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Correlation of humic substances chemical properties and their thermo-oxidative degradation kinetics

Part 1. Arrhenius equation as the temperature function

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Abstract Application of the Arrhenius equation as the temperature function in modeling of the degradation kinetics of humic substances brought a high positive Pearson correlation coefficient with the carbon content and a reasonable negative correlation with the oxygen content. Ratio C/H indicating the aromaticity degree of humic samples did not show any significant correlation. Relatively high value of correlation coefficients provided also O + N and ratios C/(O + N) and C/O, respectively. In contrast, H, N content and natural and heat generated free radical content and their ratio gave substantially lower correlation coefficients. The latter indicates that free radicals are probably not the main reason of the collapse of the secondary structure of humic substances leading to their degradation.

Keywords Humic substances · Isoconversional methods · Single step approximation · Correlation analysis · Thermal analysis

Introduction

Climate change and global warming represent the hot issue of both scientific and laical discussions. It is well known that among the most important contributors of green house gases belong emissions from natural systems having high carbon content such as soils and peats. The temperature increase promotes their faster degradation and erosion which results in a depletion of soil organic matter quality. As a result, the soils characteristics such as fertility, water holding capacity or biodiversity are strongly affected. Thus the capability of soil to fix carbon back from atmosphere by means of present plant photosynthesis is diminished.

Soil organic matter generally consists of two parts: fresh litter fall and humified pool. While the former is quickly used up as a source of energy by local macro or microorganisms, the latter is, due to its intimate contact with soil inorganic matter and high heterogeneity, more stable. Due to the disproportion in time period of soil organic matter turnover [1], the character of such stable part called humus represents a pool of soil which is thought being an indicator of whole soil "quality". It is formed by microbiological activity (i.e., breaking up of the labile part) and its turnover is related to the local conditions. As both whole soil and humic substance represent complex systems, definition of such "quality" remains still a problem. Many techniques and approaches have been employed to study chemical–physical properties of humic substances [2] and thermal analysis belongs among them [3–10].

Traditionally humic substances are divided into three groups according to their solubility. Although, such categorization is rather artificial, it reflects the nature, strength and distribution of polar groups in the humic mixture associated with their role with respect to the parent ecosystem. From the chemical composition point of view, one of the most frequently used measures is for example degree of aromaticity, often connected with the degree of humification (i.e., processes that are believed to lead to the formation of humic substances). However, aromaticity degree does not reflect the real "quality" of humic matter since some fulvic acids have higher aromaticity degree than humic acids from a different source [11].

A reliable prediction of the natural organic matter turnover is an important parameter for modeling of the carbon flux and related subjects. A perusal of literature

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showed the scattered applications of thermoanalytical techniques for soil quality evaluation. Enthalpies produced by metabolic activity of soil microorganisms was studied using calorimetry to determine their metabolic activity [12–14]. High correlations between soil chemical composition and mass loses in specific temperature regions have been demonstrated by Siewert [15] who consequently developed a method allowing calculation of C, N, clay and carbonate content in a whole soil sample using a single thermogravimetric record. Further, an interesting interrelationship between soil respiration and mass loss recorded by thermogravimetry has been also demonstrated [16]. Similar approach was used when the slope of thermogravimetric degradation curve (i.e., the degradation rate) was successfully correlated with CO_2 evolved by soil respiration [17]. Recently, isoconversional times corresponding to specific stages of soil degradation were successfully interrelated to stability/lability of soil organic matter represented by the rate of soil respiration [18].

The attempt to determine reliable kinetic parameters in solid state by methods of thermal analysis is usually associated with many problems and misunderstandings. Kinetic parameters are usually interpreted in terms of kinetics in liquid or gas state despite the fact, that processes can be complicated by diffusion processes and the conversion function is not known. Isoconversional methods using the single step approximation can overcome this problem [19, 20]. Although they do not allow elucidation of the character of studied processes from the mechanistic point of view, they can be used to make quantitative predictions. Recently, that approach was used to determine the role of individual compounds in the stability of humic acids [10]. Since they represent a complicated mixture of molecules, their degradation tends (as most of solid state processes) to occur in multiple steps that have different rates. Therefore, the application of model free kinetics appeared to be a suitable tool to recognize the role of individual molecular groups. Basically, the process of degradation was divided into several parts and their isoconversional time corresponding to stability was calculated. Obtained values of time were compared and results showed whether and how were the stability of the sample and its intermediates affected [10].

