

Humic compounds in marine ecosystems - Ecological importance in transitional Mediterranean zones

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Abstract. By using the general term ‘humic substances’ (HS), a vast category is implied of heterogeneous organic compounds that are naturally occurred, formed under certain circumstances, bearing yellow to black colour. There are distinguishable fractions of the (HS) categorized e.g. according to hydrophilicity. Humic acids (HA), comprise the soluble fraction of (HS) at high pH values whereas fulvic acids (FA) represent the soluble fraction of (HS) under all pH conditions. The humin represents the unclassified, non-soluble fraction of organic material. (HS) are separating into two well distinctive categories i.e. allochthonous & autochthonous indicating a different origin. (HS) affect bioavailability of transition metal ions including Fe, Mn, Cu, and Zn due to (HS) chelating properties and high affinity towards those metals. (HA) stimulate biodegradation and favors removal of various pollutants serving as bioremediation agents. Organic compounds leached from plastic debris into the aquatic phase, become part of the marine dissolved organic carbon (DOC) pool. Humic part characteristics are tightly bound to the anthropogenic impacts stressed over the shoreline and contribute to diagenetic processing of the marine seabed. The scope of the present essay was to outline the multi-functionality of humic substances in aquatic ecosystems with the given emphasis on the Eastern Mediterranean region.

1 Introduction

Dissolved Organic Matter (DOM) is an intrinsic, basic research frame in aquatic sciences, such as oceanology, limnology, and hydrology. Though the size limit used to categorize (DOM) is somewhat arbitrary, is globally accepted that 0.45µm porous filter cannot retain the dissolved part [1]. Human activities exert great stress to the coastal areas since considerable quantities of municipal and industrial wastewater (raw or treated) end up in the sea. Discharges occur nearby river estuaries, which result in a rapid transportation of terrigenous (DOM) into the sea [2,3]. As a result, (DOM), in the marine environment, gradually undergoes structural changes e.g. molecular structure & size, functional groups and chemical mobility. The combination of organic and inorganic pollutants affects (DOM) bioavailability (uptake part) by the aquatic microorganisms [4-5]. Photosynthetic aquatic organisms bind atmospheric CO₂ in algal cells and later on, via algal cells’ decomposition, it is released in the sea column which forms weak complexes with metal ions. It comprises the source of (DOC) pool, rich in labile polysaccharides. Microorganisms take advantage of (DOC) presence and advance with carbon fixation as a part of carbon cycle in seawater. Macroalgae determine both metal complexation and release of organic ligands. Their metabolites regulate organic matter bioavailability i.e. carbon fixation easiness [6].

Estuaries, riverine inputs, water mixing/transition zones and lagoons are the major areas where (DOM) is formed, controlled and distributed through biochemical mechanisms [4, 7]. (DOM) is mainly composed of humic substances, carbohydrates, and proteinaceous compounds [8-9]. (DOM) undergoes in both water column and sea sediment ongoing geochemical ‘*diagenetic*’ transformation alias ‘*diagenesis*’. (DOM) interacts with chemical pollution agents and enclose significant biogeochemical transformation pathways [7]. Coastal rivers discharge large quantities of terrestrial (DOM) and convey nutrients with very high primary productivity, which leads temporarily to ‘*allochthonous*’ i.e. terrestrial origin (DOM) increase [7, 10].

By using the general term humic substances (HS), a vast category is implied of heterogeneous organic compounds that are naturally occurred and formed under certain circumstances, bearing yellow to black colour. They are chemical formations of high molecular weight [11]. (HS) are the biggest reservoir of organic carbon in the aquatic and terrestrial environment [12]. (HS) represent one of the largest sources of natural organic carbon on earth, being the main component of the dissolved matter in surface and ocean waters and the natural bulk part of organic compounds in soil and peat [13-14].

