.M., Klinkhammer, G.P., Johnson, K.S., Coale, K.H., Anderson, R.F., Kumar, N., Burdige, D.J., Hammond, E.E., Brumsack, H.J., McCorkle, D.C., Rushdi, A.,

1998. Geochemistry of barium in marine sediments: implications for its use as a paleoproxy. Geochim. Cosmoch. Acta 62 (21–22), 3453-3473. https://doi.org/10.1016/S0016-7037(98)00248-8.

McQuay, E.L., Torres, M.E., Collier, R.W., Huh, C.A., McManus, J., 2008. Contribution of cold seep barite to the barium geochemical budget of a marginal basin. Deep Sea Res. Part I: Oceanogr. Res. Papers 55 (6), 801-811. https://doi.org/10.1016/j.dsr.2008.03.001.

Measures, C.I., Brown, M.T., Vink, S., 2005. Dust deposition to the surface waters of the western and central North Pacific inferred from surface water dissolved aluminum concentrations. Geochem. Geophys. Geosyst. 6, Q09M03, https://doi.org/10.1029/2005GC000922.

Measures, C.I., Landing, W.M., Brown, M.T., Buck, C.S., 2008, High-resolution Al and Fe data from the Atlantic Ocean CLIVAR-CO2 Repeat Hydrography A16N transect: extensive linkages between atmospheric dust and upper ocean geochemistry. Glob. Biogeochem. Cycles 22, GB1005. https://doi.org/10.1029/2007GB003042.

Measures, C.A., Sato, T., Vink, S., Howell, S., Li, Y.H., 2010. The fractionalsolubility of aluminium from mineral aerosols collected in Hawaii and implica-tions for atmospheric deposition of biogeochemically important trace elements. Mar. Chem. 120, 144–153. https://doi.org/10.1016/j.marchem.2009.01.014.

Megalovasilis, P., 2020. Hydrothermal fluid particle geochemistry of submarine vents in Kos Island, Aegean Sea East Mediterranean. Geochem. Internat. 58, 574-597.

Mejías, M., Ballesteros, B.J., Antón-Pacheco, C., Domínguez, J.A., Garcia-Orellana, J., Garcia-Solsona, E., Masqué, P., 2012. Methodological study of submarine groundwater discharge from a karstic aquifer in the Western Mediterranean Sea. J. Hydrol. 464–465, 27-40. https://doi.org/10.1016/j.jhydrol.2012.06.020.

Menzel, D., Van Bergen, P.F., Schouten, S., Sinninghe Damsté, J.S., 2003. Reconstruction of changes in export productivity during Pliocene sapropel deposition: a biomarker approach. Palaeogeogr. Palaeoclimatol. Palaeoecol. 190, 273-287. https://doi.org/10.1016/S0031-0182(02)00610-7.

Mercone, D., Thomson, J., Abu-Zied, R.H., Croudace, I.W., Rohling, E.J., 2001. High-resolution geochemical and micropalaeontological profiling of the most recent eastern Mediterranean sapropel. Marine Geology 177 (1–2), 25-44. https://doi.org/10.1016/S0025-3227(01)00122-0.

Merroun, M.L., Chekroun, K.B., Arias, J.M., González-Muñoz, M.T., 2003. Lanthanum fixation by *Myxococcus xanthus*: Cellular location and extracellular polysaccharide observation. Chemosph. 52 (1), 113–120. https://doi.org/10.1016/S0045-6535(03)00220-0.

Merroun, M.L., Nedelkova, M., Rossberg, A., Hennig, C., Selenska-Pobell, S., 2006. Interaction mechanisms of bacterial strains isolated from extreme habitats with uranium. Radiochim. Acta 94 (9–11), 723–729. https://doi.org/10.1524/ract.2006.94.9-11.723.

Merroun, M.L., Selenska-Pobell, S., 2001. Interactions of three eco-types of *Acidithiobacillus ferrooxidans* with U(VI). BioMetals 14 (2), 171–179. https://doi.org/10.1023/A:1016658209397.

Merroun, M.L., Selenska-Pobell, S., 2008. Bacterial interactions with uranium: an environmental perspective. J. Contam. Hydrol. 102(3–4), 285–95. https://doi.org/10.1016/j.jconhyd.2008.09.019.

Meyer, K.M., Ridgwell, A., Payne, J.L., 2016. The influence of the biological pump on ocean chemistry: implications for long-term trends in marine redox chemistry, the global carbon cycle, and marine animal ecosystems. Geobiol.y 14, 207–219. http://dx.doi.org/10.1111/gbi.12176.

Michard, G., Church, T.M., Bernat, M., 1974. The pore water chemistry of recent sediments in the western Mediterranean basin. J. Geophys. Res. 79 (6), 817– 824. http://dx.doi.org/10.1029/JC079i006p00817.

Middleburg, J.J., 2019. The Return from Organic to Inorganic Carbon. In: Mar. Carbon Biogeochem.. Springer Briefs in Earth Syst. Sci. Springer, Cham. https://doi.org/10.1007/978-3-030-10822-9_3.

Migon, C., 2005. Trace Metals in the Mediterranean Sea. In: Saliot A. (eds) The Mediterranean Sea. Handbook of Environmental Chemistry, vol 5K. Springer, Berlin, Heidelberg. https://doi.org/10.1007/b107146.

Mikhailov, V.V., Romanenko, L.A., Ivanova, E.P., 2006. The genus *Alteromonas* and related proteobacteria. In: The Prokaryotes: A Handbook on the Biology of Bacteria vol. 6. Springer, New York, 597–645. http://dx.doi.org/10.1007/0-387-30746-x_20.

Mills, M., Ridame, C., Davey, M., LaRoche, J., Geider, R.J., 2004. Iron and phosphorus co-limit nitrogen fixation in the Eastern Tropical North Atlantic. Nature 429, 292–294. http://dx.doi.org/10.1038/nature02550.

Millot C., 1999. Circulation in the Western Mediterranean Sea. J. Mar. Syst. 20 (1–4), 423-442. https://doi.org/10.1016/S0924-7963(98)00078-5.

Millot, C., 2013. Levantine Intermediate Water characteristics: an astounding general misunderstanding!. Sci. Mar. 77(2), 217-232.

Millot C., Taupier-Letage I., 2005. Circulation in the Mediterranean Sea. In: The Mediterranean Sea. In: Handbook of Environmental Chemistry, vol. 5. Springer, Berlin, Heidelberg. https://doi.org/10.1007/b107143.

Milly, P.C.D., Dunne, K.A., Vecchia, A.V., 2005. Global pattern of trends in streamflow and water availability in a changing climate. Nature 438, 347–350.

Möbius, J., Lahajnar, N., Emeis, K.C., 2015. Chemical composition of Holocene sediment core M51/3_562 from the Eastern Mediterranean Sea. Pangaea. https://doi.org/10.1594/PANGAEA.843932. In supplement to: Möbius, J. et al., 2010. Diagenetic control of nitrogen isotope ratios in Holocene sapropels and recent sediments from the Eastern Mediterranean Sea. Biogeosci. 7, 3901-3914. https://doi.org/10.5194/bg-7-3901-2010.

Moller, T., Schulz, H., Hamann, Y., Dellwig, O., Kucera, M., 2012. Sedimentology and geochemistry of an exceptionally preserved last interglacial sapropel S5 in the Levantine Basin (Mediterranean Sea). Mar. Geol. 291–294, 34–48. https://doi.org/10.1016/j.margeo.2011.10.011.

Monnin, C., Cividini, D., 2006. The saturation state of the world's ocean with respect to (Ba,Sr)SO4 solid solutions. Geochim. Cosmochim. Acta, 70, 3290-3298. https://doi.org/10.1016/J.GCA.2006.04.002 ff.hal-00662992.

Monnin, C., Jeandel, C., Cattaldo, T., Dehairs, F., 1999. The marine barite saturation state of the world's oceans. Mar. Chem. 65, 253–261. http://dx.doi.org/10.1016/S0304-4203(99)00016-X.

Monnin, C., Wheat, C.G., Dupre, B., Elderfield, H., Mottl, M.M., 2001. Barium geochemistry in sediment pore waters and formation waters of the oceanic crust on the eastern flank of the Juan de Fuca Ridge (ODP Leg 168), Geochem. Geophys. Geosyst. 2, 1008. https://doi.org/:10.1029/2000GC000073.

Moore, W., 1996. Large groundwater inputs to coastal waters revealed by ^{226}Ra enrichments. Nature 380, 612–614. https://doi.org/10.1038/380612a0.

Moore, W., 1997. High fluxes of radium and barium from the mouth of the Ganges-Brahmaputra River during low river discharge suggest a large groundwater source. Earth Planet. Sci. Lett. 150 (1–2), 141-150. https://doi.org/10.1016/S0012-821X(97)00083-6.

Moore, W., 2010. The effect of submarine groundwater discharge on the ocean. Ann. Rev. Mar. Sci. 2, 59-88. https://doi.org/10.1146/annurev-marine-120308-081019.

Moore, W., Falkner, K.K., 1999. Cycling of radium and barium in the Black Sea. J. Environ. Radioact. 43 (2), 247-254. https://doi.org/10.1016/S0265-931X(98)00095-2.

Moore, W., Sarmiento, J. Key, R., 2008. Submarine groundwater discharge revealed by ^{228}Ra distribution in the upper Atlantic Ocean. Nature Geosci. 1, 309–311. https://doi.org/10.1038/ngeo183.

Morcillo, F., 2010. *Idiomarina loihiensis MAH1*: estudio de su capacidad carbonatogénica y de fijación de lantánidos y actínidos. Tesis doctoral, Universidad de Granada. http://hdl.handle.net/10481/5640.