The main aim of this work concerns to the determination of isoconversional times (stabilities) of various humic substances during their thermo-oxidative degradation. Obtained data were correlated with the chemical composition of humic substances. In this first part, to determine isoconversional times, the Arrhenius equation was used as a conversion function. The linear correlation between obtained isoconversional times and values of primary properties of HS samples was evaluated using the Pearson product–moment correlation coefficient.

Experimental

Humic samples

The samples of humic substances (fulvic acids, humic acids and humins) were obtained from various natural sources. A set was purchased from the International Humic Substances Society (IHSS). Samples were named as follows: Elliott Soil FA (HS01), Elliott Soil HA (HS02), Pahokee Peat FA (HS03), Pahokee Peat HA (HS04), Suwannee River FA (HS05), Suwannee River HA (HS06) and Leonardite (HS15). Details on samples can be found on the web page of IHSS (www.ihss.gatech.edu).

Other three samples were soil humic acid, fulvic acid and humin provided and characterized by Department of Soil Science and Geology, Faculty of Agrobiology, Food and Natural Resources, Czech University of Life Sciences in České Budějovice. Humic substances were extracted from soil originated from Jizerské hory, Czech Republic and were extracted from O soil horizon (700 m above sea level) using standard IHSS procedure. Respecting samples were as marked as follows: fulvic acid (HS07), humic acid (HS08) and humin (HS09). HS10 were humic acids extracted from Slovak agricultural soil. Additional information can be found in ref. [21]. HS11 were humic acids obtained from North Bohemian leonardite-type lignite (so-called Oxyhumolit), mine Bílina, using standard extraction procedures. Briefly, 200 g of previously air-dried and 0.2–0.3 mm sieved lignite was mixed with 2,000 mL of 0.5 M NaOH and 0.1 M $Na_4P_2O_7$ and stirred for 2 h. After separation by centrifugation (ROTINA 46R, 4000 RPM, 15 min; in all cases), the supernatant was treated with concentrated HCl until the pH value of 1–2 was reached in order to precipitate the HA. Then approximately 40 mL of HF was added and the final solution was diluted with distilled water to 1.5 L and shaken overnight to remove the residual ashes. After that the sample was centrifuged, rinsed with distilled water, centrifuged again, dialyzed (Spectra/Por$^{®}$ dialysis tubes, 3,500 Da cut-off) against distilled water until chloride-free, and finally freeze-dried (Labconco FreeZone). Details on Oxyhumolites and properties of humic substances can be found in refs. [22–24].

The sample HS12 were humic acids extracted from Indian Black earth soil over Amazonian forest [25].

HS13 was a humic acid isolated from B horizon of ferrohumic podzol in Trojmezí stand (Bohemian Forest, Czech Republic) [26].

The last sample, HS14, was a humic acid obtained from a volcanic soil (Typic Xerofluvent) from Vico, Italy [27].

Chemical composition data are summarized in Table 1; possible sulphur traces in humic substances are included in oxygen content.

Table 1 Elemental composition of humic substances (weigh percentages), free radical content (EPR0), generated free radical content (EPRm) and their respective ratio EPRr