Distinguishable fractions of (HS) are the ‘*hydrophobic*’ and ‘*hydrophilic*’ formations. Hydrophilic part is the dominant component of (HS) [15]. Seawater

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contains approximately 1 mg.L⁻¹ dissolved organic matter (DOM), most of which is uncharacterized compounds, primarily humic polymers [16-17]. The humic fraction in seawater dissolved organic matter represents 5-15% [18]. In lake water bodies (DOC) as part of the (DOM) amounts up to 80% of humic carbon [19].

2 Scope

The scope of the present manuscript was to make a brief bibliographical review of the multi-functionality of the humic part in aquatic ecosystems. Moreover, to emphasize the interaction of (DOM) with various metal-ligands via complexation, to trace habitats' bioremediation capacity and outline the critical role of complexation mechanisms, interrelated to (DOC) in the ecological status in Eastern Mediterranean Sea.

3 Methodology

A systematic literature review was accomplished over basic issues concerning the carbonaceous particle matter in the sea column. Photosynthetic micro and macro-organisms provide organic carbon in the water column. Metal-ligand complexation transform the bioavailable carbon in consumable cycles with highly biotic importance. Bibliographical references of significant estuaries in Adriatic Sea and extended Nile deltaic system lagoons outline aspects of transition zones which characterize vital habitats with tremendous scientific & ecological interest.

Organic carbon content in sediments was determined by applying combined wet oxidation processing, digestion and titration with ferrous ammonium sulphate. Humic substances in sediments were extracted by means of acidification by applying well established lab protocols. (HA)s were extracted from sediments by applying international humic substance society methods properly modified based on solubility and pH regulation. Organic carbon origin was detected and determined by applying isotopic ratio ($\delta^{13}\text{C}$) analysis by means of the use of isotope ratio mass spectrometer coupled with a CHNSO analyzer. Fulvic acid (FA) was isolated by extracting sediments with alkaline solution, centrifugation, flocculation of mineral colloids and acidification. Later purified by using cation exchange resins and as a final stage undergo lyophilization. (DOM) was determined by employing Fourier transform infrared spectroscopy (FTIR) and samples obtained from leaching experiments. (DOC) concentrations were determined by using a Total Organic Carbon (TOC) analyzer. Different size fractions of microplastics' (DOM) samples were quantified by combining high-performance liquid chromatography systems equipped with organic carbon detectors along with 0.45 mm pore size filtering [2, 4].

4 Marine humic compounds to aquatic environment – Formation mechanism

(HS) regulate solubility, mobility, transport, concentration and accumulation of trace elements in marine environments [20]. They demonstrate great complexing ability, attributed mainly to intramolecular oxygen-based ligands, (e.g. carboxylic, phenolic and carbonyl groups), [21]. The interaction of humic matter with different metal ions changes the biogeochemical cycle of trace metals and affects their bioavailability [22]. (HS) are the structural outcome of humification processing of biotic origin. Two basic formation origins are to be distinguished as regards (DOM), as a whole, when marine sediments are to be examined. That is, the organic matter (OM) derived from aquatic, marine living organisms i.e. mainly phytoplankton, 'in situ', bacterial cells and debris production, formally named as '*autochthonous*' part and (OM) initially terrigenous, later transferred via terrestrial watersheds, defined as '*allochthonous*' [23-24]. (HS) demonstrate remarkable heterogeneity and polydispersity, as regards the molecular structure, the formation mechanism of which is not up to now fully understood in the scientific community. (HS) are consisted of aliphatic chains and aromatic rings bound with oxygen, nitrogen and sulfur ligands [25-26].

A basic categorization of (HS) is determined according to their solubility characteristics. Thus, (HS) occurred in marine sediments incorporate a great number of hydrophilic functional groups, enhancing the solubility in aquatic environment at high both pH and ionic strength values [27]. Therefore, the soluble part of (HS) denotes the Humic Acids (HA) when exposed at high pH value conditions, whereas fulvic acids (FA) is the most soluble part under any pH value and 'humins' consists the non-soluble part of the initial bound formation which cannot be affected under any pH value even in alkali. It is apparent that (HS) soluble forms are easily extracted and thus extensively, experimentally studied. Both afore mentioned extracted parts i.e. (HA) and (FA) are subjected to noticeable heterogeneity in the resulted mixtures which renders selective extraction by using chemical agents problematic [28].