Moreno, T., Querol, X., Castillo, S., Alastuey, A., Cuevas, E., Herrmann, L.,25Mounkaila, M., Elvira, J., Gibbons, W., 2006. Geochemical variations in aeolian min-eral particles from the Sahara-Sahel Dust Corridor. Chemosph. 65, 261–270, https://doi.org/10.1016/j.chemosphere.2006.02.052.

Morigi, C., 2009. Benthic environmental changes in the Eastern Mediterranean Seaduring sapropel S5 deposition. Palaeogeogr. Palaeoclimatol. Palaeoecol. 273, 258–271. https://doi.org/10.1016/j.palaeo.2008.10.010.

Morillo Perez, J.A., Garcia-Ribera, R., Quesada, T., Aguilera, M., Ramos-Cormenzana, A., Monteoliva-Sanchez, M., 2008. Biosorption of heavy metals by the exopolysaccharide produced by *Paenibacillus jamilae.* World J. Microbiol. Biotech. 24,. 2699–2704. http://dx.doi.org/10.1007/s11274-008-9800-9.

Moulin, C., Lambert, C., Dulac, F. Dayan, U., 1997. Control of atmospheric export of dust from North Africa by the North Atlantic Oscillation. Nature 387, 691–694. https://doi.org/10.1038/42679.

Moutin, T., Raimbault, P,m 2002. Primary production, carbon export and nutrients availability in western and eastern Mediterranean Sea in early summer 1996 (MINOS cruise). J. Mar. Syst. 33, 273-288. http://dx.doi.org/10.1016/S0924-7963(02)00062-3.

Müller, P.J., Suess, E., 1979. Productivity, sedimentation rate, and sedimentary organic matter in the oceans—I. Organic carbon preservation. Deep Sea Res. Part A. Oceanogr. Res. Papers 26 (12), 1347-1362. https://doi.org/10.1016/0198-0149(79)90003-7.

Mullins, T.D., Britschgi, T.B., Krest, R.L., Giovannoni, S.J., 1995. Genetic comparisons reveal the same unknown bacterial lineages in Atlantic and Pacific bacterioplankton communities. Limnol. Oceanogr. 40, 148–158. http://dx.doi.org/10.4319/lo.1995.40.1.0148.

Murat, A., 1991. Enregistrement sédimentaire des paléoenvironments quaternaires en Méditerranée Orientale. Thèse de doctorat, Université de Perpignan, 280 pp.

Murat, A., 1999. Pliocene–Pleistocene occurrence of sapropels in the Western Mediterranean Sea and their relation to Eastern Mediterranean sapropels. In: Proceedings of the Ocean Drilling Program, Scientific Results 8 vol. 161. Ocean Drilling Program, College Station, TX, 519 – 527.

Murat, A., Got, H., 2000. Organic carbon variations of the eastern Mediterranean Holocene sapropel: a key for understanding formation processes. Palaeogeogr. Palaeoclimatol. Palaeoecol. 158, 241–257. https://doi.org/10.1016/S0031-0182(00)00052-3.

Murphy, J., Riley, J.P., 1962. A modified single solution method for the determination of phosphate in natural waters. Anal. Chim. Acta 27, 31–36. https://doi.org/10.1016/S0003-2670(00)88444-5.

Murray, J.W., Top, Z., Özsoy, E., 1991. Hydrographic properties and ventilation of the Black Sea. Deep Sea Res.h Part A. Oceanogr. Res. Papers 38 (2), S663-S689. https://doi.org/10.1016/S0198-0149(10)80003-2.

Naehr, T.H., Stakes, D.S., Moore, W.S., 2000. Mass wasting, ephemeral fluid flow, and barite deposition on the California continental margin. Geology 28 (4), 315–318. https://doi.org/10.1130/0091-7613(2000)28<315:MWEFFA>2.0.CO;2.

Nagata, T., Fukuda, H., Fukuda, R., Koike, I., 2000. Bacterioplankton distribution and production in deep Pacific waters: Large-scale geographic variations and possible coupling with sinking particle fluxes. Limnology and Oceanography 2. https://doi.org/10.4319/lo.2000.45.2.0426.

Neff, J., 2002. Bioaccumulation in Marine Organisms: Effect of Contaminants from Oil Well Produced Water. In: Bioaccumulationin marine organisms. Elsevier, Oxford, 241-313.

Neff, J.M., Sauer, T.C., 1995. Barium in Produced Water: Fate and Effects in the Marine Environment. API Publication 4633. American Petroleum Institute, Washington, DC.

Neves, M.C., Jerez, S., Trigo, R.M., 2019. The response of piezometric levels in Portugal to NAO, EA, and SCAND climate patterns. J. Hydrol., 568, 1105–1117. https://doi.org/10.1016/j.jhydrol.2018.11.054.

Newsome, L., Morris, K., Lloyd, J.R., 2014. The biogeochemistry and bioremediation of uranium and other priority radionuclides. Chem. Geol. 363, 164–184. https://doi.org/10.1016/j.chemgeo.2013.10.034.

Nicholson, W.L., 2002. Roles of *Bacillus* spores in the environment. Cell. Mol. Life Sci. 59, 410–416. http://dx.doi.org/10.1007/s00018-002-8433-7.

Nieto-Moreno, V., Martínez-Ruiz, F., Willmott Puig, V., Garcia-Orellana, J., Masqué, P., Sinninghe-Damste, J., 2012. Climate conditions in the westernmost Mediterranean over the last two millennia: An integrated biomarker approach. Org. Geochem. 55, 1. https://doi.org/10.1016/j.orggeochem.2012.11.001.

Nijenhuis, I.A., De Lange, G.J., 2000. Geochemical constraints on Pliocene sapropel formation in the eastern Mediterranean. Mar. Geol. 163 (1–4), 41-63. https://doi.org/10.1016/S0025-3227(99)00093-6.

Oelkers, E.H., Gislason, S.R., Eiriksdottir, E.S., Jones, M., Pearce, C.R., Jeandel, C., 2011. The role of riverine particulate material on the global cycles of the elements. Applied Geochem. 26, S365-S369. https://doi.org/10.1016/j.apgeochem.2011.03.062.

Ollivier, P., Claude, C., Radakovitch, O., Hamelin, B., 2008. TIMS measurements of 226Ra and 228Ra in the Gulf of Lion, an attempt to quantify submarine groundwater discharge. Mar. Chem. 109 (3–4), 337-354. https://doi.org/10.1016/j.marchem.2007.08.006.

Ovchinnikov, I.M., 1974. On the water balance of the Mediterranean sea. Oceanol. 14, 198-202.

Ovchinnikov, I.M., 1983. The renewal of the principal water masses of the Mediterranean. Oceanol. 23, 719-721.

Ovchinnikov, I.M., 1984. The formation of the intermediate waters in the Mediterranean Sea. Okeanologiya 26 (2), 217-225.

Owens, W.B., Hogg, N.G., 1980. Oceanic observations of stratified Taylor columns near a bump. Deep Sea Res. A 27, 1029–1045. https://doi.org/10.1016/0198-0149(80)90063-1.

Ozsoy, E., Unluata, U., 1997. Oceanography of the Black Sea: a review of some resent results. Earth Sci. Rev. 42 (4), 231-272. https://doi.org/10.1016/S0012-8252(97)81859-4.

Packard, T.T., Minas, H.J., Coste, B., Martinez, R., Bonin, M.C., Gostan, J., Garfield, P., Christensen, J., Dortch, Q., Minas, M., Copin-Montegut, G., Copin-Montegut, C., 1988. Formation of the Alboran oxygen minimum zone. Deep Sea Res. Part A. Oceanogr. Res. Papers 35 (7), 1111-1118. https://doi.org/10.1016/0198-0149(88)90003-9.

Passier, H., Bosch, HJ., Nijenhuis, I., Lourens, L.J., Böttcher, M.E., Leenders, A., Sinninghe Damsté, J.S., De Lange, G.J., Leeuw, J.W., 1999. Sulphidic Mediterranean surface waters during Pliocene sapropel formation. Nature 397, 146–149. https://doi.org/10.1038/16441.

Passow, U., Carlson , C.A., 2012. The biological pump in a high CO_2 world. Mar. Ecol. Prog. Ser. 470, 249–271. https://doi.org/10.3354/meps09985.

Paytan, A., Griffith, E.M., 2007. Marine barite: recorder of variations in ocean export productivity. Deep-Sea Res. II 54 (5–7), 687–705. http://dx.doi.org/10.1016/j.dsr2.2007.01.007.

Paytan, A., Kastner, M., Chavez, F.P., 1996. Glacial to interglacial fluctuation in productivity in the Equatorial Pacific as indicated by marine bacteria. Science, 1355–1357. http://dx.doi.org/10.1126/science.274.5291.1355.

Paytan, A., Martinez-Ruiz, F., Eagle, M., Ivy, A., Wankel, S.D., 2004. Using sulfur isotopes to elucidate the origin of barite associated with high organic matter accumulation events in marine sediments. Sulfur Biogeochem. - Past and Present, Jan P. Amend, Katrina J. Edwards, Timothy W. Lyons.

Paytan, A., Mearon, S., Cobb, K., Kastner, M., 2002. Origin of marine barite deposits: Sr and S isotope characterization. Geol. 30 (8), 747–750. https://doi.org/10.1130/0091-7613(2002)030<0747:OOMBDS>2.0.CO;2.

Pedersen, T.F., Calvert, S.E., 1990. Anoxia vs. Productivity: What Controls the Formation of Organic-Carbon-Rich Sediments and Sedimentary Rocks?. AAPG Bulletin, 74 (4), 454–466. https://doi.org/10.1306/0C9B232B-1710-11D7-8645000102C1865D.

Peixoto, J.P., De Almeida, M., Rosen, R.D., Salstein, D.A., 1982. Atmospheric moisture transport and the water balance of the Mediterranean Sea. Water Resour. Res. 18 (1), 83–90, https://doi.org/10.1029/WR018i001p00083.