Sample/analysis	HS01[a]	HS02[a]	HS03[a]	HS04[a]	HS05[a]	HS06[a]	HS07	HS08	HS09	HS10	HS11	HS12	HS13	HS14	HS15[a]
C[b]	50.6	58.1	50.5	56.4	52.4	52.6	50.7	54.9	55.1	57.5	62.7	50.8	49.4	53.7	63.8
H[b]	3.77	3.68	3.52	3.82	4.31	4.40	3.51	4.41	4.74	4.20	3.42	4.54	2.47	4.90	3.70
N[b]	2.72	4.14	2.56	3.69	0.72	1.19	2.94	4.07	3.80	6.10	2.03	5.99	2.55	4.30	1.23
O[b]	43.7	34.1	45.5	37.3	42.2	42.5	42.9	36.6	36.4	32.2	31.8	38.6	45.5	37.1	31.3
Ash	0.86	0.88	4.61	1.12	0.46	3.10	24.5	1.80	3.20	0.32	2.60	9.20	2.80	10.0	2.58
C/H	13.4	15.8	14.3	14.8	12.2	11.9	14.4	12.5	11.6	13.7	18.4	11.2	20.0	11.0	17.2
C/O	1.16	1.71	1.11	1.51	1.24	1.24	1.18	1.50	1.51	1.79	1.97	1.32	1.08	1.45	2.04
C/(O + N)	1.09	1.52	1.05	1.37	1.22	1.20	1.11	1.35	1.37	1.50	1.86	1.14	1.03	1.30	1.96
EPR0 × 10^{16} [c]	3.50	56.4	3.25	37.6	8.45	1.72	12.6	29.5	10.6	34.3	22.6	56.4	10.7	110	32.0
EPRm × 10^{16} [c]	3.01	329	7.86	127	5.41	2.20	17.1	182	32.9	114	59.1	286	32.9	366	206
EPRr	0.86	5.83	2.42	3.38	0.64	1.29	1.35	6.17	3.09	3.33	2.62	5.06	3.09	3.32	6.42

[a] Data provided by International Humic Substances Society (www.ihss.gatech.edu)

[b] In mass %

[c] In number of spins per gram of HS

Thermal analysis

Thermogravimetric analysis of each sample in oxidative atmosphere was performed at several heating rates using Shimadzu DTG-60. The furnace was calibrated using fusion transition temperatures of indium, tin, lead and zinc (156.6, 231.93, 327.46 and 419.53 °C, respectively). Samples were carefully homogenized in an agate mortar and weighed to approximately 2–3 mg and measured in an open aluminum pan. The oxygen flow rate was set at 20 mL per minute.

The heating rates were 0.5, 1, 3, 5, 7, 10 and 15 K per minute. The measurement was carried out from room temperature to 600 °C. Obtained results were evaluated by means of TA-60 software.

Data treatment

The thermogravimetric records showed two steps of mass loss, the first one attributable to the loss of water, whereas the second one (in Fig. 1 marked with a vertical line) to the degradation of organic matter. The latter step was used to assess the stability of examined samples. The beginning as well as the end of the process was also verified by the first derivative of the TG curve (DTG). The mass losses obtained at different heating rates were recalculated to conversion intervals (0–100%). Within this interval, several conversions were selected and for those the isoconversional times were calculated (given in Table 2) as described elsewhere [28–31]. Briefly, the calculation considers the linear increase of temperature, combination of general kinetic equation (1) with the Arrhenius equation and a plausible assumption of independence of the conversion α_i, corresponding to (the end of) some specific isoconversional time t_i, on the temperature regime. The single-step approximation [28] employs the assumption that the rate process function Φ in Eq. 1 can be expressed as a product of two separable functions independent of each other, the first one $k(T)$ depending solely on temperature T and the other one, $f(\alpha)$, depending solely on the conversion, α.

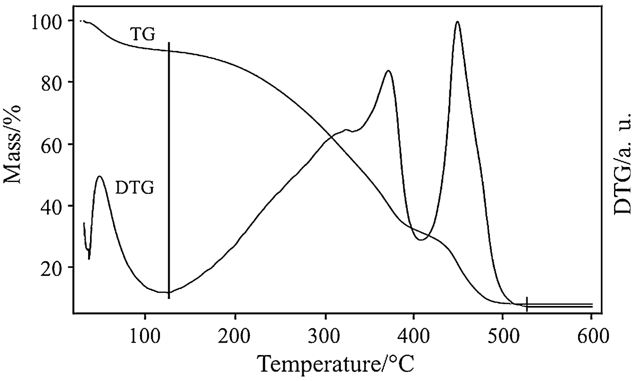

Fig. 1 An example of thermo-oxidative degradation of HS03 at heating rate 3 °C per min. The vertical line indicates the degradation onset

$$\Phi(T, \alpha) = k(T) f(\alpha) \tag{1}$$

Almost exclusively, the Arrhenius equation is used as the temperature function $k(T)$, i.e., $k = A_k \exp\left[-\frac{E_a}{RT}\right]$. Two parameters, A and B, are involved in the calculations and defined as follows:

