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HUMIC ACID: COMPREHENSIVE REVIEW OF PRODUCTION, STRUCTURE, CHARACTERIZATION AND APPLICATIONS

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Abstract

Humic acid represents an intricate heterogeneous compound of organic compounds of large molecular weight, which are synthesized in protracted soil throughout the humification of plant and animal deposits. This actuarial review in basic to applied approach considers humic acid, definition, sources, production process, structure properties, analytical estimation, physicochemical properties with multifunctional his use in agricultural, industrial and environmental areas etc. higher peat, soil organic materials, leonardite and lignite (80-50%) in ancient deposite of coal. The processes involved in the production consist of the usual alkaline extraction through the natural beds and the recent production processes based on biotechnology which involves a fungal fermentation. Molecularly, the chemical structure of the HAs is an aromatic and aliphatic backbone with functional groups of carboxylic, phenolic, quinone and methoxy functional groups and provides them with chemical characteristics of novel chelating, buffering, and bioactivity. The characterization is conducted analytically with the aid of a series of methodologies such as elemental, UV-visible (E4/E6-ratio), FTIR, NMR, and colorimetric. Part of this is in the form of molecular weights between the 1,700- 6,000Da, amphiphilic as well as pH buffering and metal binding capabilities. Applications Uses Agricultural soil amendments, plant growth bidder, industrial applications- batteries, ceramics, environmental cleanup and cosmetic supplements/ food supplements. The possible effects of limitations to the environment are by affecting the quality of water, cost, regulatory, as well as lack of full knowledge of the ecological effects in the long run. The review gives an overview of the available knowledge covering science and technology of humic acid with of primary concerns of its enhanced sustainable use in the varied fields through highlighting of opportunities and challenges.

Keywords: *Humic acid, Humification, Organic matter, Alkaline extraction, Molecular characterization, Agricultural applications.*

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I. INTRODUCTION

1.1 What is Humic Acid

Humic acid is a group of thick organic molecules developed as a result of decomposed dead biomass which numbers concurrently because of biochemical and ligation-exchange reactions. Such is matter that is common, or, at any rate, common physically - stacking up in most soils, in water-logged tundras, stratified layers of lignity, and in a more minor degree in the meromictic profile of rivers and in the meromictic profile of saltwater.

1.2 Definition and Origin

One of the large molecules in the humic substances are the humic acid, which has been formed as a result of the decaying of organic patience, which are long overdue. In thousands of millions of years that chemical change with the assistance of microorganisms of the plant and animal remains which is worth gallons per acre is totally covarying the quality of that organic substance in the humic acid. Get it, though, anywhere there is decay still on progress, but the richest sources are old peat, the carbon stake of humus formations of lignites and some brownish to purplish layers sold as Leonardites. In addition to these concentratitons humic acid is softly diffused in the mineral soils, in the mysticism of such fresh waters and in the depths of the ocean that so shines. If not, the data integrity might be kept at a high level [1,2,3].

1.3 Structure and Properties

The compound of the humic acid is an oversized and heavyweight composed organic polymer, which lacks a precise and unchanging structuring by the climate, age, and procedures that lead it to its creation. It has a mosaic pattern of functional groups, most conspicuously, quinones, phenols and carboxylic acids which endow it not only with a fondness to water, but also with a dislike of it - to put it technically, then, it is amphiphilic. Consequently, it readily solubilizes in medium-alkaline solutions, dissolves to a restricted extent in the more unworried pH of water, and precipitates in highly acidic conditions, in which the protonation of the carboxylic acids is effectively masking the acidic areas. This shape provides humic acid with an extraordinary adsorption capabilities capacity in addition to its capability to form secure chelates with metals as well as self-assemble into dispersed and micelle-like systems that are able to sequester within their interior a range of lesser solute associates.

1.4 Functions and Applications

Among other effects, humic acid is critical towards soil health such as helping in increasing soil structure, soil retention, nitrogen availability, and aiding in the nourishment of soil microbes. It can also be viewed as the anti-inflammatory, detoxifying and metal- Chelating agent tends to have an anti-oxidant effect and be used in agriculture, cleaning up pollution, medicine and in the synthesis of green catalysts during chemical synthesis [2,3,5].

Humic acid is an extraordinarily universal product which is utilized in soil health, plant farming, environmental fixing and industry aspects.