Penaud, A., Eynaud, F., Sánchez-Goñi, M., Malaizé, B., Turon, J.L., Rossignol, L., 2011. Contrasting sea-surface responses between the western Mediterranean Sea and eastern subtropical latitudes of the North Atlantic during abrupt climatic events of MIS 3. Mar. Micropaleont. 80 (1–2), 1-17. https://doi.org/10.1016/j.marmicro.2011.03.002.

Pérez-Asensio, J.N., Frigola, J., Pena, L.D., Sierro, F.J., Reguera, M.I., Rodríguez-Tovar, F.J., Dorador, J., Asioli, A., Kuhlmann, J., Huhn, K., Cacho, I., 2020. Changes in western Mediterranean thermohaline circulation in association with a deglacial Organic Rich Layer formation in the Alboran Sea. Quat. Sci. Rev. 228. https://doi.org/10.1016/j.quascirev.2019.106075.

Pérez, F.F., Mouriño, C., Fraga, F., Ríos, A.F., 1993. Displacement of water masses and remineralization rates off the Iberian peninsula by nutrient anomalies. J. Mar. Res. 51, 869-892. https://doi.org/10.1357/0022240933223891.

Perissoratis, C., Piper, D.J.W., 1992. Age, regional variation, and shallowest occurrence of S1 sapropel in the northern Aegean Sea. Geo-Mar. Lett. 12, 49–53. https://doi.org/10.1007/BF02092108.

Pfeifer, K., Kasten, S., Hensen, C., Schulz, H., 2001. Reconstruction of primary productivity from the barium contents in surface sediments of the South Atlantic Ocean. Mar. Geol. 177 (1–2), 13–24. https://doi.org/10.1016/S0025-3227(01)00121-9.

Picotti, V., Negri, A., Capaccioni, B., 2014. The Geological Origins and Paleoceanographic History of the Mediterranean Region: The Mediterranean Sea. Springer, Dordrecht. https://doi.org/10.1007/978-94-007-6704-1_1.

Piela, C., Lyle, M., Marcantonio, F., Baldauf, J., and Olivarez Lyle, A., 2012. Biogenic sedimentation in the equatorial Pacific: Carbon cycling and paleoproduction, 12–24 Ma. Paleocean. 27, PA2204, https://doi.org/10.1029/2011PA002236.

Pinardi, N., Arneri, E., Crise, A., Ravaioli, M., Zavatarelli, M., 2006. The physical, sedimentary and ecological structure and variability of shelf areas in the Mediterranean sea. In: The Sea. Harvard University Press 14, 1245–1331.

Pinardi, N., Cessi, P., Borile, F., Wolfe, C.L.P., 2019. The Mediterranean Sea Overturning Circulation. J. Physic. Oceanogr. 49(7), 1699-1721. https://journals.ametsoc.org/view/journals/phoc/49/7/jpo-d-18-0254.1.xml

Pinardi, N, Masetti, E., 2000. Variability of the large scale general circulation of the Mediterranean Sea from observations and modelling: a review. Palaeogeogr. Palaeoclimatol. Palaeoecol. 158 (3–4), 153-173. https://doi.org/10.1016/S0031-0182(00)00048-1.

Pinhassi, J., Zweifel, U., Hagström, A., 1997. Dominant marine bacterioplankton species found among colony-forming bacteria. Appl. Environ. Microbiol. 63, 3359–3366.

Planchon, M., Cavagna, A.J., Cardinal, D., André, L., Dehairs, F., 2013. Late summer particulate organic carbon export and twilight zone remineralisation in the Atlantic sector of the Southern Ocean. Biogeosciences 10, 803–820. http://dx.doi.org/10.5194/bg-10-803-2013.

POEM Group, 1992. General Circulation of the Eastern Mediterranean. Earth Sci. Rev. 32, 285-309. https://doi.org/10.1016/0012-8252(92)90002-B.

Porob, S., Nayak, S., Fernandes, A., Padmanabhan, P., Patil, B.A., Meen, R.M., Ramaiah, N., 2013. PCR screening for the surfactin (sfp) gene in marine *Bacillus* strains and its molecular characterization from *Bacillus tequilensis NIOS11*. Turk. J. Biol. 37, 212–221. http://dx.doi.org/10.3906/biy-1206-40.

Powley, H.R., Krom, M.D., Van Cappellen, P., 2016. Circulation and oxygen cycling in the Mediterranean Sea: Sensitivity to future climate change. J. Geophys. Res. Oceans, 121, 8230– 8247. http://dx.doi.org/10.1002/2016JC012224.

Prospero, J.M., 1989. Mineral aerosol transport to the Pacific Ocean. Chem . Oceanogr. 10, 188-218.

Pruysers, P.A., De Lange, G.J., Middelburg, J.J., 1991. Geochemistry of eastern Mediterranean sediments: Primary sediment composition and diagenetic alterations. Mar. Geol. 100, 137–154. https://doi.org/10.1016/0025-3227(91)90230-2.

Psarra, S., Zohary, T., Krom, M.D., Mantoura, R.F.C., Polychronaki, T., Stambler, N., Tanaka, T., Tselepides, A., Thingstad, T.F., 2005. Phytoplankton response to a Lagrangian phosphate addition in the Levantine Sea (Eastern Mediterranean). Deep Sea Res. Part II: Topical Studies in Oceanogr. 52 (22–23), 2944-2960. https://doi.org/10.1016/j.dsr2.2005.08.015 .

Pujo-Pay, M., Conan, P., Oriol, L., Cornet-Barthaux, V., Falco, C., Ghiglione, J.-F., Goyet, C., Moutin, T., Prieur, L., 2011. Integrated survey of elemental stoichiometry (C, N, P) from the western to eastern Mediterranean Sea. Biogeosci. 8, 883–899. https://doi.org/10.5194/bg-8-883-2011.

Pyle, K.M., Hendry, K.R., Sherrell, R.M., Legge, O., Hind, A.J., Bakker, D., Venables, H., Meredith, M.P., 2018. Oceanic fronts control the distribution of dissolved barium in the Southern Ocean. Mar. Chem. 204, 95–106. https://doi.org/10.1016/j.marchem.2018.07.002.

Rademaker, J.L.W., Louws, F.J., Versalovic, J., de Bruijn, F.J., 2004. Characterization of the diversity of ecologically important microbes by rep-PCR genomic fingerprinting. In: Molecular Microbial Ecology Manual. Kluwer Academic Publishers, Dordrecht, 611–644.

Rahmstorf, S., 1998. Influence of Mediterranean Outflow on climate. Eos Trans. AGU, 79 (24), 281– 282. http://dx.doi.org/10.1029/98EO00208.

Rama and Moore, W.S., 1996. Using the radium quartet for evaluating groundwater input and water exchange in salt marshes. Geochim. Cosmochim. Acta 60,4645-4652.

Reguera, I., 2004. Respuesta del Mediterráneo occidental a los cambios bruscos ocurridos durante el último ciclo glacial: Estudio de las asociaciones de foraminíferos, Ph.D. thesis, 231 pp., Univ. de Salamanca, Salamanca, Spain.

Rivadeneyra, M.A., Martín-Algarra, A., Sanchez-Roman, M., Sanchez-Navas, A., Martín-Ramos, J.D., 2010. Amorphous Ca-phosphate precursors for Ca-carbonate biominerals mediated by Chromohalobacter marismortui. ISME J. 4 (7), 922–932. http://dx.doi.org/10.1038/ismej.2010.17.

Robertson, A.H.F., 1990. Geochemistry and mineralogy of iron- and trace-metal-rich Messinian and Pliocene mudstones cored on the Sardinian Margin: ODP Leg 107, Tyrrhenian Sea (central Mediterranean). In: Proceedings of the Ocean Drilling Program, Scientific Results, College Station, TX (Ocean Drilling Program), 107, 245-253. https://doi.org/10.2973/odp.proc.sr.107.191.1990.

Robinson, C.E., Xin, P., Santos, I.R., Charette, M.A., Li, L., Barry, D.A., 2018. Groundwater dynamics in subterranean estuaries of coastal unconfined aquifers: Controls on submarine groundwater discharge and chemical inputs to the ocean. Adv. in Water Resources 115, 315-331. https://doi.org/10.1016/j.advwatres.2017.10.041.

Rodellas, V., Garcia-Orellana, J., Garcia-Solsona, E., Masqué, P., Domínguez, J.A., Ballesteros, B.J., Mejías, M., Zarroca, M., 2012. Quantifying groundwater discharge from different sources into a Mediterranean wetland by using 222Rn and Ra isotopes. J. Hydrol. 466–467, 11-22 https://doi.org/10.1016/j.jhydrol.2012.07.005.

Rodellas, V., Garcia-Orellana, J., Masqué, P., Feldman, M., Weinstein, Y., 2015. SGD to the Mediterranean Sea. Proceed. of the National Academy of Sci. Mar. 112 (13), 3926-3930. https://doi.org/10.1073/pnas.1419049112.

Rodellas, V., Garcia-Orellana, J., Tovar-Sánchez, A., Basterretxea, G., López-Garcia, J.M., Sánchez-Quiles, D., Garcia-Solsona, E., Masqué, P., 2014. Submarine groundwater discharge as a source of nutrients and trace metals in a Mediterranean bay (Palma Beach, Balearic Islands). Mar. Chem. 160, 56-66, https://doi.org/10.1016/j.marchem.2014.01.007.

Rodellas, V., Garcia-Orellana, J., Trezzi, G., Masqué, P., Stieglitz, T.C., Bokuniewicz, H., Cochran, J.K., Berdalet, E., 2017. Using the radium quartet to quantify submarine groundwater discharge and porewater exchange. Geochim. Cosmochim.a Acta 196, 58-73. https://doi.org/10.1016/j.gca.2016.09.016.

Rodrigo-Gámiz, M., 2012. High-frequency Climate Variability and Forcing Mechanisms in the Western Mediterranean During the Last 20,000 yrs: Reconstructing Atmospheric and Oceanic Responses. PhD, Andalusian Institute of Earth Sciences (IACT), Spanish National Research Council-University of Granada (CSIC-UGR).