Table 2 Isoconversional times (years) of thermo-oxidative degradation of humic substances samples extrapolated to 100 °C

Sample/conversion	HS01	HS02	HS03	HS04	HS05	HS06	HS07	HS08	HS09	HS10	HS11	HS12	HS13	HS14	HS15
0 ($\times 10^3$)	6.300	0.066	0.172	3.960	1.410	1.430	0.079	9.960	0.830	1.740	1.950	0.007	1.070	1.670	14.60
10	0.120	0.240	1.250	0.200	0.340	0.610	0.040	0.370	0.280	2.900	11.05	1.020	0.720	2.420	8.180
20	0.830	1.480	4.830	1.100	2.850	1.740	0.740	1.790	9.620	14.42	61.32	10.27	9.070	8.300	32.53
30	4.250	12.96	9.150	3.440	5.210	3.110	7.770	3.070	14.56	29.40	80.83	29.23	15.18	12.16	130.3
40	12.50	91.35	13.74	14.93	9.290	10.24	114.3	7.060	21.46	77.63	162.5	116.4	17.72	56.72	795.7
50	21.90	687.7	29.06	132.1	12.09	29.15	631.9	44.20	104.2	304.9	781.5	270.7	64.28	256.6	3141
60 ($\times 10^{-1}$)	17.00	487.6	5.222	87.01	5.112	12.58	120.5	16.80	29.96	183.1	96.07	80.25	30.66	29.87	614.5
70 ($\times 10^{-3}$)	1.990	17.52	0.183	3.317	0.833	1.412	7.582	0.697	0.614	10.07	5.890	1.192	2.362	0.478	12.81
80 ($\times 10^{-3}$)	17.10	52.73	1.38	8.581	27.02	8.178	53.01	1.302	1.172	38.06	42.76	2.262	5.438	20.02	32.46
90 ($\times 10^{-4}$)	18.00	162.4	0.927	1.560	38.01	2.738	6.24	0.325	0.602	15.35	4.280	0.841	1.281	1.283	8.231
100 ($\times 10^{-5}$)	3.580	5.400	3.491	2.422	3.694	2.459	5.867	1.649	2.058	5.310	5.019	1.539	1.443	1.917	198.4

$$A = \frac{F(\alpha) - F(0)}{A_k} \tag{2}$$

$$B = \frac{E_a}{R} \tag{3}$$

Here, A_k is the pre-exponential factor in Arrhenius equation, E_a is the (apparent) activation energy, R stands for the gas constant, and $F(\alpha)$ is the integrated inverted conversion function [19–22]. Due to the above assumption, also the value of the integrated function $F(\alpha)$ at the point α_i, i.e., $F(\alpha_i)$, is independent of temperature. The parameters are related to the applied temperature increase (scan) rate (β) according to the following equation:

$$\beta = \int_{T_0}^{T_i} \frac{dT}{A \exp(B/T)} \tag{4}$$

T_0 is the starting temperature of the measurement, equal to zero in our case, T_i is the isoconversional temperature, i.e., temperature corresponding to the conversion α_i. Calculated parameters A and B are listed in Table 1.

The parameters describe the temperature dependence of the isoconversional time [19–22]:

$$t_i = A \exp(B/T) \tag{5}$$

The isoconversional time t_i is the interval in which conversion α_i at temperature T is reached. This method has been successfully used for the prediction of stability of various materials at defined temperature [28–30, 32–34]. In this work, using Eq. 5, the parameters A and B served for the evaluation of stability at 100 °C. That temperature was adopted since many problems can appear when data obtained at higher temperatures are extrapolated to lower ones [32]. Further, properties of humic substances (Table 1) were correlated with the calculated isoconversional times reported in Table 2.

All calculations reported here were carried out by the software KINPAR [28–30, 32, 33]. Except the conversion 0%, where some problems appeared with onset determination, the difference between theoretical and measured values never exceeded 2 °C.

From the non-isothermal measurements, the kinetic parameters A and B have been obtained by minimizing the sum of squares between experimental and theoretical values of heating rates by a simplex method.