2. Multifaceted Applications of Humic Acid in Agriculture, Environmental Remediation, and Industry

One of such principals is humic acid (HA) that enhances water infiltration, cation exchange capacity, and soil structure and nutrient availability significantly. It enhances the physical, chemical and biological qualities of soil, existence microbes, recycling of other nutrients etc. HA as a biostimulant has also been reported in the promoting elongation of roots and shoots, productive rate and adaption to stress conditions through recovery of the hormone (auxin, cytokinin and gibberellin) pathways and expression of stress genes in plants [4,7,8]. Besides that, HA fosters the release of organic matter in the roots and disrupts the rhizosphere chemistry and micrograms [9].

2.1 Environmental and Tox Remediation Position.

HA is high molecular weight heterogeneous compound which is an excellent natural adsorbent that can bind heavy metals, organic contaminant and toxins through electrostatic, hydrophobic and hydrogen bonding. Due to this property, the chelating is applied in the soil and water remediation to eliminate the toxicity and bioavailability of pollutants. More so, HA contributes to changing and sequestration of such pollutants as polycyclic aromatic hydrocarbons, phenols, and heavy metals when used in combination or synergetically with microbial biodegradation [10,11].

2.3 Industrial and Health Care Applications.

HA is chelating, anti-inflammatory and antioxidant which is why it is in use in the pharmaceutical field, medicine and animal feed. Additionally, HA (hundunlingschroeder (2000)) also finds application in anaerobic digestion processes so as to develop the process of methanogenesis and to improve the production of hydrogen by acting as an electron shuttling product as well as other manipulations over the populations of the microbes. To tackle this issue, it is necessary to employ more affordable and dependable electric vehicles [12,15].

3. SOURCE OF HUMIC ACID

Table 3: Humic acid is mainly formed from plant and animal remains from soils, peat, lignite (brown coal), compost and organic wastes generally.

Source Type	Description & Examples	Reference
Soil and Peat	Formed naturally through the humification of plant and animal residues in soils and peatlands.	[3,4,5]
Lignite and Coal	Extracted from low-rank coals (lignite, leonardite) via alkaline extraction; widely used in agriculture and industry.	[5,20]
Compost and Manures	Produced during composting, vermicomposting, and from farmyard or sheep manure; can be extracted for agricultural use.	[43]
Agricultural Residues	Crop residues, cow dung, rice husks, and landfill compost are effective sources, especially for soil amendment.	[13,38]
Aquatic and Algal Matter	Found in natural waters, sediments, and can be extracted from decaying plant/algal material.	[3,4]
Artificial/Synthetic	Produced by controlled oxidation or hydrothermal treatment of biomass, food waste, or coal.	[20,38]

4. EXTRACTION AND VARIABILITY

Humic acids can be readily obtained by extracting (e.g., alkaline extraction using KOH or NaOH) or extracting by oxidizative conditions (artificially processed treated oxidized coal) or by hydrothermal conditions (artificially processed hydrolyzed biomass). Humic acids are commonly derived as food waste or Biomass artificially under oxidative conditions or artificially under hydrothermal conditions. The extraction method or source is important to determine its structure and use either in agriculture, environmental use or industrial use.

4.1 Percentage of Humic Acid

Average Percentage of Humic Acid in commerce and agricultural situations.

Humic acid can vary significantly in different roots of different origin, in its extraction method, and its use. The commercial products of humic acid are mostly 30 to 65 percent of humic acid and in agriculture, the usage is in solutions ranging at 1 to 3 percent.

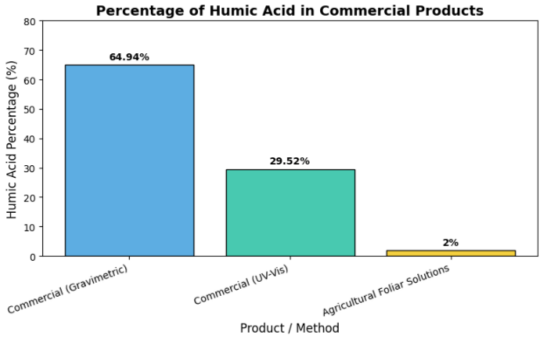

Fig 1: Percentage of Humic Acid in Commercial Products. Comparative analysis of gravimetric method and UV-Vis spectrophotometric showed that commercial humic acids contained 64.94% (gravimetric) and 29.52% (UV-Vis) humic acid with differences occurring depending on the analysis methodology and origin of products

Agricultural Application Rates

Foliar application studies on crops such as pomegranate trees commonly use humic acid concentrations of 1%, 2%, and 3% to improve yield and fruit quality these lower percentages are typical for direct application to plants, not for the raw or commercial product [19].

