

Fluorescence and photochemical properties of humic acids

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To obtain new information on the properties of humic acids (fractions of humic substances), we have studied the fluorescence and photochemical properties of various samples of humic acids alone and the photodegradation of phenolic ecotoxins in the presence of humic acids. The influence of different light sources and wavelengths of the exciting radiation has been studied, as well as the capability of humic acids to affect the photolysis in different ways depending on their origin.

Introduction

Processes, occurring in nature, are the subject for studies in the physics and chemistry of the environment. In considering the processes taking place in the biosphere, especially, in aqueous media, it is necessary to take into account the presence of humic substances (HSs), since these are almost always present in natural waters. The HSs are very important class of natural high-molecular compounds and form the basis for vital activity of living organisms. These natural organic compounds with a complex composition attract the growing attention due to their properties. All natural waters contain the dissolved organic matter. Marsh waters are very rich in humic substances. The HSs can absorb solar energy and transport it to other substrates, in some cases affecting significantly the photolysis of xenobiotics. It has been found that in water and soil HSs act as photosensitizers, produce active forms of oxygen, and can photoinitiate the transformations of the ecotoxins. The HSs can also act as quenchers or as photostabilizers.

The HSs have an irregular structure, which is not fully known yet. The natural selection of the most stable products determines their very complex structure. However, their structures have some inherent internal regularity. All these macromolecules contain an aromatic carbon skeleton, enriched with functional groups and alkyl radicals; the periphery part of carbohydrate-peptide fragments; and mineral components. The HSs include a large set of different functional groups: ketyl, quinoid phenol, amino-, carboxy-, methoxy-groups, which assume the possibility of various photochemical processes to occur, although HSs in general have relatively high light fastness. A great attention is paid to the study of the HS composition and structure parameters by various methods (ESR, NMR, and others), and certain progress has been achieved in this field.^{1,2} The situation with the study of HS functional properties is more difficult, although it is known that these organic substances compensate, to a significant degree, for the negative effect of the anthropogenic impact.³ As an example, the decrease in the content of mobile forms of some heavy metals,⁴ as well as the

regulation of the effect of acid atmospheric precipitation⁵ can be mentioned. At the same time, the data obtained using laser-induced fluorescence show⁶ that some amounts of humic and fulvic acids are present even in the purified tap water.

In the biosphere, humic substances have various important functions, like accumulation, transport, regulation, protection, physiological, and many other functions.^{7,8} The HSs form the group of natural compounds, which is one of the most difficult to study, and numerous functions of these substances are still poorly investigated. The HSs act in many ways, for instance as sorbents, catalysts in hydrolysis processes, solubilization agents; they also influence microbiological processes and serve photosensitizers and quenchers.⁹

The photochemical properties of HS are poorly studied, although it is known that humic substances can absorb light and transport the light energy to other components of aqueous solutions, strongly affecting the photolysis of xenobiotics in some cases.¹⁰ It has been found¹¹ that HSs can act as photosensitizers if exposed to the radiation with the wavelength longer than 290 nm. The capability of these substances to produce active forms of oxygen was reported,¹² as well as the capability to induce herbicide transformations under the action of light.¹³

It is known¹⁴ that the photodecomposition of ecotoxins in pure water under the exposure to sunlight takes about 85 h, while in the natural water it takes only about 60 h. The photochemical activity of HS is still poorly studied, although these properties are directly connected with the problem of redistribution of the photoexcitation energy in molecules, which is one of the most important problems in basic science.¹⁵ There are some contradictory data about the mutual influence of HSs and organic ecotoxins in the process of photolysis. The investigation of basic regularities of the HS photolysis (in particular, in the presence of ecotoxins of various structure) is extremely urgent, because it helps to reveal the influence of optical radiation of the natural and anthropogenic origin on the environmental processes. As Lord George Porter, a prominent photochemist, noted, the

optical radiation is of principal significance for the functioning of the biosphere.

Experiment

The HSs consist of three fractions: fulvic acids (FAs) soluble in water at any pH, acid-precipitated humic acids (HAs) soluble in water at pH > 2, and water-insoluble humins. According to contemporary conceptions, just humic acids form the most representative group of humic substances.⁸ Therefore, the results obtained from the study of humic acids are of greatest interest for investigators.