Statistical analysis

The correlation values between the isoconversional times and values of primary properties of HS samples were calculated using the Pearson product–moment correlation coefficient, which is obtained by dividing the covariance of the two variables by the product of their standard deviations. The correlation is defined only if both of the standard deviations are finite and both of them are nonzero. It is a corollary of the Cauchy–Schwarz inequality that the correlation cannot exceed 1 in absolute value. The correlation is 1 in the case of an increasing linear relationship, −1 in the case of a decreasing linear relationship, and some value in between in all other cases, indicating the degree of linear dependence between the variables. The closer the coefficient is to either −1 or 1, the stronger the correlation between the variables. If the variables are totally independent then the correlation is 0, but the converse is not true because the correlation coefficient detects only linear dependencies between two variables. Correlation coefficients were computed using MS© Excel.

Electron paramagnetic resonance

The Electron Paramagnetic Resonance (EPR) spectra of all HS samples were obtained on solid samples packed in quartz EPR tubes using a SpectraNova ESR 70-03 XD/2 spectrophotometer. The scans were performed to determine the natural content of free radicals (time 0) denoted as EPR0 and then for 3 h in 20 min intervals. This temperature of isothermal heating was chosen with respect to previous investigation [8–10], i.e., when no significant heat evolution or mass loss has been registered. The relative area of EPR signals was obtained by double integration and compared with EPR measurements of standard sample of known spin per gram content (strong pitch supplied by Bruker). The maximal free radicals content is denoted as EPRm. The ratio of free radicals content between EPR0 and EPRm, i.e., EPRr was calculated.

Results and discussion

Calculated values of isoconversional times at specific conversions have meaning of stabilities of samples and their corresponding intermediates and reflect mutual differences in kinetics of degradation of measured samples. The aim was to avoid any transformation of experimental data by linearization of functional dependences to obtain reliable values of isoconversional time since such approach leads to deformation of distribution errors, heteroskedasticity and a shift in the position in minima of the sum squares between experimental and calculated values [28].

Humic substances samples were chosen to cover perhaps all the possible sources and thus various chemical–physical character reflecting the conditions of their formation. They include relatively fresh raw material such as soil, aqueous systems such as river and peat and those that underwent partial processes of coalification, i.e., lignites and leonardites. The purpose of this work was not to establish any equations enabling calculation of elemental analysis data from thermogravimetric records but (i) to shed light on the processes of thermo-oxidative degradation of humified pool of natural organic matter; (ii) to search for an interrelationships and trends between primary (analytical) and secondary (physical) structure of humic substances and (iii) to search for the suitable temperature function providing data with the highest correlation coefficient. In this part we have focused on the application of the Arrhenius relationship as a temperature function $k(T)$.

Information on elemental composition, ash and free radical content are listed in Table 1. The contents of C, N, H and O are expressed as mass percentages. As expected, the highest C content possessed lignite and leonardite humic acids HS11 and HS15, respectively. In contrast sample of humin HS09, which was expected, in accordance with literature data, to have high content of C showed only an average value. The lowest C content showed soil humic acid HS13 and fulvic acids HS01, HS03 and HS07. N content in humic substances is regularly attributed to hydrolysable amino and imino groups [11] and therefore the "oldest" lignite and leonardite samples possessed relatively low values. Distribution of O in humic substances is connected with carboxylic groups, phenolic and alcoholic OH groups, ketones and chinones. That is the reason of highest content in fulvic acids samples, i.e., the most soluble humic substances fraction.