Certain scientific publications outline striking differences regarding (HS) bulk molecular characteristics, between soil and marine sediments [29]. Therefore, lignin's phenols have been identified as major part of terrestrial humic acids [30-31], whereas marine derived humic acids are less aromatic and have higher carbohydrate and protein content [32]. Although lignin polymers are the basic source of terrestrial humic substances, yet carbohydrates are more abundant in marine environment. In general, allochthonous (DOM) sources increase respiration within marine ecosystems up to a level that exceeds photosynthetic carbon uptake by phytoplankton and aquatic macrophytes [33]. It incurs gradually a shift in marine (DOM) characteristics, turning from terrestrial to autochthonous due to aquatic biota metabolism (e.g. excretion, biogenic debris), phytoplankton and other photosynthetic organisms' primary production [34-35]. Thus, autochthonous sources contribute to the ongoing (DOM) transformation, resulting in the increase of chromophoric (DOM) (i.e. amino acids, humic acids and aromatic proteins) turning the water colour to black (or brown) accordingly, (see Fig.

1), in the water transition area [36]. In **Table 1** is illustrated (DOM) classification for different pore size filtration. The origin of humic substances in soil, lakes, and seawater and the role of melanoidins in humification, have been discussed thoroughly in several reviews [37-39]. The interaction of humic matter with different trace metal ions affects the biogeochemical cycle of these trace metals, and consequently their bioavailability [22].

Table 1. Attributes of various (DOM) classes derived from different pore size filter (modified [39])

	DOM-I	DOM-II	DOM-III
Pore size (µm)	<0.2	0.2-6.0	>6.0
Water 'type'	cohesive/adhesive	cohesive	gravitational/ cohesive
Water tension (kPa)	<1,500	1,500 to 50	>50
(%) of water at WHC ^(a)	~30	~50	~20
Transport mechanism	diffusion	diffusion> convection	convection> diffusion
Metabolism	abiotic, exoenzymes	microbial	biotic
Relative turn over	slow	moderate	rapid
Effect of drought ^(b)	weak	moderate	strong

^(a) WHC is the water holding capacity, approximate values
^(b) The row does not apply to water saturated systems

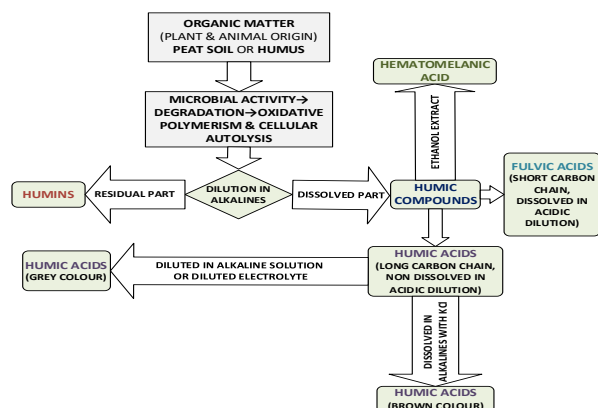


Figure 1. Organic matter dilution/extractation in alkalines and acidic condition (modified [40]).

4.1 Complexation of DOM with Al & Si

The interaction between (Si) and organic matter (OM) was considered in the past as non-existent or very weak. Later on, fluorescence experiments confirmed the existence of weak (HA-Si) interactions. However, through several complementary techniques, it was proved that stronger (HA-Si) interactions exist in a chain scheme with the presence of an intermediate ion such as (Al³⁺) which contributes to the formation of a ternary complex (HA-Al-Si) and the bridging of (Al) in a (Organic Matter-Al-Si) bond. (DOM-Al-Si) complexes play a vital role in 'pedogenesis' wherever dissolved organic matter (DOM) is abundant, in soils that podzolic processes occur.

The presence of silicon (Si) significantly promotes the maintenance of both (Al) and (DOM) in solution, which

enhances the modification or even the kinetics of the processes of dissolution/precipitation of minerals, thereof pedogenesis' outcome. The importance of (DOM-Al-Si) complexes in the dynamics of the systems will however have to be clarified further [41].