Rodrigo-Gámiz, M., Martínez-Ruiz, F., Jiménez-Espejo, F., Gallego-Torres, D., Nieto-Moreno, V., Romero, O., Ariztegui, D., 2011. Impact of climate variability in the western Mediterranean during the last 20,000 years: oceanic and atmospheric responses. Quat. Sci. Rev. 30 (15–16), 2018-2034. https://doi.org/10.1016/j.quascirev.2011.05.011.

Rodriguez-Navarro, C., Jimenez-Lopez, C., Rodriguez-Navarro, A., Gonzalez-Muñoz, M.T., Rodriguez-Gallego, M., 2007. Bacterially mediated mineralization of vaterite. Geochim. et Cosmochim. Acta 71 (5), 1197–1213. https://doi.org/10.1016/j.gca.2006.11.031.

Rodriguez-Navarro, C., Jroundi, F., Schiro, M., Ruiz-Agudo, E., González-Muñoz, M.T., 2012. Influence of Substrate Mineralogy on Bacterial Mineralization of Calcium Carbonate: Implications for Stone Conservation. Applied and Environ. Microbiol. 78 (11), 4017–4029. http://dx.doi.org/10.1128/AEM.07044-11.

Rodwell, M., Rowell, D., Folland, C., 1999. Oceanic forcing of the wintertime North Atlantic Oscillation and European climate.

Nature 398, 320–323. https://doi.org/10.1038/18648.

Roether, W., Klein, B., Beitzel, V., Manca, B.B., 1998. Property distributions and transient-tracer ages in Levantine Intermediate Water in the Eastern Mediterranean. J. Mar Syst. 18 (1–3), 71-87. https://doi.org/10.1016/S0924-7963(98)00006-2.

Roether, W., Klein, B., Hainbucher, D., 2014. The eastern mediterranean transient: Evidence for similar events previously?. Geoph. Monograph Series 202, 75-83.

Roether, W., Klein, B., Manca, B.B., Theocharis, A., Kioroglou, S., 2007. Transient Eastern Mediterranean deep waters in response to the massive dense-water output of the Aegean Sea in the 1990s. Progr. Oceanogr. 74 (4), 540-571. https://doi.org/10.1016/j.pocean.2007.03.001.

Roether, W., Manca, B.B., Klein, B., Bregant, D., Georgopoulos, D., Beitzel, V., Kovacevic, V., Luchetta, A., 1996. Recent changes in Eastern Mediterranean Deep Waters. Science 271 (5247), 333-335. http://dx.doi.org/10.1126/science.271.5247.333.

Roether, W., Schlitzer, R., 1991. Eastern Mediterranean deep water renewal on the basis of chlorofluoromethane and tritium data. Dyn. Atmosph. Oceans 15 (3–5), 333-354. https://doi.org/10.1016/0377-0265(91)90025-B.

Roether, W., Well, R., 2001. Oxygen consumption in the Eastern Mediterranean. Deep Sea Res. Part I: Oceanogr. Res. Papers 48 (6), 1535-1551. https://doi.org/10.1016/S0967-0637(00)00102-3.

Roeske, T., Bauch, D., Rutgers, M., Loeff, V.D., Rabe, B., 2012. Utility of dissolved barium in distinguishing North American from Eurasian runoff in the Arctic Ocean. Mar. Chem. 132–133, 1–14. https://doi.org/10.1016/j.marchem.2012.01.007.

Rogerson, M., Cacho, I., Jimenez-Espejo, F., Reguera, M. I., Sierro, F. J., Martinez-Ruiz, F., Frigola, J., and Canals, M., 2008. A dynamic explanation for the origin of the western Mediterranean organic-rich layers. Geochem. Geophys. Geosyst. 9, Q07U01, https://doi.org/10.1029/2007GC001936.

Rogerson, M., Rohling, E. J., Weaver, P.P.E., 2006. Promotion of meridional overturning by Mediterranean-derived salt during the last deglaciation, Paleoceanogr. 21, PA4101. https://doi.org/10.1029/2006PA001306.

Rohling, E.J., 1991. A simple two-layered model for shoaling of the eastern Mediterranean pycnocline due to glacio-eustatic sea-level lowering. Paleoceanogr. 6, 537–541. http://dx.doi.org/10.1029/91PA01328.

Rohling, E.J., Abu-Zied, R.H., Casford, J.S.L., Hayes, A. and Hoogakker, B.A.A., 2009. The Marine Environment: Present and Past. In, Woodward, J.C. (ed.) The Physical Geography of the Mediterranean. Oxford, UK. Oxford University Press, 33-67.

Rohling, E.J., Cane, T.R., Cooke, S., Sprovieri, M., Bouloubassi, I., Emeis, K.C., Schiebel, R., Kroon, D., Jorissen, F., Lorre, A., Kemp, A.E.S., 2002. African monsoon variability during the previous interglacial maximum. Earth Planet. Sci. Lett. 202, 61-75. http://dx.doi.org/10.1016/S0012-821X(02)00775-6.

Rohling, E.J., Gieskes, W.W.C., 1989. Late Quaternary changes in Mediterranean intermediate water density and formation rate. Paleoceanogr. 4(5), 531– 545. http://dx.doi.org/10.1029/PA004i005p00531.

Rohling, E.J., Hilgen, FJ., 1991. The eastern Mediterranean climate at times of sapropel formation: a review. Geol. Mijnbouw, 70, 253-264.

Rohling, E.J., Hopmans, E.C., Sinninghe-Damsté, J.S., 2006. Water column dynamics during the last interglacial anoxic event in the Mediterranean (sapropel S5). Paleoceanogr. 21, PA2018, http://dx.doi.org/10.1029/2005PA001237.

Rohling, E.J., Marino, G., Grant, K.M., 2015. Mediterranean climate and oceanography, and the periodic development of anoxic events (sapropels). Earth-Science Reviews 143, 62–97. https://doi.org/10.1016/j.earscirev.2015.01.008.

Rohling, E.J., Sprovieri, M., Cane, T.R., Casford, J.S.L., Cooke, S., Bouloubassi, I., Emeis, K.C., Schiebel, R., Hayes, A., Jorissen, F.J., Kroon, D., 2004. Reconstructing past planktic foraminiferal habitats using stable isotope data: a case history for Mediterranean sapropel S5. Mar. Micropaleontol. 50, 89-123. http://dx.doi.org/10.1016/S0377-8398(03)00068-9.

Romero-González, M., Nwaobi B.C., Hufton, J.M., Gilmour, D.J., 2016. Ex-situ Bioremediation of U(VI) from Contaminated Mine Water Using *Acidithiobacillus ferrooxidans* Strains. Frontiers in Environ. Sci. 4, 39. https://www.frontiersin.org/article/10.3389/fenvs.2016.00039.

Rossignol-Strick, M., 1985. Mediterranean Quaternary sapropels, an immediate response of the African monsoon to variation of insolation. Palaeogeogr. Palaeoclimatol. Palaeoecol. 49 (3–4) 237-263. https://doi.org/10.1016/0031-0182(85)90056-2.

Rossignol-Strick, M., Nesteroff, W., Olive, P., Vergnaud-Grazzini, C., 1982. After the deluge: Mediterranean stagnation and sapropel formation. Nature 295, 105–110. https://doi.org/10.1038/295105a0.

Rossignol-Strick, M., Paterne, M., 1999. A synthetic pollen record of the eastern Mediterranean sapropels of the last 1 Ma: implications for the time-scale and formation of sapropels. Mar. Geol. 153 (1–4), 221–237. https://doi.org/10.1016/S0025-3227(98)00080-2.

Roy-Barman, M., Pons-Branchu, E., Levier, M., Bordier, L., Foliot, L., Gdaniec, S., Ayrault, S., Garcia-Orellana, J., Masque, P., Castrillejo, M., 2019. Barium during the GEOTRACES GA-04S MedSeA cruise: The Mediterranean Sea Ba budget revisited. Chem. Geol. 511, 431–440. https://doi.org/10.1016/j.chemgeo.2018.09.015.

Rožič, P.Z., Dolenec, T., Baždarić, B., Karamarko, V., Kniewald, G., Dolenec, M., 2012. Major, minor and trace element content derived from aquacultural activity of marine sediments (Central Adriatic, Croatia). Environ. Sci. Pollut. Res. 19, 2708–2721. https://doi.org/10.1007/s11356-012-0769-4.

Rubin, S.I., King, S.L., Jahnke, R.A., Froelich, P.N., 2003. Benthic barium and alkalinity fluxes: Is Ba an oceanic paleo-alkalinity proxy for glacial atmospheric CO_2? Geophys. Res. Lett. 30 (17), 1885. https://doi.org/10.1029/2003GL017339.

Rushdi, A., McManus, J., Collier, R.W., 2000. Marine barite and celestite saturation in seawater. Mar. Chem., 69 (1–2), 19–31. https://doi.org/10.1016/S0304-4203(99)00089-4.

Rutsch, H.J., Mangini, A., Bonani, G., Dittrich-Hannen, B., Kubik, P.W., Suter, M., Segl, M.,

1995. ^{10}Be and Ba concentrations in West African sediments trace productivity in the past. Earth Planet. Sci. Lett. 133 (1–2), 129-143. https://doi.org/10.1016/0012-821X(95)00069-O.

Rutten, A., De Lange, G.J., Ziveri, P., Thomson, J., Van Santvoort, P.J.M., Colley, S., Corselli, C., 2000. Recent terrestrial and carbonate fluxes in the pelagic eastern Mediterranean; a comparison between sediment trap and surface sediment. Palaeogeogr. Palaeoclimat. Palaeoecol. 158 (3–4), 197-213. https://doi.org/10.1016/S0031-0182(00)00050-X.