Table 2 reports the calculated isoconversional times (in years) for the set of humic substances consisting of 4 fulvic acids, 10 humic acids and 1 humin. Their respective coefficients of mutual linear correlations are given in Fig. 2. As can be seen in Fig. 2a correlation of isoconversional times with C content gave positive values of correlation while H, N, and O almost exclusively have negative values. The exceptions represent values at 70% of degradation for N and 90% for H and O. Nevertheless, those values are so small also for C that one can conclude that there is either no correlation between correlated parameters or, which is more probable, an unexpected experimental error occurred. The highest value (0.77) of linear correlation of kinetics of degradation showed carbon content at 10% and 30% and except of above-mentioned 90% of degradation, the values were spread within correlation coefficient 0.5–0.8 having a slow decreasing tendency with proceeding degradation. Relatively high correlation was also observed for O content. The highest value was determined for 10, 30 and 60% of degradation. Those correlation coefficients were obtained within the range 0.3–0.6 which indicates the destabilization effect of oxygen on overall humic substances structure stability mainly at the beginning of degradation processes. Content of N and H showed a weak influence on the stability of humic substances. The maximal linear correlation values

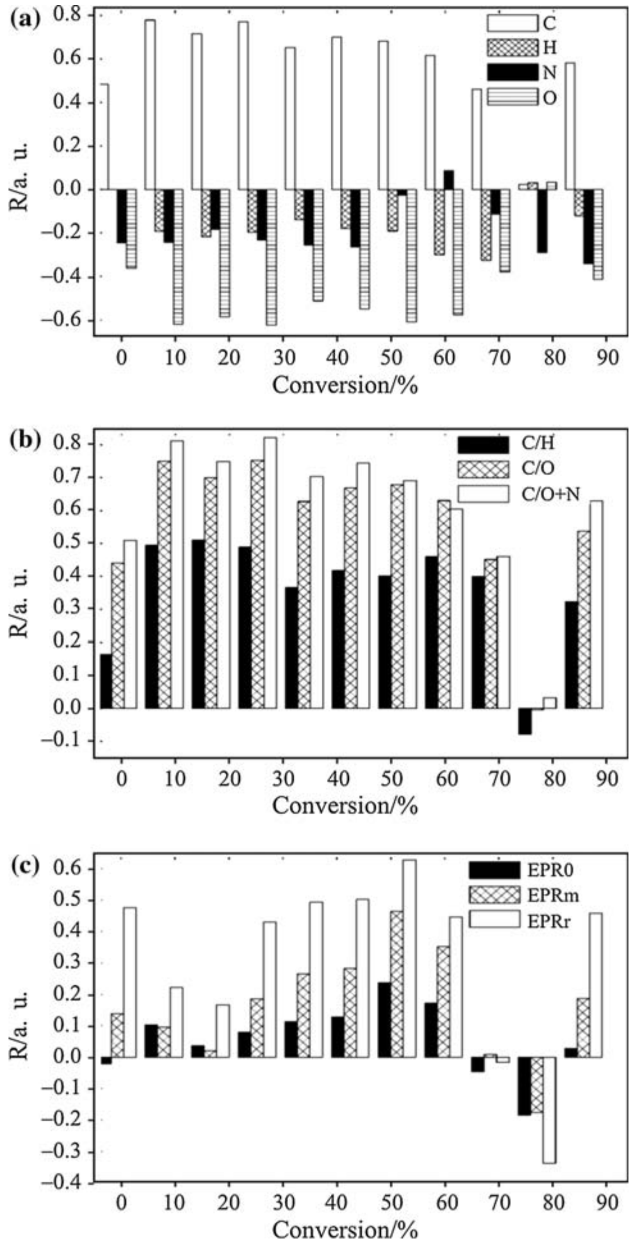

Fig. 2 Values of Pearson correlation coefficients between isoconversional times and **a** C, H, N and O contents, **b** ratios C/O, C/H and C/O + N and **c** free radical content (EPR0), generated free radical content (EPRm) and EPR0/EPRm (EPRr)

were 0.32 (80% of degradation) and 0.34 (at 100%) for H and N, respectively. The increase of the latter value by the end of degradation can be associated with the stability and degradation of heterocyclic compounds which are present in humic substances as a minority [11] and exhibit high thermo-oxidative stability [8]. Interestingly, the correlation between sum of O and N with isoconversional times brought high negative correlation coefficients varying from -0.43 to -0.74 which is higher than for O itself (graph not shown). Further correlations were carried out with the ash content but no significant result was observed (results not shown). Additional correlations were also carried out with C/O, C/H and C/(O + N) and some significant results were observed (Fig. 2b). In fact, C/O and C/(O + N) reflect the role of the organic carbon content in samples and C/H aromaticity degree of humic substances and they are frequently used as a distinguishing characteristics of soil, sediment and sedimentary rock [11]. A number of relatively great values of linear correlations can be identified in Fig. 1b. From 10 to 70% the values of linear correlation reached values within 0.6–0.81 while the value decreased with increasing conversion. At the same conversions, parameter C/O gave the highest correlation 0.63–0.75 as well. In contrast, C/H reached the correlation maximally 0.51 at 30% of degradation. This is in line with recent statements [4, 9] that the aromaticity alone is not the only indicator of humic substances stability. It seems that during the degradation of humic substances, the presence and distribution of polar groups play an important role. Probably, mutual weak interactions have more stabilizing effect than the chemical nature of present hydrocarbons.