5. PRODUCTION OF HUMIC ACID

Both natural and inorganic humic acid (HA) is produced by natural and artificial/engineered processes to transform organic material into stable compounds containing high molecular weight of humic acids. Naturally, the process of humification involves the production of humic acid because the remains of plants and any other microorganisms is exposed to biochemical degradation and polymerization during aerobic or anaerobic conditions over a long period of time. However, it is primarily produced on top of various carbonaceous resources such as lignite, leonardite, peat, brown coal, compost and organic wastes in the extraction and synthesis of HA in the industry and agricultural sector [21,53].

The classic form of production method involves the utilization of solution of sodium hydroxide (NaOH) or potassium hydroxide (KOH) that is employed in extracting the humic acid in the fulvic and soluble fraction and then the extract is made acidic using the hydrochloric or sulfuric vacuum [22]. Speed of this process is conditioned by extent of oxidation of the raw material, the temperature, length of extraction and concentration of alkali utilized. The product of high levels of oxidation such as leonardite results in the production of leveraged humic acid which is approximately 40-90 percent however peat or less developed organic based materials will have a relatively low production [53].

During the recent years, the efficiency of extraction, the environmental sustainability has been improved by the scientists who employ more advanced technologies of processing. These are ultrasound-assisted extraction (UAE), microwave-assisted extraction (MAE), and hydrodynamic cavitation- assisted extraction (HCAE) which enhance the role of disruption of the cell wall, solvent penetration and reduction in time and chemical used 51. Indicatively, at optimal conditions (300 mW cm 2/90 min) of ultrasound-assisted peat extraction, one was able to get a higher amount of HA compared to regular batch extraction. Similarly, oxidative pretreatment with hydrogen peroxide (H_2O_2), nitric acid (HNO_3) or Ozone was left to depolymerize the complex organic matrices and increase the solubility and yield of HA, was also demonstrated in the same turn [53].

Violent to humic acid production, other than by extract, biological and synthetic techniques of fermentation sandwich are also being paid attention to. The application of microbial consortia; Bacillus, ligninolytic fungi, and Streptomyces can be used to induce biological humification to catalyze the oxidation polymerization of lignin precursors and other aromatic compounds to form humic like substances [53]. A biological humification analogy can then be provided by composting as well as vermicomposting of organic residues to produce humic like substances with similar properties as HA, which is a naturally existing substance. On the other hand, a different approach to artificial humification that has been boasted of is hydrothermal carbonization (HTC). It converts farm waste or lignocellulosic bi-mass into the humic acid type products upon less than critical temperature of water (180-250 degC) [52]. the production of HTC in two stages, the history incorporates the corn stalk hydrochar and the production yield is humic acid approximately 68 percent thus making it an efficient alternative source of production and is also friendly to the environment.

An integrated approach toward sustainable production of humic acids by making integrated chemical-biological systems and working with renewable feedstock is also there. In addition to enhancing yield and purity, these inventions will minimize the reliance on fossil needs, e.g., lignite and coal. As the requirements of humic acid in the world increase with its application in agriculture, bioremediation as well as in industry, the contemporary manufacturing methods continue to shift towards cost saving and eco-friendly technological procedures [51,52,53].

Main Methods for Humic Acid Production

Table 2: Humic acid production methods and innovations

Method	Raw Materials	Key Features/Process	Yield/Advantages	Reference
Alkaline Extraction	Lignite, leonardite, peat	Extraction with NaOH/KOH, acid precipitation; efficiency depends on extractant	Up to 50% with NaOH; simple structure	[20]
Controlled Oxidation	Coal, low-rank coals	Air, oxygen, ozone, or nitric acid oxidation	Produces artificial humic acids	[20]
Hydrothermal Conversion	Biomass, food waste, grains	Acid/alkaline hydrothermal process, sometimes microwave-assisted	Up to 43.5% yield; fast, efficient	[38,45]
Composting (Biotic)	Plant/animal residues	Microbial decomposition in reactors	35–248 g/kg (lab/pilot scale)	[13]
Catalytic Oxidation	Carbonaceous wastes	Catalysts and free radical mechanisms to enhance synthesis	High yield, energy efficient	[3]
Chemical Synthesis	Biomass pretreatment liquor	H_2O_2 oxidation, other chemical treatments	High metal-binding, electron transfer	[13]