Ryan, W.B.F., Hsü, K.J., Cita, M.B., Dumitrica, P., Llort, J., Maync, W., Nesteroff, W.D., Pautot, G., Stradner, H., Wezel, F.D., 1973. Initial Reports of the Deep Sea Drilling Project, Volume 13: Washington (U.S. Government Printing Office).Sancetta C. (1999) - The Mistery of the sapropels. Nature 398, 6722.

Rykaczewski, R.R., Dunne, J.P., 2010. Enhanced nutrient supply to the California Current Ecosystem with global warming and increased stratification in an earth system model. Geophys. Res. Lett. 37, L21606. http://dx.doi.org/10.1029/2010GL045019.

Saccà, C., Saccà, D., Nucera, P., De Fazio, A., 2011. Composition and geochemistry of clay sediments offshore the northeastern Sicilian coast (Southeastern Tyrrhenian Sea, Italy). Estuar. Coast. Shelf Sci. 92 (4), 564-572. https://doi.org/10.1016/j.ecss.2011.02.012.

Sadowsky, M.J., Hur, H.G., 1998. Use of endogenous repeated sequences to fingerprint bacterial genomic DNA. In: Bacterial Genomics: Physical Structure and Analysis. Chapman and Hall, New York, 399–413.

Sajih, M., Bryan, N.D., Livens, F.R., Vaughan, D.J., Descostes, M., Phrommavanh, V., Nos, J., Morris, K., 2014. Adsorption of radium and barium on goethite and ferrihydrite: A kinetic and surface complexation modelling study. Geochim. Cosmochim. Acta 146, 150-163. https://doi.org/10.1016/j.gca.2014.10.008.

Sanchez-Moral, S., Luque, L., Cañaveras, J.C., Laiz, L., Jurado, V., Hermosin, B., Saiz-Jimenez, C., 2004. Bioinduced barium precipitation in St. Callixtus and Domitilla catacombs. Ann. Microbiol. 54 (1), 1–12.

Sánchez-Román, A., Criado-Aldeanueva, F., García-Lafuente, J., Sánchez, J.C., 2008. Vertical structure of tidal currents over Espartel and Camarinal sills, Strait of Gibraltar. J. Mar. Syst. 74 (1–2), 120-133. https://doi.org/10.1016/j.jmarsys.2007.11.007.

Santos, I.R., Burnett, W.C., Misra, S., Suryaputra, I.G.N.A., Chanton, J.P., Dittmar, T., Peterson, R.N., Swarzenski, P.W., 2011. Uranium and barium cycling in a salt wedge subterranean estuary: The influence of tidal pumping. Chem. Geol. 287, 114–123. https://doi.org/10.1016/j.chemgeo.2011.06.005.

Sanyal, S.K., Shuster, J., Reith, F., 2019. Biogeochemical gold cycling selects metal-resistant bacteria that promote gold particle transformation. FEMS Microbiol. Ecol. 95 (7), fiz078. https://doi.org/10.1093/femsec/fiz078.

Saravanan, P., Nancharaiah, Y.V., Venugopalan, V.P., Rao, T.S., Jayachandran, S., 2006. Biofilm formation by *Pseudoalteromonas ruthenica* and its removal by chlorine. Biofouling 22, 371–381. http://dx.doi.org/10.1080/08927010601029103.

Sarmiento, J.L., Gruber, N., McElroy, M.B., 2007. Books-Ocean Biogeochemical Dynamics. Physics Today 60(6), 63.

Sawabe, T., Tanaka, R., Iqbal, M.M., Tajima, K., Ezura, Y., Ivanova, E.P., Chisten, R., 2000. Assignment of *Alteromonas elyakovii KKM 162T* and five strains isolated from spotwounded fronds of *Laminaria japonica* to *Pseudoalteromonas elyakovii* comb. nov. and the extended description of the species. Int. J. Syst. Evol. Microbiol. 50 (1), 265–271. http://dx.doi.org/10.1099/00207713-50-1-265.

Schäfer, H., Servais, P., Muyzer, G., 2000. Successional changes in the genetic diversity of a marine bacterial assemblage during confinement. Arch. Microbiol. 173 (2), 138–145. http://dx.doi.org/10.1007/s002039900121.

Schenau, S.J., Antonarakou, A., Hilgen, F.J., Lourens, L.J., Nijenhuis, I.A., Van der Weijden, C.H., Zachariasse, W.J., 1999. Organic-rich layers in the Metochia section (Gavdos, Greece): evidence for a single mechanism of sapropel formation during the past 10 My. Mar. Geol. 153 (1–4), 117-135. https://doi.org/10.1016/S0025-3227(98)00086-3.

Schenau, S.J., Prins, M.A., De Lange, G.J., Monnin, C., 2001. Barium accumulation in the Arabian Sea: controls on barite preservation in marine sediments. Geochim. Cosmochim. Acta 65 (10), 1545–1556. https://doi.org/10.1016/S0016-7037(01)00547-6.

Schlitzer, R., 2013. Ocean Data View, http://odv.awi.de.

Schlitzer, R., Roether, W., Oster, H., Junghans, H.G., Hausmann, M., Johannsen, H., Michelato, A., 1991. Chlorofluoromethane and oxygen in the Eastern Mediterranean. Deep Sea Res. Part A. Oceanogr. Res. Papers 38 (12), 1531-1551. https://doi.org/10.1016/0198-0149(91)90088-W.

Schmitz, B., 1987. Barium, equatorial high productivity, and the northward wandering of the Indian continent. Paleoceanogr. 2(1), 63– 77. https://doi.org/10.1029/PA002i001p00063.

Schoepfer, S.D., Shen, J., Wei, H., Tyson, R.V., Ingall, E., Alge, A.J., 2015. Total organic carbon, organic phosphorus, and biogenic barium fluxes as proxies for paleomarine productivity. Earth-Sci. Rev. 149, 23–52. https://doi.org/10.1016/j.earscirev.2014.08.017.

Schroeder, T., Devlin, M.J., Brando, V.E., Dekker, A.G., Brodie, J.E., Clementson, L.A., McKinna, L., 2012. Inter-annual variability of wet season freshwater plume extent into the Great Barrier Reef lagoon based on satellite coastal ocean colour observations. Mar. Pollution Bull. 65 (4–9), 210-223. https://doi.org/10.1016/j.marpolbul.2012.02.022.

Schroeder, K., Ribotti, A., Borghini, M., Sorgente, R., Perilli, A., and Gasparini, G.P., 2008. An extensive western Mediterranean deep water renewal between 2004 and 2006. Geophys. Res. Lett. 35, L18605. https://doi.org/10.1029/2008GL035146.

Schubert, M., Knöller, K., Stollberg, R., Mallast, U., Ruzsa, G., Melikadze, G., 2017. Evidence for Submarine Groundwater Discharge into the Black Sea—Investigation of Two Dissimilar Geographical Settings. Water *9*, 468. https://doi.org/10.3390/w907046.

Schwarzkopf, T.A., 1993. Model for prediction of organic carbon content in possible source rocks. Mar. Petrol. Geol. 10 (5), 478-492. https://doi.org/10.1016/0264-8172(93)90049-X.

Senko, J.M., Campbell, B.S., Henriksen, J.R., Elshahed, M.S., Dewers, T.A., Krumholz, L.R., 2004. Barite deposition resulting from phototrophic sulfide-oxidizing bacterial activity. Geochim. Cosmochim. Acta 68 (4), 773–780. http://dx.doi.org/10.1016/j.gca.2003.07.008.

Sharp, J.H., Benner, R., Bennett, L., Carlson, C.A., Fitzwater, S.E., Peltzer, E.T., Tupas, L.M., 1995. Analyses of Dissolved Organic Carbon in Seawater: The Jgofs Eqpac Methods Comparison. Mar. Chem. 48, 91–108. https://doi.org/10.1016/0304-4203(94)00040-K.

Shaw, T.J., Moore, W.S., Kloepfer, J., Sochaski, M.A., 1998. The flux of barium to the coastal waters of the southeastern USA: the importance of submarine groundwater discharge. Geochim. Cosmochim. Acta 62 (18), 3047-3054. https://doi.org/10.1016/S0016-7037(98)00218-X.

Seshadri, S., Prakash, A., Kowsik, M., 2012. Biosynthesis of silver nanoparticles by marine bacterium, *Idiomarina sp. PR58-8.* Bull. Mater. Sci. 35(7), 1201–1205. http://dx.doi.org/10.1007/s12034-012-0417-0.

Siddall, M., Rohling, E., Almogi-Labin, A., Hemleben, C., Meischner, D., Schmelzer, I., Smeed, D.A., 2003. Sea-level fluctuations during the last glacial cycle. Nature 423, 853–858. https://doi.org/10.1038/nature01690.

Singer, D.M., Griffith, E.M., Senko, J.M., Fitzgibbon, K., Widanagamage, I.H., 2016. Celestine in a sulfidic spring barite deposit - A potential biomarker?. Chem. Geol. 440, 15–25. https://doi.org/10.1016/j.chemgeo.2016.06.022.

Sillitoe, R.H., 2005. Supergene oxidized and enriched porphyry copper and related deposits. In: Economic Geol. One Hundredth Anniversary Volume, 1905–2005. Littleton, Colorado , Society of Economic Geol. 723–768.

Singh, A.K., Marcantonio, F., Lyle, M., 2020. An Assessment of xsBa Flux as a Paleoproductivity Indicator and Its Water-Depth Dependence in the Easternmost Equatorial Pacific Ocean. Paleocean. Paleoclim. 35 (12). https://doi.org/10.1029/2020PA003945.

Singh, S.P., Singh, S.K., Bhushan, R., 2013. Internal cycling of dissolved barium in water column of the Bay of Bengal. Mar. Chem. 154, 12-23. https://doi.org/10.1016/j.marchem.2013.04.013.