Free radicals are thought to be a main factor influencing stability of various materials [35, 36]. In this work it was measured both the natural content of free radicals and content generated by the isothermal heating. Free radicals in humic substances are mainly of semi-chinoidal origin and their quantity is fluctuating in dependence on surrounding conditions and usually reported within 10^{16} to 10^{18} spin/g of humic substances [37]. Processes of heat-induced radical generation follow the natural processes and uncover the potential of accessibility of benzenediol-like structures in humic materials. Their most important role is associated with the sequestration of organic contaminants, stabilization due to interactions with other compartments, and in soil are thought to be a "shuttle" of electrons for soil microorganisms. In soils their content slightly decreases with the depth of the sample [38]. In earlier work [39] it was demonstrated that the spin concentration of the humic acids samples exhibits a highly significant positive correlation with mean annual rainfall in samples from Argentine Pampa.

The free radical content, generated radical content and their ratio were correlated with isoconversional times as in the case of elemental analysis. Obtained data showed mainly positive linear correlations exceptionally for EPR0 for conversion 0, 80 and 90%, at 80 and 90% for EPRr and at 90% for EPRm. In contrast the greatest values of linear correlations gave EPRr at 60% of humic substances degradation (0.63). Other values reached maximally 0.5 and showed rather scattered trend. Other parameters, especially EPR0 showed insignificant values of linear correlation.

Correlations at higher conversions were supposed to give less reliable data since the far-reaching extrapolations frequently give unrealistic values due to exponential form of Eq. 5 derived from the Arrhenius equation [28]. The trend of decreasing values of correlation coefficient with conversion can be partly identified in Fig. 2. Since the calculations were performed for 100 °C the highest correlation coefficients were obtained for conversions up to 30% of degradation. Nevertheless, after the increase of temperature in Eq. 5 to 200 °C and correlation with values 70, 80 and 100% conversion, no significant shift in correlation coefficient have been observed.

As stated previously, kinetic parameters A_k and E_a in Eqs. 2 and 3, respectively, are frequently interpreted as a quantity with a physical meaning. Recently it has been demonstrated that they do not have any mechanistic significance and represent only points on the kinetic hypersurface (i.e., dependence of conversion on temperature and time) [28]. In this work parameters A and B form Eqs. 1 and 2, respectively, were also correlated with basic properties of humic substances and unlike the isoconversional times, no significant correlations have been found (results not reported).

Conclusions

Generally it can be concluded that using the Arrhenius equation as a temperature function, parameters of elemental analysis (C and O content and ratios as C/(O + N) and C/O) are linearly correlated with the thermo-oxidative degradation kinetics/stability of humic substances. In contrast, H, N content and natural and heat generated free radical content and their ratio gave substantially lower correlation coefficients. The latter indicates that free radicals are probably not the main reason of humic substances secondary structure collapse leading to degradation. Although the correlation coefficients did not reach the highest possible values, in case of such a complex natural product as humic substances are, many obtained values were relatively high. Moreover, it should be taken into account that humic substances were collected from different sources, and extracted and purified using sometimes slightly different procedures.

Further, unlike the approach used by Siewert [15, 16], we used specific conversions (with an increment of 10%) which should not necessarily covered the ranges with maximal linear correlations. However, our aim was to point out the possible interrelationships which play role in thermo-oxidative stability of humic substances and find the right temperature function.

To sum it up, it seems that stability and degradation of C and O fixed in organic matter as humic substances obey the Arrhenius kinetics.

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