6. STRUCTURE OF HUMIC ACID

The chemistry of the structure of humic acid (HA) is amongst the complicated and best researched areas of soil biomass. Humic acid is not a pure, crystalline substance, rather it is a heterogeneous product of various organic substances which are developed by the microbial and chemical processes of plant and animal residues during the humification. It is built in the form of a three-dimensional network of aromatic and aliphatic molecules linked together by different oxygen-, nitrogen-, and sulfur-based functional groups includes hydroxyl (-OH), carboxyl (-COOH), carbonyl (C=O), phenolic (-PhOH), amide (-CONH2) and methoxyl (-OCH3) within its molecular structure [21,53]. These properties make the humic acids possess a highly reactive surface chemistry that enables them to react with metal, nutrients and organic pollutants in terms of complexation, adsorption, and ion-exchanged reactions [24].

In previous models, humic acid was regarded as a big and covalently bonded macromolecule which is a result of polymerization of lignin, tannins, and phenols. Returning to the spectroscopic findings of modernity, however, has led to the supramolecular model and thus conceptualizes HA as an assembly of smaller non-homogenous molecules that are retained by weak non-covalent forces like hydrogen bonding, van der Waals forces, and hydrophobic interactions [23,25]. This is a model used to explain reversible aggregation and solubility of humic acids at different pH conditions as well as different ionic conditions. Humic acid fractions easily have a molecular weight of hundreds to thousands of Daltons, whereas this depends on the extraction procedure and the extent of humification [25].

Spectroscopic and chromatographic studies have depicted that humic acids have a backbone of condensed aromatic ring with aliphatic chains and oxygenated groups. Total solid-state ^13C nuclear magnetic resonance (NMR) spectroscopy has revealed that 30-60 percent of humic acids is usually comprised of aromatic carbon and 20-40 percent comprised of aliphatic carbon with the remaining part being comprised of carbohydrate- and peptide-like structure [24]. The aromatic component is mainly extracted out of lignin and polyphenolic precursors whereas the aliphic ones are extracted out of the waxes, lipids, and polysaccharides 21. The appearance of carboxyl, carbonyl, and phenolic groups stated by Fourier-transform infrared spectroscopy (FTIR) and ultraviolet-visible spectroscopy (UV-Vis) is what suggests the high light absorption capacity and cation exchange ability of humic acids [53].

The heterogeneity of HA structure is highly composed of it origin and isolation technique. The humic acid present in lignite and leonardite is more likely to be aromatic and hydrophobic whereas the acid in soil, compost, and peat aremore lean to oxygenated and aliphatic groups, and hence, are more polar and prone to reaction [24]. Their physicochemical behavior is governed by this structural variability, which affects their solubility, affinity to bind and reactivity with the environment. This is possible due to the appearance of both hydrophilic (carboxylic and phenolic) and hydrophobic (aliphatic and aromatic) domains, which allow humic acids to act as natural amphiphilic surfactants, which stabilize the colloids and also serve to transport both nutrients and pollutants in the soil-water systems [22].

Table 3: Key Structural Features of Humic Acid

Feature	Description	Role	Reference
Aromatic rings	Provide rigidity, π–π interactions	It enhances adsorption and complex formation, also provide chemical stability.	[5]
Aliphatic	Add	It improves	[3,5]

Feature	Description	Role	Reference
chains	flexibility, hydrophobicity	aggregation and structure stability.	
Carboxyl groups	Metal binding, acidity, solubility	Takes place in acidity, cation exchange capacity (CEG).	[5,49]
Phenolic/ quinone groups	Antioxidant, redox, radical scavenging	HA will act as antioxidant and radical scavengers.	[2,5]
Supramolecular assembly	Aggregation, colloid/micelle formation	HA exists as supramolecular structure.	[5]