Siokou-Frangou, I., Christaki, U., Mazzocchi, M.G., Montresor, M., Ribera d'Alcalá, M., Vaqué, D., Zingone, A., 2010. Plankton in the open Mediterranean Sea: a review. Biogeosci. 7, 1543–1586. https://doi.org/10.5194/bg-7-1543-2010.

Smith, D., Simon, M., Alldredge, A., Azam, F., 1992. Intense hydrolytic enzyme activity on marine aggregates and implications for rapid particle dissolution. Nature 359, 139–142. https://doi.org/10.1038/359139a0.

Somot, S., Sevault, F., Déqué, M., Crépon, M., 2008. 21st century climate change scenario for the Mediterranean using a coupled atmosphere–ocean regional climate model. Global and Planet. Change 63 (2–3), 112-126. https://doi.org/10.1016/j.gloplacha.2007.10.003.

Soto-Navarro, F.J., 2012. ExchangethroughtheStraitofGibraltarinrelationtotheclimaticforcingovertheMediterraneanSea. Tesis doctoral, Universidad de Málaga.

Soto-Navarro, J., Criado-Aldeanueva, F., García-Lafuente, J., Sánchez-Román, A., 2010. Estimation of the Atlantic inflow through the Strait of Gibraltar from climatological and in situ data. J. Geophys. Res. 115, C10023. https://doi.org/10.1029/2010JC006302.

Spangenberg, J.V. Cherr, G.N., 1996. Developmental effects of barium exposure in a marine bivalve (*Mytilus californianus*).

Environ. Toxicol. and Chem. 15, 1769-1774. https://doi.org/10.1002/etc.5620151018.

Sparnocchia, S., Gasparini, G.P., Astraldi, M., Borghini, M., Pistek, P., 1999. Dynamics and mixing of the Eastern Mediterranean outflow in the Tyrrhenian Sea. J. Mar. Syst. 20, 301–317. https://doi.org/10.1016/S0924-7963(98)00088-8.

Spötl, C., Nicolussi, K., Patzelt, G., Boch, R., 2010. Humid climate during deposition of sapropel 1 in the Mediterranean Sea: Assessing the influence on the Alps. Global Planet. Change 71 (3–4), 242–248. https://doi.org/10.1016/j.gloplacha.2009.10.003.

Stanford, J.D., Hemingway, R., Rohling, E.J., Challenor, P.G., Medina-Elizalde, M., Lester, A.J., 2011. Sea-level probability for the last deglaciation: A statistical analysis of far-field records. Global Planet. Change 79 (3–4), 193–203. https://doi.org/10.1016/j.gloplacha.2010.11.002.

Stein, R., 1986. Organic carbon and sedimentation rate — Further evidence for anoxic deep-water conditions in the Cenomanian/Turonian Atlantic Ocean. Mar. Geol. 72 (3–4), 199–209. https://doi.org/10.1016/0025-3227(86)90119-2.

Sternberg, E., Tang, D., Ho, T.Y., Jeandel, C., Morel, F.M.M., 2005. Barium uptake and adsorption in diatoms, Geochim. Cosmochim. Acta 69 (11), 2745-2752. https://doi.org/10.1016/j.gca.2004.11.026.

Stevens, E.W.N., Bailey, J.V., Flood, B.E., Jones, D.S., Gilhooly, W.P., III, Joye, S.B., Teske, A., Mason, O.U., 2015. Barite encrustation of benthic sulfur-oxidizing bacteria at a marine cold seep. Geobiol. 13, 588–603. https://doi.org/10.1111/gbi.12154.

Stoll, M.H.C., Bakker, K., Nobbe, G.H., Haese, R.R., 2001. Continuous-Flow Analysis of Dissolved Inorganic Carbon Content in Seawater. Anal. Chem. 73 (17), 4111–4116. https://doi.org/10.1021/ac010303r.

Stratford, K., Williams, R.G., 1997. A tracer study of the formation, dispersal, and renewal of Levantine Intermediate Water. J. Geophys. Res. 102 (C6), 12539– 12549. https://doi.org/10.1029/97JC00019.

Strickland, J.D.H., Parson, T.R., 1968. A manual of seawater analysis. Bull. Fisheries Res. Board Can. 167, 311.

Struglia, M., Mariotti, A., Filograsso, A., 2004. River Discharge into the Mediterranean Sea: Climatology and Aspects of the Observed Variability. Journal of Climate, 17 (24), 4740-4751.

Sugiyama, M., Matsui, M., Nakayama, E., 1984. Direct determination of barium in Sea water by inductively coupled plasma emission spectrometry. J. Oceanogr. Soc. of Japan 40, 295–302. https://doi.org/10.1007/BF02302522.

Sun, X., Wang, S., Zhang, X., Chen, J.P., Li, X., Gao, B., Ma, Y., 2009. Spectroscopic study of Zn2+ and Co2+ binding to extracellular polymeric substances (EPS) from aerobic granules. J. Colloid and Interface Sci. 335 (1), 11-17. https://doi.org/10.1016/j.jcis.2009.03.088.

Supić, N., Grbec, B., Vilibić, I., Ivančić, I., 2004. Long-term changes in hydrographic conditions in northern Adriatic and its relationship to hydrological and atmospheric processes.Ann. Geophys. 22, 733–745. https://doi.org/10.5194/angeo-22-733-2004, 2004.

Sur, H., Ozsoy, E., Ilyin, Y.P., Unluata, U., 1996. Coastal/deep inter-actions in the Black Sea and their ecological/environmen-tal impacts. J. Mar. Syst. 7, 293–32.

Suzuki, M.T., Rappé, M.S., Haimberger, Z.W., Winfield, H., Adair, N., Strobel, J., Giovannoni, S.J., 1997. Bacterial diversity among small-subunit rRNA genes clones and cellular isolates from the same seawater sample. Appl. Environ. Microbiol. 63, 983–989. http://dx.doi.org/10.1128/AEM.63.3.983-989.1997.

Sweet, W.V., Zervas, C., 2011. Cool-Season Sea Level Anomalies and Storm Surges along the U.S. East Coast: Climatology and Comparison with the 2009/10 El Nino, Mon. Weather Rev. 139 (7), 2290– 2299. ttps://doi.org/10.1175/MWR-D-10-05043.1.

Swingedouw, D., Colin, C., Eynaud, F., Ayache, M., Zaragosi, S., 2019. Impact of freshwater release in the Mediterranean Sea on the North Atlantic climate. Clim. Dyn. 53, 3893–3915. https://doi.org/10.1007/s00382-019-04758-5.

Tachikawa, K., Vidal, L., Cornuault, M., Garcia, M., Pothin, A., Sonzogni, C., Bard, E., Menot, G., Revel, M., 2015. Eastern Mediterranean Sea circulation inferred from the conditions of S1 sapropel deposition. Clim. Past. 11, 855–867. https://doi.org/10.5194/cp-11-855-2015.

Tamura, K., Dudley, J., Nei, M., Kumar, S., 2007. MEGA4: molecular evolutionary genetics analysis (MEGA) software version 4.0. Mol. Biol. Evol. 24, 1596–1599. http://dx.doi.org/10.1093/molbev/msm092.

Tanaskovski, B., Petrović, M., Kljajic, Zo., Degetto, S., Stankovic, S., 2014. Analysis of major, minor and trace elements in surface sediments by X-ray fluorescence spectrometry for assessment of possible contamination of Boka Kotorska Bay, Montenegro. Maced. J. Chem. and Chem. Engin. 33, 139–150. http://dx.doi.org/10.20450/mjcce.2014.423.

Tanhua, T., Hainbucher, D., Schroeder, K., Cardin, V., Álvarez, M., Civitarese, G., 2013. The Mediterranean Sea system: a review and an introduction to the special issue. Ocean Sci. 9, 789–803. https://doi.org/10.5194/os-9-789-2013.

Tesi, T., Asioli, A., Minisini, D., Maselli, V., Dalla Valle, G., Gamberi, F., Langone, L., Cattaneo, L., Montagna, P., Trincardi, F., 2017. Large-scale response of the Eastern Mediterranean thermohaline circulation to African monsoon intensification during sapropel S1 formation. Quat. Sci. Rev. 159, 139–154. https://doi.org/10.1016/j.quascirev.2017.01.020.

Theocharis, A., 2009. Variability of the thermohaline properties in the Eastern Mediterranean during the post-EMT period (1995–2008) and SST changes in the Aegean (1985–2008). Dynamics of Mediterranean deep waters, CIESM Workshop Monographs 38, 35-40.

Theodosi, C., Markaki, Z., Pantazoglou, F., Tselepides, A., Mihalopoulos, N., 2019. Chemical composition of downward fluxes in the Cretan Sea (Eastern Mediterranean) and possible link to atmospheric deposition: A 7 year survey. Deep Sea Res. Part II: Topical Studies in Oceanogr. 164, 89-99. https://doi.org/10.1016/j.dsr2.2019.06.003.

Thingstad, T.F., Krom, M., Mantoura, R., Flaten, G.A., Groom, StS., Herut, B., Kress, N., Law, C., Pasternak, A., Pitta, P., Psarra, S., Rassoulzadegan, F., Tanaka, T., Tselepides, A., Wassmann, P., Woodward, E., Riser, C., Zodiatis, G., Zohary, T., 2005. Nature of Phosphorus Limitation in the Ultraoligotrophic Eastern Mediterranean. Science 309, 1068-71. https://doi.org/10.1126/science.1112632.

Thomson, J., Croudace, I.W., Rothwell, R.G., 2006. A geochemical application of the ITRAX scanner to a sediment core containing eastern Mediterranean sapropel units. Geological Society, London, Special Publications 267, 65-77. https://doi.org/10.1144/GSL.SP.2006.267.01.05.