4.2 Complexation of (HS) with metal ions

(HS) regulates the bioavailability of transition metal ions including (Fe), (Mn), (Cu), and (Zn), due to their chelating properties and high chemical affinity [42]. However, the impact of those properties has been underestimated, especially in case of iron. Iron is essential for all living beings and its poor bioavailability limits biota potential growth and affects their physiology. Moreover, (HS) affects plants' photosynthesis and the formation of chlorophyll and carbohydrates [43-45]. The bioavailability of iron in soils is strongly regulated by the pH value and oxygen content. At neutral pH and under aerobic conditions iron occurs mainly as iron oxide. The general term iron oxide comprises various chemical forms such as ferric oxyhydroxide FeO(OH) and amorphous iron hydroxide Fe(OH)₃. The latter is poorly soluble in water and therefore cannot be taken up by plants and microorganisms.

In cases that the oxygen content in soil is low, trivalent iron (III) can be reduced to the far more accessible and soluble bivalent iron (II) [46]. More problematic tends to be iron bioavailability in aquatic systems. Most of aquatic systems are highly oxygenated, at least in regions of significant photosynthetic activity. Due to high dissolved oxygen (DO) values combined with the low solubility of inorganic trivalent iron (III) salts, iron is often the limited growth factor for the productivity of aquatic primary producers [47]. Furthermore, seawater high ionic strength and pH value (pH ~8.2) contribute to rapid precipitation of iron [48-50]. As a result, marine iron cycle is linked to the carbon cycle via primary production including algae uptake mechanisms. It is therefore, very important to be cleared out the limitation pathways and environmental aspects of iron bioavailability. The analysis of the dissolved iron in seawater revealed that 99.99% of it is engaged in organic ligand structures [51-52]. Humic acids (HA) are natural chelators and important factors for iron supply to marine organisms [53].

Several years before, the impact of humic substances on iron bioavailability was underestimated due to the assumption that most of the organic material precipitates in salt water conditions [54-55]. Nonetheless, the assumption turned out to be incorrect since approximately 20% of aquatic humic substances resist precipitation and remain as dissolved part in seawater body [56-58]. Humic acid-based iron complexes have been located practically in all open seas. Moreover, (HS) originate from diverse plants and animal residues and thus are very complex and demonstrate high structural variety [58]. In general, they are divided into three groups according to their size, properties and solubility [59]. The group of humic substances with a good aqueous solubility (fulvic acids) is of great interest regarding iron complexation and transport.

Humic acid - iron complexes consist of rather small molecules with a molecular mass of about 0.5–2.0 kDa and relatively high oxygen content [60]. In a nutshell, it is crucial to be cleared out iron binding mechanisms and releases from those complexes and the role of functional groups which determine the iron uptake by microorganisms.

4.3 Bioremediation by means of (HA) in aquatic ecosystems

Dissolved organic matter (DOM), such as humic or fulvic acids, generates under solar radiation, highly reactive chemical forms capable to oxidize pollutant agents [61].

Humic acids (HA) are complex structured polyelectrolyte compounds and constitute between 50 and 75% of dissolved organic matter in the natural environment regulating mechanisms in aquatic ecosystems [62]. (HA) enhance the rate of removal of various pollutants by stimulating biodegradation along with the removal of various chemical agents such as congo red [63], polycyclic aromatic hydrocarbons (PAH) [64], photodegradation of synthetic hormone (EE2) [65], reduction of the removal rate of glyphosate and acephate [66-67], pharmaceuticals [68], and Bisphenol A (BPA), an agent used in plastics' manufacturing [69]. Other macromolecules with similar characteristics (humic-like substances, HLS) have been demonstrated to be useful for the same purposes [70].