We have studied the spectral-luminescent properties of humic acids before and after the UV irradiation, as well as their interaction with phenol ecotoxicants. The absorption and fluorescence spectra were recorded following standard technique on a Specord M40 spectrophotometer and Hitachi M850 and CM 2203 spectrofluorimeters. The sources of UV radiation for photochemical studies were: 1) OKN-11M high-pressure mercury lamp; 2) a spontaneous radiation source – pulsed barrier-discharge-pumped KrCl exciplex lamp (of the U-type with the following parameters: $\lambda = 222$ nm, $\Delta\lambda = 5$ –10 nm, $W = 18$ mW/cm², $f = 200$ kHz), developed at the Institute of High-Current Electronics SB RAS, in the lab headed by Professor V.F. Tarasenko.¹⁶

We studied the samples of humic acids of different degrees of humification, kindly placed at our disposal by L.I. Inisheva (Siberian Scientific Research Institute of Peat, Russian Academy of Agricultural Sciences), extracted at the Institute of Petroleum Chemistry SB RAS in the group headed by N.V. Yudina. The objects for study were extracted from low-moor peat (pH = 4.9, Tagan, Tomsk Region). We have also studied the samples of HAs of different origin, kindly placed at our disposal by V.V. Demin (Department of Soil Science, Moscow State University). These samples were extracted from peat (produced by Merck Co.), ordinary chernozem from the bicentennial fallow land of Kamennaya Steppe (Talovo District, Voronezh Region); from the sod mesopodzol on the moraine loam (Lesnaya test cottage of the K.A. Timiryazev Moscow Agricultural Academy); from the intensively weathered brown coal of the Kansk-Achinsk coal field (TU 211-06-18-94 an industrial preparation of sodium humate); from the podzol pseudogley peated soil (white podzol) from the Tsentral'no-Lesnoy State Natural Biospheric Reserve, Tver Region.

Discussion of the results

The electron spectra (UV and visible regions – from 220 and to 750 nm) explain the dark color of the humic substances. The HSs are characterized by the intense absorption in the UV spectral region, and the absorption decreases gradually with the increasing wavelength.¹⁷ The HS spectra look like curves, gently sloping toward long wavelengths, with almost no peaks and dips (very low peaks are observed sometimes nearby 430, 448, 568, and 613 nm).

Some authors explain this shape of the curves by the fact that such molecules have a developed chain of coupled double carbon–carbon bonds. The destruction of this chain upon the photochemical degradation leads to the gradual loss of the color. Our data show that the absorption spectra of sedge HSs and cotton-grass HSs are almost identical, but in the region of 280–350 nm the sedge HS molecules absorb more intensely. In the spectra of humified HS molecules of sedge and cotton-grass, the absorption intensity decreases all over the spectrum. For the samples taken from the peat, we see the increase of the absorption intensity in the region of 220–360 nm. From the data of the absorption spectra of water-alkaline solutions of HSs from peat (Merck), chernozem, sod-podzol, brown coal, podzol (MSU), we can see that the absorption spectra of HSs from the peat and the podzol soil are virtually identical, but in the region of 42500–30000 cm⁻¹ molecules of the latter absorb more intensely (Fig. 1). For other HSs, the absorption intensity increases all over the spectrum.

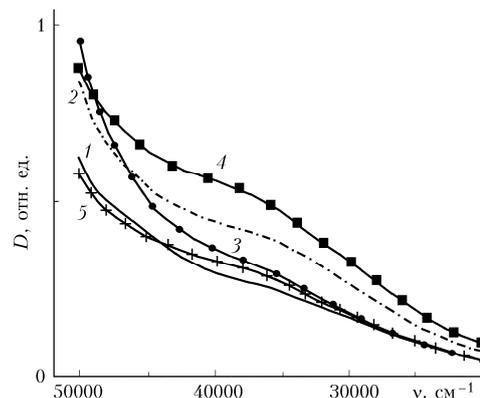


Fig. 1. Absorption spectrum of HS in the water-alkaline solution at $C_{\text{HS}} = 0.0098$ g/l, $C_{\text{NaOH}} = 0.0019$ m/l: HS from the Merck peat (1), chernozem (2), sod-podzol (3), brown coal (4), and podzol (5).