Thomson, J., Higgs, N.C., Wilson, T.R.S., Croudace, I.W., De Lange, G.J., Van Santvoort, P.J.M., 1995. Redistribution and geochemical behaviour of redox-sensitive elements around S1, the most recent Eastern Mediterranean sapropel. Geochim. Cosmochim. Acta 59, 3487-3501. https://doi.org/10.1016/0016-7037(95)00232-O.

Thunell, R.C., Williams, D.F., Kennett, J.P., 1977. Late Quaternary paleoclimatology, stratigraphy and sapropel history in eastern Mediterranean deep-sea sediments. Mar. Micropal. 2, 371-388. https://doi.org/10.1016/0377-8398(77)90018-4.

Tixeront, J., 1970. Le bilan hydrologique de la mer Noire et de la mer Méditerranée. Cah. Oceanogr 22 , 227–237.

Tolmazin, D., 1985. Changing Coastal oceanography of the Black Sea. I: Northwestern Shelf. Progr. Oceanogr. 15 (4), 217-276. https://doi.org/10.1016/0079-6611(85)90038-2.

Tornos, F., Oggerin, M., de los Ríos, A., Rodriguez, N., Amils, R., Sanz, J.L., Rojas, P., Velasco, F., Escobar, J.M., Gómez, C., Slack, J.F., 2019. Do microbes control the formation of giant copper deposits?. Geol. 47 (2), 143–146. https://doi.org/10.1130/G45573.1.

Torres, M.E., Barry, J.P., Hubbard, D.A., Suess, E., 2001. Reconstructing the history of fluid flow at cold seep sites from Ba/Ca ratios in vesicomyid clam shells. Limnol. Oceanogr. 7. https://doi.org/10.4319/lo.2001.46.7.1701.

Torres, M.E., Bohrmann, G., Suess, E., 1996. Authigenic barites and fluxes of barium associated with fluid seeps inthe Peru subduction zone. Earth Planet. Sci. Lett. 144, 469–481. https://doi.org/10.1016/S0012-821X(96)00163-X.

Torres, M.E., McManus, J., Huh, C.A., 2002. Fluid seepage along the San Clemente Fault scarp: basin-wide impact on barium cycling, Earth and Planet. Sci. Lett. 203 (1), 181-194. https://doi.org/10.1016/S0012-821X(02)00800-2.

Toucanne, S., Soulet, G., Freslon, N., Silva Jacinto, R., Dennielou, B., Zaragosi, S., Eynaud, F., Bourillet, J.F., Bayon, G., 2015. Millennial-scale fluctuations of the European Ice Sheet at the end of the last glacial, and their potential impact on global climate. Quat. Sci. Rev. 123, 113–133. https://doi.org/10.1016/j.quascirev.2015.06.010.

Tourney, J., Ngwenya, B.T., 2014. The role of bacterial extracellular polymeric substances in geomicrobiology. Chem. Geol. 386, 115–132.

Tovar-Sánchez, A., Arrieta, J.M., Duarte, C.M., Sañudo-Wilhelmy, S.A., 2014. Spatial gradients in trace metal concentrations in the surface microlayer of the Mediterranean Sea. Front. Mar. Sci. 1, 79. https://doi.org/10.3389/fmars.2014.00079.

Tremblay, L., Larocque, M., Anctil, F., Rivard, C., 2011. Teleconnections and interannual variability in Canadian groundwater levels. J. Hydrol. 410 (3–4), 178-188. https://doi.org/10.1016/j.jhydrol.2011.09.013.

Trezzi, G., Garcia-Orellana, J., Rodellas, V., Masqué, P., A., Garcia-Solsona, Andersson, P.S., 2017. Assessing the role of submarine groundwater discharge as a source of Sr to the Mediterranean Sea. 2017. Geochim. Cosmochim. Acta 200, 42-54. https://doi.org/10.1016/j.gca.2016.12.005.

Trezzi, G., Garcia-Orellana, J., Rodellas, V., Santos-Echeandia, J., Tovar-Sánchez, A., Garcia-Solsona, E., Masqué, P., 2016. Submarine groundwater discharge: a significant source of dissolved trace metals to the North Western Mediterranean Sea. Mar. Chem. 186, 90–100. https://doi.org/10.1016/j.marchem.2016.08.004.

Triantaphyllou, M.V., Ziveri, P., Gogou, A., Marino, G., Lykousis, V., Bouloubassi, I., Emeis, K.C., Kouli, K., Dimiza, M., Rosell-Melé, A., Papanikolaou, M., Katsouras, G., Nunez, N., 2009. Late Glacial–Holocene climate variability at the south-eastern margin of the Aegean Sea. Mar. Geol. 266 (1–4), 182-197.https://doi.org/10.1016/j.margeo.2009.08.005.

Tsimplis, M.N., Josey, S.A., 2001. Forcing of the Mediterranean Sea by atmospheric oscillations over the North Atlantic. Geophys. Res. Lett. 28 (5), 0094-8276. https://doi.org/10.1029/2000GL012098.

Tsimplis, M.N., Marcos, M., Somot, S., 2008. 21st century Mediterranean sea level rise: Steric and atmospheric pressure contributions from a regional model. Global Planet. Change 63 (2–3), 105-111. https://doi.org/10.1016/j.gloplacha.2007.09.006.

Tugrul, S., Basturk, O., Saydam, C., Yilmaz, A., 1992. Changes in the hydrochemistry of the Black Sea inferred from water density profiles. Nature 359, 137–139. https://doi.org/10.1038/359137a0.

Turekian, K., Tausch, E., 1964. Barium in Deep-sea Sediments of the Atlantic Ocean. Nature 201, 696–697. https://doi.org/10.1038/201696b0.

Turley, C.M., Bianchi, M., Christaki, U., Conan, P., Harris, J.R.W., Psarra, S., Ruddy, G., Stutt, E.D., Tselepides, A., Van Wambeke, F., 2000. Relationship between primary producers and bacteria in an oligotrophic sea-the Mediterranean and biogeochemical implications. Mar. Ecol. Prof. Ser. 193, 11–18. https://doi.org/10.3354/meps193011.

Tyson, R.V., 1995. Sedimentary Organic Matter: Organic Facies and Palynofacies. Chapman and Hall, London. https://doi.org/10.1007/978-94-011-0739-6.

Tyson, R.V., 2001. Sedimentation rate, dilution, preservation and total organic carbon: some results of a modelling study. Org. Geochem. 32 (2), 333-339. https://doi.org/10.1016/S0146-6380(00)00161-3.

Tyson, R.V., 2005. The “productivity versus preservation” controversy; cause, flaws, and resolution. In: Deposition of Organic-carbon-rich Sediments: Models, Mechanisms, and Consequences. Society for Sedimentary Geology (SEPMSSG) Special Publication 82, 17–33.

Tziperman, E., Speer, K., 1994. A study of water mass transformation in the Mediterranean Sea: Analysis of climatological data and a simple three-box model. Dynam. Atmosph. and Oceans 21 (2-3), 53-82.

Unluata, U., Oguz, T., Latif, M.A., Özsoy, E., 1990. On the Physical Oceanography of the Turkish Straits. In: The Physical Oceanography of Sea Straits, NATO/ASI Series, Kluwer, Dordrecht.

Van Beek, P., Francois, R., Conte, M., Reyss, J.L., Souhau,t M., Charette, M., 2007. ^{238}Ra/^{226}Ra and ^{226}Ra/Ba ratios to track barite formation and transport in the water column. Geochimic. Cosmochim. Acta 71, 71–86. http://dx.doi.org/10.1016/j.gca.2006.07.041.

Van Beek, P., Sternberg, E., Reyss, J.L., Souhaut, M., Robin, E., Jeandel, C., 2009.

228Ra/226Ra and 226Ra/Ba ratios in the Western Mediterranean Sea: Barite formation and transport in the water column. Geoch. Cosmoch. Acta 73 (16), 4720–4737. https://doi.org/10.1016/j.gca.2009.05.063.

Van Cappellen, P., Powley, H.R., Emeis, K.C., Krom, M.D., 2014. A biogeochemical model for phosphorus and nitrogen cycling in the Eastern Mediterranean Sea: Part 1. Model development, initialization and sensitivity. J. Mar. Syst. 139, 460-471. https://doi.org/10.1016/j.jmarsys.2014.08.016.

Van Der Wal, A., Norde, W., Zehnder, A.J.B., Lyklema, J., 1997. Determination of the total charge in the cell walls of Gram-positive bacteria. Colloids and Surfaces B: Biointerfaces 9 (1–2), 81–100. https://doi.org/10.1016/S0927-7765(96)01340-9.

Van Helmond, N., Hennekam, R., Donders, T.H., Bunnik, F.P.M., De Lange, G.J., Brinkhuis, H., Sangiorgi, F., 2015. Marine productivity leads organic matter preservation in sapropel S1: palynological evidence from a core east of the Nile River outflow. Quat. Sci. Rev. 108, 130–138. https://doi.org/10.1016/j.quascirev.2014.11.014.

Van Os, B.J.H., Middelburg, J.J., De Lange, G.J., 1991. Possible diagenetic mobilization of barium in sapropelic sediment from the eastern Mediterranean. Mar. Geol. 100 (1–4), 125-136. https://doi.org/10.1016/0025-3227(91)90229-W.

Van Santvoort, P.J.M., De Lange, G.J., Langereis, C.G., Dekkers, M.J., Paterne, M., 1997. Geochemical and paleomagnetic evidence for the occurrence of “missing” sapropels in eastern Mediterranean sediments. Paleoceanogr. 12(6), 773–786. https://doi.org/10.1029/97PA01351.

Van Santvoort, P.J.M., De Lange, G.J., Thomson, J., Cussen, H., Wilson, T.R.S., Krom, M., Strohle, K., 1996. Active post-depositional oxidation of the most recent sapropel (S1) in sediments of the eastern Mediterranean. Geochim. Cosmochim. Acta 60, 4007–4024.