4.4 Humic – like substances

A roughly 30% of the marine (DOC) pool is composed of labile polysaccharides, some of which form weak complexes with iron (Fe). A further decomposition of cell-derived debris contributes to a longer-lived (DOC) pool known as humic-like substances, which compose around 50% of DOC, a smaller fraction of which (around 5% of DOC) can be traced bound to iron, forming siderophore complexes. Humic ligands form iron complexes of intermediate strength. Iron complexation in estuarine and coastal waters dominated by means of humic-like material, mostly of terrestrial origin. The stability of iron-humic complexes allows iron to be transported long distances by ocean currents [71].

However, despite many indications that humic ligands regulate to a certain degree the iron cycle, there have been difficulties to provide sufficient explanation of the interconnections among humic substances i.e. iron and iron ligand pool in ocean water. Humics comprise a heterogeneous mixture of soluble and colloidal-sized compounds of various origins and a great variety of iron-binding pathways [71].

Marine (DOM) is mainly derived from photosynthetic production by phytoplankton, whose organic matter is mainly composed of labile biomolecules such as carbohydrates, proteins and lipids. However, uncharacterized and bio-refractory organic matter (i.e. organic compounds which resist decomposition by ordinary biological mechanisms) accounts for a major part of oceanic (DOM) [40].

Bio-refractory organic matter provoked in the past vivid interest in the scientific community regarding 'diagenesis' geoprocessing and transformation of phytoplankton organic matter into bio-refractory (DOM). There are numerous publications during the past few decades over that topic [72-74].

Marine humic substances (HS) are known to be one of the main components of bio-refractory (DOM) [75]. Since they have a specific fluorescent property, their distribution and dynamics have recently been acknowledged as marine humic-like fluorescent (DOM).

4.5 (DOC) & plastic material interaction

Plastics due to their durability and cost-effective manufacturing, has been accumulated in the aquatic environment. Plastic debris affects negatively marine macro- and micro-organisms in marine ecosystems [76]. High production polymers in the plastic industry are expected to be the most abundant in the ocean i.e. low- and high-density polyethylene (LDPE/HDPE), polypropylene (PP) and polystyrene (PS), which account for ~70 % of the total plastic debris encountered [77]. They are petroleum-based, of high molecular weight, with degrading time periods of hundreds of years.

Starch-based plastics and starch blends, such as starch-polyethylene blends or polylactic acid (PLA) are promising alternatives of petroleum-based plastics, with shorter degradation periods and less harmful environmental impact [78]. However, the biodegradability of these plastics is controversial since many of them need high temperatures to be degraded, far from the conditions occurred in the ocean [79].

Furthermore, synthetic polymers produced by the plastic industry are accompanied by a variety of chemical additives, such as plasticizers, flame retardants (inorganic fillers), metals, antioxidants and other stabilizers, pro-oxidants, surfactants and pigments [80]. All these additives are part of the plastic manufacture processing in order to improve plastic's quality and performance [81]. Many of these additives are not covalently bound to the plastic polymer [82] and can be eventually migrated from plastics to the seawater column i.e. 'leached' into the surrounding water and enter aquatic organism's body during the plastic degradation and fate process [76]. Leaching is enhanced by solar (UV) radiation along with physical abrasion of wave kinetic energy forces which lead to fragmentation of the plastic material into small debris [4, 83-85]. Plastics can also be degraded biotically i.e. by microorganisms [82]. Then the structure of the polymers weakens leading to cracks, roughness and molecular changes.

Organic compounds leached from plastics become part of the marine dissolved organic carbon (DOC) pool [86]. The basic source of natural pool of carbon is phytoplankton's primary production. The main decrease occurs during heterotrophic respiration, when most of the (DOC) is being decomposed by bacterioplankton (i.e. bacterial component of the drifting plankton) in the ocean surface [87]. Part of the (DOC) released from plastics follow the same fate along with the freshly produced

natural (DOC) since it can be taken up by heterotrophic bacteria stimulating their growth [84-86].

5 Humic matter in the Eastern Mediterranean Sea - Discussion

Certain researches were conducted in the Eastern Mediterranean Basin, mostly in transitional zones e.g. estuaries, in lagoons nearby the coastline and shallow formation with saline waters. The northern Adriatic Sea pelagic ecosystem reveals certain peculiarities. It considered to be a critical productive area of the Mediterranean aquatic basin.