7. ESTIMATION OF HUMIC ACID

Precise determination of a humic acid (HA) in soils and sediments, biomass and commercial products involvesintegration of fractionative methods of classical fractionation with spectroscopy and new sophisticated instrumental measurements since HA is no singular compound but a blend of several compounds. The common and used method adheres to the IHSS-fashioned alkaline extraction (0.1 M NaOH or KOH), acid correction to pH1 and gravimetric recovery of the precipitated HA portion; this gravimetric scheme is the standard platform of disclosing HA content however is known to be spoilt by co-precipitation, ashes interference and variability with extraction factors. The quickest, most frequently used approach to rapidly estimate concentrations of HA and the degree of humification: total HA (or HA-containing extracts) is measured using UV-visible spectrophotometry and structural proxies like the ratio of E4/E6(A465/A665) are used to estimate averages of the two additional parameters of average lobular size and aromaticity, though both methods need to be calibrated on the specific feedstock and extraction regime. Chemical titrations and colorimetric assays (such as total acidity, carboxyl/phenolic titrations and Folin-Ciocalteu phenol assays) are used to supplement the estimates of concentration because of the quantitative extrinsic groups in terms of which the reactivity and quality of HA is determined. To achieve increased confidence and overcome material heterogeneity, contemporary research combines spectrophotometry / gravimetry with other instrumental analyses - elemental (CHNS/O) analysis, FTIR, solid-state -13C NMR, fluorescence spectra, size-exclusive chromatography or HPLC, and mass spectrometry which combine to yield data on a compositional value (C/N/O ratios, functional groups, molecular-weight distributions) improving the procedure of quantifying and assessing the quality of HA fractions. Recent developments in method development (with both validated JoVE protocols and comparative studies) focus on (1) reporting HA on a non-ash-contaminated basis with or without (2) and without validating spectrophotometric calibrations with an independent validation by gravimetric or chromatographic standard and (3) workflows based on multiple techniques, since single methods are systematic, over- or under-estimators of HA based on the but not limited to the nature of the matrix and treatment. These best-practice suggestions are gradually becoming the norm in the soil, sapropel, lignite and commercial humate tests to enhance the reproducibility of the results across the laboratories and in quality control of the products.

Table 4: Comparison of major humic acid estimation methods and their accuracy.

Method	Principle/ Approach	Accuracy/ Limitations	Reference
CDFA, ISO 5073, NSM	Standardized extraction & gravimetry	CDFA and ISO 5073 tend to underestimate HA; NSM is more consistent and often used as a reference	[26,28]
Colorimetric	Color reaction with HA	Overestimates HA (by up to 64%) due to interference from soluble organics	[26]
Modified Extraction	Optimizedalkaline extraction	High correlation with validated methods; efficient and reproducible	[27,28]
Electroanalytical	Stochastic sensor/electroanalysis	Fast, no sample prep, high sensitivity, suitable for mud/water	[54]
Spectroscopic (UV, NMR, DTA)	Measures aromatic C, structure, or thermal properties	Useful for characterization and source discrimination, not direct quantificatioN	[29,31]
Potentiometric Titration	Acid-base titration for functional groups	Provides acid group content, not total HA mass	[33]

7.1 Key Insights and Best Practices

Comparable controls, as applicable in the determination of HA in products, are the application of NSM and gravimetric analysis where refining control parameters (e.g., 0.2 mol/L NaOH, 2-hours, sample solution ratio) presented by refined parameters increases accuracy and consistency. There are also similar origins of satisfactions that can lead to pain, instead of happiness [26,27].

Electroanalytical methods are sensitive, fast and lack preparation particularly when the samples are related to the environment such as aqueous and sediment samples [54].

Spectroscopic and titration assays are appropriate to analyze HA structure and functional groups and not as specific in the determination of protein amounts of total HA. It is linked to increased heightened consciousness exhibiting spontaneity and measurable affective characteristics including euphoria and paranoia [29,30].

The determination of humic acid depends upon the choice of the method and the sample. The most quantitative results are provided by gravimetric and optimized extraction using coded protocols and the colorimetric and some spectral methods will undergo a large error. Field application Electroanalytical techniques are quite suitable in quick field measurements [31].