As to the fluorescence-based methods those are characterized by the high sensitivity, as well as the convenient time intervals as the fluorescence occurs roughly 10^{-8} s (10 ns) after absorption. For this interval, many different molecular processes can take place, and they can affect the spectral characteristics of the fluorescing compound. Such a combination of the high sensitivity with the suitable time range has led to the situation that the fluorescence methods are used widely in various fields of science, in particular, in the studies of humic substances.^{2,18} The fluorescence spectra of the HS samples of different degree of humification we have studied are in the long-wavelength region with peaks near 490 nm.

The data obtained allow us to conclude that, in the process of humification, the fluorescence intensity decreases, while the region of fluorescence does not change significantly. The change in the intensity is connected with the structure change of the molecules and the change in the component percentage. The

fluorescence spectra ($\lambda_{\text{pump}} = 365 \text{ nm}$) of the solutions of HSs of different origin differ much more significantly than their absorption spectra (Fig. 2).

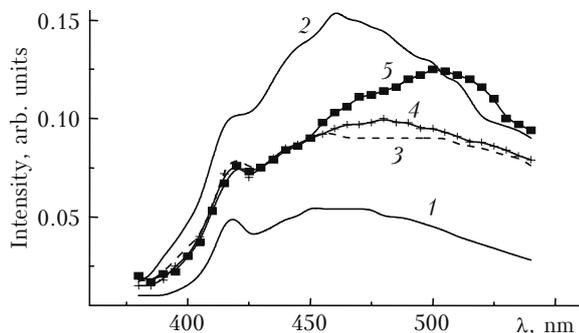


Fig. 2. Fluorescence spectrum of HSs in water-alkaline solutions $C_{\text{HS}} = 0.0098 \text{ g/l}$ and $C_{\text{NaOH}} = 0.0019 \text{ m/l}$; $\lambda_{\text{pump}} = 365 \text{ nm}$; HS from the Merck peat (1), chernozem (2), sod-podzol (3), brown coal (4), and podzol (5).

The peak of the fluorescence band of the HS from the peat and HS from the chernozem occurs at 460 nm, and that of the HS from the sod-podzol, coal, and podzol is at 480 nm. This shift is connected with the increase of aromatic fragments in macromolecules.

Let us now discuss the properties of irradiated samples. No significant spectral changes were observed in HSs of different degree of humification exposed to the mercury lamp radiation for 4 h. Consequently, the considered HSs are rather photostable to the irradiation at $\lambda = 365 \text{ nm}$. The absorption spectrum of the sedge HSs has an inflexion in the range of 250–350 nm, and in the process of irradiation the absorption intensity in this range increases. The changes in the fluorescence spectra are more significant than those in the absorption spectra.

The most significant photodestruction of HSs occurs under the exposure to short-wavelength radiation of the excilamp (222 nm). Consequently, the use of the radiation with this wavelength is promising for water cleaning from organic impurities. It has been found that humic acids can serve both as photostabilizers and photosensitizers for organic microimpurities (phenol and its substitutes were considered) depending on the characteristics of the pump source.

Such a complicated photochemical behavior of humic substances can be explained based on the results obtained at studying the properties of HS triplet states using spectra of light absorption from excited states.¹⁹ The laser photolysis of HSs has revealed the possibility of generating a number of triplet states with the use of different pump wavelengths, and therefore the HS photochemical properties depend on the characteristics of pump radiation.

Humic acids, being the very important component of peat of different composition, influence natural ecosystems of the biosphere in different ways. The existing classifications are based on the descriptive properties and cannot be used for

scientific analysis and prediction. The database, we do compile, will include the data on the elemental, fraction, and aminoacid composition of peat, along with the visible and IR spectra of the HSs. The obtained results on the fluorescence and photochemical properties of the HSs will also be included in the database of the properties of HSs of different origin.

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