Ravenna lagoon (see **Fig. 2**) sediments in Adriatic Sea is a place of vivid scientific interest on account of the interactions of anthropogenic pollutants e.g. (PAHs) and the intrinsic Humic Organic Matter abbr. (HOM) in the sediments of the lagoon. Thus, high abundance of (PAHs), some of which were very uncommon, were expected to pose a significant environmental threat to lagoon bottom feeders, such as benthic organisms, with implications in aquatic life. Emphasis was laid to monitor the mobilization of hydrophobic, immobile (PAHs) in dissolved humic organic matter with terrestrial origin, anthropogenic characteristics via carbon isotopic ratios [88-89].

(PAH) molecular structure obtained by pyrolysis' methodology [89], appeared to be significantly different from the (PAH) structure obtained from the solvent extracts in the lab. Heavy road traffic justifies the presence of fossil fuel remnants i.e. pyrolysis derived (PAHs). Though findings provided convinced that molecular structures covalently bound inside (HOM) remained chiefly unaffected by contamination, still was evidenced, that the very uncommon (PAH) structures obtained by pyrolysis, pointed out the formation of bound residues based on activated (PAHs) as a detoxification pathway. Furthermore, the results obtained from the analyses of molecular markers provide indications that methanotrophic and sulfate reducing bacteria were involved in biochemical transformations of the pollutants. Humification pathways were traced along with the formation of bound residues and anthropogenic sources of (HOM) [89-90]. Considerable quantities of nutrients are ending up into the sea from rivers the most significant of which is the river Po and the estuarine formed, not far from Ravenna lagoon (see **Fig. 2**) in Adriatic Sea. Transferred organic matter affects the local ecosystem leaning towards eutrophication phenomena, affecting all sea water quality characteristics. Sea bed sediments' status is the proper material for scientists to apply a well based methodology upon assessing the local environmental quality and measuring anthropogenic exerted pressure.

Apart from e.g. sediments resuspension and bioturbation, the presence of strong chemical gradients i.e. redox, pH, salinity and organic matter concentration, along with humic substances (HS) in organic matter hold a key role in the accumulation and bio-availability of pollutants, regarding mostly metals bound in sediments [91].

Since the significance of river Po runoff was early acknowledged for the sake of local ecosystems' quality status assessment, a strategy was established, via steadily programmed sampling and monitoring by means of measuring certain ecological parameters. The scope was the estimation of ecological impact of Po river's plume transferring fresh inland surface water. Practical Salinity Unit (PSU) gradient, Dissolved Oxygen (DO) concentration, Redox potential, Total Phosphorus (TP), Total Organic Carbon (TOC), Humic Acids and Fulvic Acids content, influence the biogeochemical cycle of the organic matter. Furthermore, remarkable sediment accumulation takes place on yearly basis. The final goal was the redox state of sediments to be investigated in area zones under the influence of both fresh and treated waste water discharges. (HS) derivations i.e. humic acids (HA) & fulvic acids (FA), and non-soluble part in alkaline environment (humins), were the major organic constituents of sediments. Both, (HA) and (FA) incorporate polar and non-polar substituents, thus they are able to bind with hydrophobic and hydrophilic ligands simultaneously. More importantly, they performed as stimulatory agents for marine phytoplankton growth by acting as electron donors in photo-oxidation reactions in marine environments.

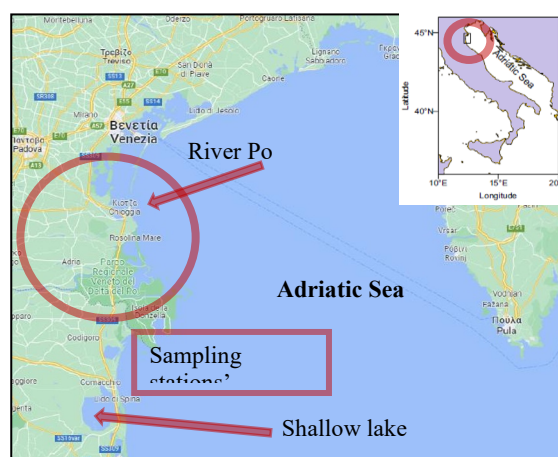


Figure 2. Ravenna lagoon and stations in a sampling zone in proximity to the coastline bounded by Po River estuarine and Reno River runoff [91].