8. CHARACTERISTICS OF HUMIC ACID

Studying of humic acid (HA) must be characterized to assist in determining the complexity of the form it happened to act and functional group and how reactive it is and how it explains its importance in the environment and agriculture. At present state of analysis, they are able to not only figure out the structural features of that compound, but also give an account in an exhaustive data about the same using all available spectroscopic, chromatographic, and thermal techniques. The preliminary characters have been feasible with extensive use of UV-Visible spectroscopy and fluorescence spectroscopy whereby fast indicators of aromaticity, molecular condensation and the degree of humification based on spectral proportion such as E4/E6 and fluorescence indices are represented. These parameters indicate that conjugated aromatic systems exists and the humic measurements of maturities of humic objects 53. FTIR spectroscopy is extensively employed by the way of identifying functional groups of characteristics of carboxyl (-COOH), hydroxyl (-OH), carbonyl (C=O), and phenolic (-PhOH), via which HA takes part in the functions of ion-exchange and chelating 34.With the help of solid-state 13C NMR used in NMR spectroscopy effectively yields semiquantitative data of distribution of different forms of aliphatic, aromatic and oxygenated carbon as the level of oxidation and aromatic condensation of the humic structure [35].

Elemental analysis (CHNS/O) and thermal stability are detected together with spectroscopy to apply elemental analysis and Thermogravimetric analysis (TGA) to the sample, respectively, and finally overall display of the organic and inorganic composition of HA is provided36. High-resolution mass spectrometry (FT-ICR-MS) breadth and other chromatographic techniques such as high-performance liquid chromatography (HPLC) and size-exclusion chromatography (SEC) are gaining factual presence in determining the issue of molecular-weights and locating the specific inventions of humic acids sources and processing change [37]. As per recent studies it is assumed to apply multi-technique characterization models which is a mix of spectroscopy, NMR, and mass spectrometry to provide a more precise, reliable and all encompassing structural heterogeneity of the structure and functionality in HA [53].

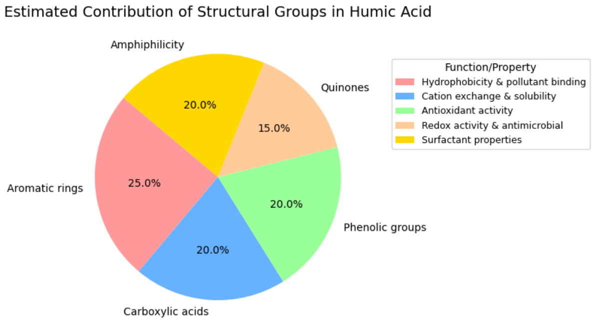

Fig 1: Key structural groups and their functions in humic acid

Humic acid is a structurally large, amphiphilic macromolecule with major roles in soil health, pollutant remediation, and various technological applications.

9. APPLICATION OF HUMIC ACID

Major Application Areas

Humic acid (HA) is a multipurpose natural compound whose applications find extensive usage in the agricultural, environmental cleanup, medical, as well as latest technology sectors. HA is very crucial in modifying the soil structure in terms of improving aeration, water retention and distribution of nutrients in the soil in amendments and fertilizer application. It improves cation exchange capacity (CEC) and higher bioavailability of nutrients, improving the development of crops (maize, wheat, coffee, broccoli, and thyme), yield, and stress resilience [38,39,40] and Noroozishar Besides acting as a soil conditioner, humic acid decelerates biochemical and biological roles including hormonal regulation (auxin and cytokinin regulation), photosynthetic activity and enzymatic activity, acting as a plant biostimulant. All these impacts increase the level of tolerance of plant to the abiotic stresses like drought, salinity and toxication by heavy [4,9]. HA application has also been reported to either enrich vital oil content and alter compositional content in aromatic and medicinal plants (thyme) and declare its impact on secondary metabolism and phytochemical accumulation [42].

Humic acid is essential in the ecological context of detoxification and sustainability of the environment. It adsorbs and immobilizes numerous pollutants important to the soil and water systems, including organic pollutants of pesticides, antibiotics, and heavy metals (Pb, Hg, Cd, Cu) [10,43,55]. Synthetic and natural HAs, as well, are utilizing these agents as soil wash agents in the elimination of heavy metals but they performed better than a few traditional agents used in remediation [44]. In addition, humic acids made synthetic with means of biomass have demonstrated potential in climate change reduction since it results in carbon sequestration and enhances soil fertility, thereby contributing to sustainable carbon management practices [52,55]. In addition to its application in the environment, HA demonstrates antiviral, antioxidant, and anti-inflammatory activity, which is why it can become a promising biomedical agent (wound healing and antiviral therapy [5,46]. Moreover, sodium humate has also been tested in phototheranostic implementation, as it is a biocompatible photothermal reagent used in the imaging and treatment of cancer since it has a high photothermal conversion efficiency and is not as cytotoxic [47].