Van Trappen, S., Tan, T.L., Yang, J.,Mergaert, J., Swings, J., 2004. *Alteromonas stellipolaris* sp. nov., a novel, budding, prosthecate bacterium from Antarctic seas, and emended description of the genus *Alteromonas*. Int. J. Syst. Evol. Microbiol. 54 (Pt 4), 1157–1163. http://dx.doi.org/10.1099/ijs.0.02862-0.

Varnavas, S.P., Cronan, D.S., 1991. Hydrothermal metallogenic processes off the islands of Nisiros and Kos in the Hellenic Volcanic Arc. Mar. Geol. 99 (1–2), 109-133.https://doi.org/10.1016/0025-3227(91)90086-J.

Vasiliev, I., Karakitsios, V., Bouloubassi, I., Agiadi, K., Kontakiotis, G., Antonarakou, A., Triantaphyllou, M., Gogou, A., Kafousia, N., de Rafélis, M., Zarkogiannis, S., Kaczmar, F., Parinos, C., Pasadakis, N., 2019. Large sea surface temperature, salinity, and productivity-preservation changes preceding the onset of the Messinian Salinity Crisis in the eastern Mediterranean Sea. Paleoceanogr. Paleoclimatol. 34, 182–202. https://doi.org/10.1029/2018PA003438.

Veit, E., Conrad, C.P., 2016. The impact of groundwater depletion on spatial variations in sea level change during the past century. Geophys. Res. Lett. 43, 3351– 3359. https://doi.org/10.1002/2016GL068118.

Versalovic, J., de Bruijn, F.J., Lupski, J.R., 1998. Repetitive sequence-based PCR (rep-PCR) DNA fingerprinting of bacterial genomes. In: Bacterial Genomes: Physical Structure and Analysis. Chapman and Hall, New York, 437–454.

Versalovic, J., Koeuth, T., Lupski, J.R., 1991. Distribution of repetitive DNA sequences in eubacteria and application to fingerprinting of bacterial genomes. Nucleic Acids Res. 19, 6823–6831. http://dx.doi.org/10.1093/nar/19.24.6823.

Voelker, A.H.L., Lebreiro, S.M., Schönfeld, J., Cacho, I., Erlenkeuser, H., Abrantes, F., 2006. Mediterranean outflow strengthening during northern hemisphere coolings: A salt source for the glacial Atlantic?. Earth Planet. Sci. Lett., 245 (1–2), 39– 55. https://doi.org/10.1016/j.epsl.2006.03.014.

Volk, T., Hoffert, M.I., 1985. Ocean carbon pumps: analysis of relative strengths and efficiencies in ocean-driven atmospheric CO2 changes. In: The carbon cycle and atmospheric CO: natural variations Archean to present. Chapman conference papers, 99–110. American Geophysical Union, Geophysical Monograph 32.

Von Breymann, M.T., Emeis, K.C., Suess, E., 1992. Water depth and diagenetic constraints on the use of barium as a paleoproductivity indicator. In: Upwelling Systems: Evolution since the Early Miocene. Geol. Soc. London Spec. Publ. 64, 273–284. https://doi.org/10.1144/GSL.SP.1992.064.01.18.

Von Damm, K., Edmond, J., Grant, B., Measures, C., Walden, B., Weiss, R., 1985. Chemistry of submarinehydrothermal solutions at 21°N, East Pacific Rise.Geochim. Cosmochim. Acta 49, 2197–2220. https://doi.org/10.1016/0016-7037(85)90222-4.

Wallace, A.F., 2019. Organic interfaces enhance strontium content of marine barite. Proc. Nat. Acad. Sci. 116 (27), 13161–13162. https://doi.org/10.1073/pnas.1908013116.

Wang, F., Polcher, J., 2019. Assessing the freshwater flux from the continents to the Mediterranean Sea. Sci Rep 9, 8024. https://doi.org/10.1038/s41598-019-44293-1.

Warning, B., Brumsack, H., 2000. Trace metal signatures of eastern Mediterranean sapropels. Palaeogeogr. Palaeoclimatol. Palaeoecol. 158, 293-309. https://doi.org/10.1016/S0031-0182(00)00055-9.

Wehausen, R., Brumsack, H.J., 1999. Cyclic variations in the chemical composition of eastern Mediterranean Pliocene sediments: a key for understanding sapropel formation. Mar. Geol. 153 (1–4), 161–176. https://doi.org/10.1016/S0025-3227(98)00083-8.

Weldeab, S., Emeis, K.C., Hemleben, C., Schmiedl, G., Schulz, H., 2003. Spatial productivity variations during formation of sapropels S5 and S6 in the Mediterranean Sea: evidence from Ba contents. Palaeogeogr. Palaeoclimatol. Palaeoecol. 191(2), 169–190. https://doi.org/10.1016/S0031-0182(02)00711-3.

Weldeab, S., Menke, V., Schmiedl, G., 2014. The pace of east African monsoon evolution during the Holocene. Geoph. Res. Let. 41, 1724–1732. https://doi.org/10.1002/2014GL059361.

Wilson, J.D., Arndt, S., 2017. Modeling radiocarbon constraints on the dilution of dissolved organic carbon in the deep ocean. Global Biogeoch. Cycles 31 (5), 775–786. https://doi.org/10.1002/2016GB005520.

Widanagamage, I., Waldron, A., Glamoclija, M., 2018. Controls on Barite Crystal Morphology during Abiotic Precipitation. Minerals 8, 480. 10.3390/min8110480.

Williams, P.J.L.B., 2014. Plankton respiration, net community production and the organic carbon cycle in the oceanic water columnA2—Holland. In: Treatise on

Geochemistry, 2nd ed., edited by K. K. Turekian, 593–612, Elsevier, Oxford, U. K.

Wolgemuth, K., Broecker, W.S., 1970. Barium in sea water. Earth Planet. Sci. Lett. 8 (5), 372-378. https://doi.org/10.1016/0012-821X(70)90110-X.

Wu, J., Böning, P., Pahnke, K., Tachikawa, K., De Lange, G.J., 2016. Unraveling North-African riverine and eolian contributions to central Mediterranean sediments during Holocene sapropel S1 formation. Quat. Sci Rev. 152, 31–48. https://doi.org/10.1016/j.quascirev.2016.09.029.

Wu, J., Filippidi, A., Davies, G.R., De Lange, G.J., 2018. Riverine supply to the eastern Mediterranean during last interglacial sapropel S5 formation: A basin-wide perspective. Chemi. Geol. 485, 74–89. https://doi.org/10.1016/j.chemgeo.2018.03.037.

Yacobi, Y.Z., Zohary, T., Kress, N., Hecht, A., Robarts, R.D., Waiser, M., Wood, A.M., Li, W.K.W., 1995. Chlorophyll distribution throughout the southeastern Mediterranean in relation to the physical structure of the water mass. J. Mar. Syst. 6 (3), 179-190. https://doi.org/10.1016/0924-7963(94)00028-A.

Yang, Y., Chen, S., Li, S., Chen, M., Chen, H., Liu, B., 2014. Bioleaching waste printed circuit boards by *Acidithiobacillus ferrooxidans* and its kinetics aspect. J. of Biotech. 173, 24–30. https://doi.org/10.1016/j.jbiotec.2014.01.008.

Yiğiterhan, O., Murray, J.W., 2008. Trace metal composition of particulate matter of the Danube River and Turkish rivers draining into the Black Sea. Mar. Chem. 111 (1–2), 63-76. https://doi.org/10.1016/j.marchem.2007.06.019.

Yogev, T., Rahav, E., Bar-Zeev, E., Man-Aharonovich, D., Stam-bler, N., Kress, N., Beja, O., Mulholland, M.R., Herut, B., Berman-Frank, I., 2011. Is dinitrogen fixation significant in the Levantine Basin, East Mediterranean Sea?. Environ. Microbiol. 13, 854–87.

Zektser, I.S., 2006. Groundwater as a Component of the Environment. In: Geology and Ecosystems. Springer, Boston, MA. https://doi.org/10.1007/0-387-29293-4_8.

Zhou, M., Liu, Y., Zeng, G., Li, X., Xu, W., Fan, T., 2007. Kinetic and equilibrium studies of Cr (VI) biosorption by dead *Bacillus licheniformis* biomass. World J. Microbiol. Biotech. 23 (1), 43–48. https://doi.org/10.1007/s11274-006-9191-8.

Zhou, Y.Q., Sawyer, A.H., David, C. H., Famiglietti, J.S., 2019. Fresh submarine groundwater discharge to the near-global coast. Geophysical Research Letters 46, 5855– 5863. https://doi.org/10.1029/2019GL082749

Zohary, T., Herut, B., Krom, M.D., Fauzi, R., Mantoura, C., Pitta, P., Psarra, S., Rassoulzadegan, F., Stambler, N., Tanaka, T., Thingstad, T.F., Woodward, E.M.S., 2005. P-limited bacteria but N and P co-limited phytoplankton in the Eastern Mediterranean—a microcosm experiment. Deep Sea Res. Part II: Topical Studies in Oceanogr. 52 (22–23), 3011-3023. https://doi.org/10.1016/j.dsr2.2005.08.011.

Zúñiga, D., Calafat, A., Sanchez-Vidal, A., Canals, M., Price, B., Heussner, S., Miserocchi, S., 2007. Particulate organic carbon budget in the open Algero-Balearic Basin (Western Mediterranean): Assessment from a one-year sediment trap experiment. Deep Sea Res. Part I: Oceanogr. Res. Papers 54 (9), 1530-1548. https://doi.org/10.1016/j.dsr.2007.06.001.

Zwiep, K.L., Hennekam, R., Donders, T.H., Van Helmond, N., De Lange, G.J., Sangiorgi, F., 2018. Marine productivity, water column processes and seafloor anoxia in relation to Nile discharge during sapropels S1 and S3. Quat. Sci. Rev. 200, 178–190. https://doi.org/10.1016/j.quascirev.2018.08.026.