The presence of humic acids in coastal sediments indicates an early stage of 'diagenesis' processing. High values of (TOC), (TP) and (HS) measured from sampling stations (see **Fig. 2**), indicate an anthropogenic input, which may be ascribed to the river Po in particular [91]. Data acquired concerning (HS) structures denote that they are largely autochthonous. River Po can act both as labile organic matter supplier leading to the formation of humic compound and as carrier of humified compounds. Moreover, humic and fulvic acid concentrations were inversely correlated to the distance from the coast and directly correlated to the (TOC) and (TP) content [91].

Later on, another scientific research [92], quantified (HA) contribution to sedimentary (OM) and investigated (HA) origin in a shallow continental plateau influenced by Po's riverine runoffs. Organic compounds' ratio was estimated which characterize the degree of the ongoing

humification. As a result, the (HA)-carbon extracted from the surface and subsurface sediments constitute a relevant fraction (17%) of the total organic matter sedimentary. Sediment sampling located between the Po di Goro and the Ravenna harbour in central Italy in depth ranging from 4 to 18 m.

The sampling conducted at grain size analyses and certain physico-biochemical parameters i.e. pH, oxidative-reductive potential (Eh), organic carbon (C_{org}) and total nitrogen (N) were determined in both surface and subsurface sediments. (HA) was measured as well, from sediments extracted by combining the use of a CHNS-O Elemental Analyzer and a specific methodology with carbon isotopes. The scope was to assure the overall impact of Po and Reno rivers on the sea area.

The negative relationship between HA content and redox potential in the sub-surface sediments, which were expected to be in a reduced state, indicated that organic compounds are functioning as electron acceptors for microbial respiration and take part in the ongoing mineralization process in anaerobic environments.

'Diagenesis' in sediments favors specific organic intramolecular structure and a preferential degradation of the more reactive fraction of humic compounds along with an increase of C_{org}/N ratio in the HA. Chemical characteristics support the hypothesis of their mixed origin: riverine & continental source, probably derived from lignin compounds and marine input derived from degradation products of plankton.

Dissolved humic substances (HS) reveal critical ecological balance mechanisms in sensitive habitats such as the shallow Lake Edku in Egypt (Fig. 3). It's a coastal lagoon of the Nile delta in northern Egypt. The lake covers an area of 115 km² and has an average depth of 1.0 m (ranges from 0.40 to 1.80 m).



Figure 3. Lake Edku, a shallow coastal lagoon of Nile estuarine in northern Egypt.

Monitoring of the ecological status of the lake entails measuring of certain parameters to verify and understand the origin, distribution and fate of (HS), the seasonal variability and habitat interactions. Therefore, the shallow lake was subdivided into three individual basins to facilitate the scope of investigation [93].

Humic substances were extracted from sediments of lake Edku taken from all subdivisions, and fractionated to their humic and fulvic acid constituents according to their physicochemical characteristics regarding optical density, solubility, chemical composition incl. elemental composition, molecular functional groups etc.

Fulvic acids were characterized by their lower carbon and higher oxygen content. C/H and C/O ratios were lower in fulvic acids although C/N ratio was comparable in both components. Fulvic acids were also more acidic due to a higher carboxyl group content which contributed

to the total acidity. The geographical distribution of the physical and chemical characteristics did not reveal distinct and systematic features that could be related to the origin of the material. This was mainly attributed to the predominance of the internally produced humic substances over the terrigenous derived material.

Lake Edku dominant source of in-situ-produced organic matter is the macrophytes; these aquatic plants probably have an organic composition which does not differ significantly from the organic matter derived from arable soil. Moreover, both land-derived and in-situ-produced organic matter by macrophytes are exposed to microbial transformation activity.