The distinctive redox-active functional groups of humic acid create the opportunity to use it in the sphere of electrochemical and energy storage in the form of a redox mediator or buffer substance in the sensor, and energy storage [4,13]. All of these applications support the diversity of uses and environment adaptability of humic acids. Its multifunctional nature is a consequence of its complex molecular architecture, full of carboxylic, phenolic, and quinone functionalities, to place it in the most significant role in the sustainable use of agriculture, ecology, medical research, and green technology formation [13,31].

10. DRAWBACKS OF HUMIC ACID

Limitations of Humic Acid: Main Risks and Restrictions

High concentration of humic acid poses a major disadvantage to biological wastewater treatment system especially those that utilize specific consortia of microorganisms in the treatment of wastewater. The basic anaerobic oxidation of ammonia (Anammox) process, on which it relies to get nitrogen in advanced wastewater treatment, is gravely impaired as soon as the level of humic acid goes above the threshold. According to CPA et al. (2018), humic acid shows considerable inhibitory properties against Anammox bacteria, a vital part of the microbial population involved in the transformation of ammonium into nitrogen gas at concentrations over 70 mg/L. The inhibitory process acts in several lines: humic acid imposes barriers to the passage of substrate to bacterial cells, and accumulates substrate toxicity, which does not allow the application of effective metabolic activity. The applied implications are cataclysmic, at 200 mg/L levels, ammonium removal efficiency has dropped by nearly 57 percent, which is a dismal loss in treatment efficiency. In addition to direct bacterial inhibition, long term exposure to the large levels of humic acid results in undesirable ecosystem-scale modifications in the wastewater treatment reactor. The microbial community should experience dysbiotic shift in which the abundance of helpful symbiontic bacterial species such as Candidatus Brocadia fulgida--a primary species of Anammox aerobic organism- is reduced and, at the same time, propagation of unwanted heterotrophic denitrifiers that are not involved in the effective elimination of nitrogen is fostered. This displacement is unfriendly to the ecological balance of the biological treatment system, and thus when introduced the community structure has been broken and to reestablish the functionality becomes difficult or impossible. Studies have shown that humic concentrations below 150mg/L may in fact cause anammox activity, whereas above 150mg/L a shift towards inhibition occurs that clearly shows that there is a concentration-dependent threshold effect. The practical implications are that the humic acid level should be highly monitored to ensure that the treatment of wastewater is still operational and therefore close monitoring and control measures should be taken to avoid any failure of the system [48].

The second significant limitation of humic acid pertains to its metal-complexation not only it creates system instabilities that significantly depend on the pH value and environmental conditions, but also these instabilities have extreme values. Although the idea that the humic acid can bind the metal ions like iron is generally accepted to have positive effects on the environmental remediation and sequestration of metals, the same property leads to a paradox when the pH fluctuates. The stability of the humic acid-metal complexes is highly pH dependent, at pH 5 the stability constant of a humic acid-iron complex is nearly 5.26 whereas at pH 7, the stability constant is slightly lower at 5.18. More importantly, the absolute amount of iron complexed with humic acid at a pH of 7 rises to about 117 cmol/kg as compared to a rise by merely 57 cmol/kg to stability of the complex at a pH of 5. This seeming contradiction is caused by the fact that the formation of iron hydroxide precipitates at neutral to slightly alkaline pH ranges leading to a destabilizing effect on the system by co-precipitation. This instability has its structural foundation in the coordination chemistry of humic acid functional groups of iron ions. In humic acid at pH 5, the high-affinity chelate (bidentate) coordination modes of the cationic crossover atom, iron are used, in which case the electrostatic repulsion of carboxylic and phenolic and the negatively charged groups secures the complexes, resulting in stability of the solution. However, with a further rise in pH towards a neutral (pH 7), the mode of coordination changes to the bridging position, a less stable structural motif by nature. At these elevated PH level, the iron is more likely to form insoluble iron hydroxide [$Fe(OH)3$], used as a flocculant and the metal and the organic matter become insoluble together in the solution [49].

In contrast, pH reduction gives rise to another, however, just as bad destabilization process. The negatively charged functional groups of the humic acid are protonated in acidic pH and neutralized by competing metal cations (H + and other divalent cations including Ca 2 + and Mg 2 +) and neutralizes the electrostatic charges holding the complexes together. Such neutralization brings together the electric layer around the humic acids to decrease the repulsion between the charges and enhance aggregation and precipitation of the humic acids. What is obtained is the loss of solution stability and flocculation of the humic acid-metal complexes. Also, steric constraints in the three -dimensional arrangement of the humic acid further limit the ability to bind metals. The intra-molecular clusters and crowding effects imply that only a small proportion of carboxylic and phenolic functional groups are free to contribute to the coordination of the metal; about 80 per cent of carboxyl and phenolic groups in pH 7 and 90 per cent in pH 5, are not occupied because of steric hindrance. This is a drawback that makes the theoretical maximum of the possibility of complexation unachievable. The real-life implications of such pH dependent instabilities are enormous, especially in the soil and water environment where pH inherently varies. The structure of humic acid is further condensed at high ionic strength in estuarine systems where the salinity and ionic strength varies significantly and this further, decreases iron-binding capacity and facilitates complex dissociation and co-precipitation. Experiments have shown that the iron binding capacity of humic acid was found to have a negative relationship with salinity ranging between 5 and 40 ppt with the capacity following a pattern of 41.40 nmol Fe on 31.50 nmol Fe on 1 mg HA which is about a 24 percent loss in the potential of complexation. The use of humic acid in conditions with changing pH or ionic strength cannot. Therefore be considered dependable in offering equal amounts of metal sequestration or complexation and this is essentially the main limitation of its use as an environmental remediation process due to the varying environmental conditions that occur unpredictably. Perturbation of the system by the change in pH values implies that the intended changes of subjecting the metals to solution to make them bioavailable or elimination of the solution to trap contamination would not be guaranteed unless the environmental change is strictly controlled which might not be feasible in most field scenarios [50].

11. CONCLUSION

In a small nutshell, humic acid is the complex organic material of different meanings in the fields of science, agricultural field, industrial sector, and in environmental science. Humic acid was generated as a result of the humifying vegetal and animal leftovers and soils, leonardites, lignites, are inherently rich in humic acid, fragment of humic acid has become commonplace and subsequently there has been wide usage of the substance. Although, classical approach to alkaline extraction is the most competent procedure in production, innovative production means have given rise to biotechnology based methods of production, which are both greener and clean. Use of the unusual supramolecular topology which is determined by a heterogeneous environment of aromatic and aliphatic molecules, including carboxyl, phenolic, quinone, and methoxy functional groups, is supported by amphiphilicity of humic acid, its high level of chelation ability, and its buffering pH. Analytical reagents which are authored such as those of an UV-vis spectroscopy, FTIR, NMR, elemental analysis, and colorimetric assays, make the process of accurate quantification and structure elucidation possible that may ultimately lead to quality control and formulation development.

Physicochemically, humic acids occur at molecular weights of 1. 7-6 kDa, colloidal stability and in all the nutrients and the pollutants. Their multifunctional usage is informatic: as biostimulants, to catalyze nutritionally in soil: biostimulating nutrient uptake and counteracting or mitigating plant tolerance; as active ingredient in ceramic, battery, cosmetic and nutraceutical product production. The technology however has its problems; diversification of quality of raw material, a possible ecological effect on the water quality, extraction costs, and energy use, industry uncertainty, necessitates the employment of conventional methodologies, the life cycle analyses, and the extensive field studies.

The direction of studying in future must improve the bioprocessing process of humic acid production to its useful form through a sustainable fashion with an aim of characterization of the form and structure of the structures with respect to determining the relations of the structures with functionality and a development of its application in a sustainable way of maximizing the effectiveness and the minimal impact to the environment. With the cross-disciplinary expertise of soil science, chemistry, biotechnology, and environmental engineering continued to be sharpened the whole possibility of humic acid will be used to permit sustainable agricultural systems, the circuitous economy and development of green technologies that are new.

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14. CONFLICT OF INTEREST

The authors declare no conflict of interest.

15. INFORMED CONSENT

Informed consent was obtained from participants

16. ETHICAL STATEMENT

Confirm that the study was approved by a recognized ethics committee,

17. AUTHOR CONTRIBUTION

Data collection, analysis

18. REFERENCE

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