Drainage water transfers tonnes of particulate organic carbon per year into the lake. The annual input (in tonnes) of dissolved humic substances was estimated [93], part of which, about 65%, is recycled or trapped (HS) inside the lake system. The second source is the primary production of the lake. According to other researchers [94-95], 10% of the carbon from primary productivity is transferred to humic dissolved organic carbon. Furthermore, it was estimated [96] that about 50% of the organic matter produced in lakes of high sedimentation rate are recycled in the lake. A great amount of carbon derived from the primary productivity is accumulating annually as organic debris in the lake bottom sediment. Thus, it is concluded that more than 85% of the sedimentary humic substances are of internal origin.

Some particular geographical variations could easily be related to the deposition occurred with the expected influence on the humification processes [97]. For example, humic acid sampling from the middle and eastern parts has higher total acidity, carboxyl groups and carboxyl content per unit carbon, and lower C/H and C/N ratios than the rest of the lake. Fulvic acid showed the same trend except for C/N. Sediments of these stations were the highest in mud content (~80%). Moreover, sedimentation rates are expected to be high due to the proliferation of macrophytes. These sustained conditions would favour the development of a poorly oxygenated or even anoxic environment which retards (HS) transformation processes [98].

Humic substances are categorized by their chemical interactions. It has been shown [12] that humic acids of marine sediments are stronger complexing substances for certain trace metals compared to terrestrial-derived ones. Furthermore, the results of the elemental analysis showed that humic acids have a higher carbon and lower oxygen content than fulvic acids which are in accordance with the results obtained by other researchers [99]. In general, (HA) demonstrate sulphur content within the range (0–2%) [100], whereas, sulphur content in fulvic acids is remarkably higher [101-102]. Several arguments explain the high sulphur content in the humic material, the most solid argument of which is the prevailing of reducing conditions during deposition [102]. Elemental sulphur and hydrogen sulphides can be incorporated into the organic molecules.

It is widely accepted that higher phenolic, aromatic and lignin entities are more abundant and characterize (HS) of terrestrial origin, whereas (HS) in the marine environment (planktonic origin & algal-derived material)

are mostly aliphatic and have a higher N/C and H/C atomic ratios due to their higher protein and aliphatic content [102].

6 CONCLUSION

Humic substances (HS) incorporate a vast category of heterogeneous organic compounds that are naturally occurred. There are distinguishable regarding the degree of hydrophilicity they present. Humic acids (HA), comprises the soluble fraction of (HS) at high pH values whereas the fulvic acids (FA), denote the soluble fraction of (HS) under all pH conditions. The humin, represents the non-soluble fraction of (HS).

They are holding a protagonist role in aquatic and marine ecosystems with the emphasis on biota. (HS) are separated into two well distinctive categories i.e. allochthonous & autochthonous indicating a different origin. (HS) regulates the mechanisms for the bioavailability of transition metal ions including Fe, Mn, Cu, and Zn due to their chelating properties and high affinity towards those metals. (HA) stimulate biodegradation and removal of various pollutants functioning as bioremediation agents. The organic compounds leached from plastic debris become part of the marine dissolved organic carbon (DOC) pool.

The most important products formed during the composition of organic substances are the humic acids. Therefore, the resulting biopolymers are expected to demonstrate characteristics which are more or less related to the source component parts. The distribution of humic and fulvic acids in the bulk humic substance may vary according to the source material. (C), (H), (O), (N) constituents and their ratios are characteristics tightly bound to geographical and anthropogenic impacts and contribute to marine diagenetic processing.

Scientific research outcome in Ravenna Lagoon in central Italian coastline, in Po river estuarine in Adriatic Sea and in Lake Edku (Egypt), a coastal lagoon located in Nile estuarine, confirms the multi-functionality of humic substances in aquatic ecosystems and outline the necessity of the investigation of qualitative characteristics of (HS) to draw conclusions about the ecological status and integrated management of sensible habitats in Eastern Mediterranean coastline.

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