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Fractionation of Humic Acids upon Adsorption on Montmorillonite and Palygorskite

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Abstract—The adsorption of three humic acid (HA) preparations by clays—montmorillonite (Wyoming, USA) and palygorskite (Kolomenskoe district, Moscow oblast)—has been studied. The HA preparations were isolated from samples of the humus-accumulative horizons of a leached chernozem (Voronezh) and a chestnut soil (Volgograd), and a commercial preparation of sodium humate (Aldrich) was also used. The solid-state ^{13}C NMR spectroscopy and IR spectroscopy revealed the selective adsorption of structural HA fragments (alkyls, O-alkyls (carbohydrates), and acetal groups) on these minerals. As a result, the aromaticity of the organic matter (OM) in the organic−mineral complexes (OMCs) and the degree of its humification have been found to be lower compared to the original HA preparations. The fractionation of HAs is controlled by the properties of the mineral surfaces. The predominant enrichment of OMCs with alkyls has been observed for montmorillonite, as well as an enrichment with O-alkyls (carbohydrates) for palygorskite. A decrease in the C : N ratio has been noted in the elemental composition of the OM in complexes, which reflected its more aromatic nature and (or) predominant sorption of N-containing structural components of HA molecules. The adsorption of HA preparations by montmorillonite predominantly occurs on the external surface of mineral particles, and the interaction of nonpolar alkyl groups of HAs with this mineral belongs to weak (van der Waals, hydrophobic) interactions. The adsorption of HA preparations by palygorskite is at least partly of chemical nature: Si−OH groups of minerals are involved in the adsorption process. The formation of strong bonds between the OM and palygorskite explains the long-term (over 300 million years) retention of fossil fulvate-type OM in its complex with palygorskite, which we revealed previously.

Keywords: palygorskite, montmorillonite, humic acids, ^{13}C NMR spectroscopy, IR spectroscopy, organic-mineral complexes

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INTRODUCTION

It is known that 50−75% of the organic matter (OM) in the soils of the temperate zone is bound to minerals and is a part of organic−mineral (clay) complexes (OMCs) [33]. Organic−mineral processes participate in the formation of soil aggregates [3, 8, 15, 44] and largely determine the behavior of pollutants [18] and the fate of the soil OM by affecting the rate and evolution of its mineralization and protecting some of its components. The OM of OMCs forms passive (or stable) pools of soil organic carbon (C_{org}), whose turnover time was recently assessed at a few thousand years [27]. This fact explains the resistance of some humus components to diagenesis and their preservation in paleosols, including ancient fossil soils [2, 6]. The mechanisms of the OM's interaction with the mineral surfaces depend on the properties of the organic molecules and the mineral component. Three types of interactions are considered as the main interactions: (1) ligand exchange; (2) the formation of polyvalent cation bridges; and (3) hydrophobic, H−, and van der Waals bonds [32]. The ligand exchange

provides the maximum protection of the OM, and each soil has some total protecting function [27]. It is proposed to consider the number of OH groups in iron and aluminum hydroxides and silicates potentially able to participate in ligand exchange as a measure for the content of OM, which the soil can stabilize as organic−minerals complexes [26].

Another aspect of the organic−mineral interactions is the selective adsorption of humic substances by different minerals. In 1980, Aleksandrova showed that humic acids (HAs) and fulvic acids (FAs) adsorbed on bentonite have smaller molecular weights compared to the original preparations of humic substances [1]. A new tide of interest in studies in this field arose with the development of solid-state NMR spectroscopy, which allows directly studying the structure of organic molecules without their decomposition. Kogel-Knabner et al. [27] showed that the OM of soil OMCs is depleted of lignin and phenols and enriched with alkyls and O-alkyls. The content of aromatic structures in them is low, except for coalified OM. In loamy soils, the contents of polysaccharides and protein

structures are higher than those in sandy soils. The authors supposed that the features of the OM in complexes depend on the soil genesis and mineralogy. Other authors [29] revealed that the OM in association with separate soil subfractions differing in their mineralogical parameters (2–0.2, 0.2–0.02, and <0.02 μm) varies in composition. For example, fine colloids are enriched with monosaccharides, amino acids, and fatty acids. At the interpretation of the data, the authors advance two points of view: (1) clay minerals affect the humification of OM, and (2) the sorption of OM on clay minerals is accompanied by its fractionation. The first point of view is supported by Zavarzina [46]. The results of other authors for pure minerals showed that kaolinite and montmorillonite predominantly sorb aliphatic components of HAs, and Fe hydroxide (goethite) sorbs components containing carboxylic groups [20, 21, 41, 42, 45]. Some authors showed a difference in the affinity of nitrogen-rich and -poor components (amino acids, amino sugars, and lignin phenols) for mineral surfaces [34]. This results in a shift of the C : N ratio for the OM of OMCs.

This work continues our earlier studies [2, 4], in which we found that the fossil soils of the late Carboniferous time (about 300 million years old), which have palygorskite mineralogy, retain up to 1.5% C_{org} represented by fulvate-type humus. The study of these components by solid-state ^{13}C NMR spectroscopy showed that the complexes contain similar proportions of aliphatic (alkyls, carbohydrates) and aromatic structures. The composition of the OM in the revealed complexes radically differs from that of kerogen (the prevalent form of C_{org} in ancient sedimentary rocks having a predominantly aliphatic nature, as well as humin [5, 38, 42]). Paleosols of the same region and age having a smectite composition contain significantly less C_{org} (<0.5%), which is represented by only aromatic groups ("coal"). The revealed fundamental difference between the humus of the OMCs from the fossil soils of the same age but different mineralogy formed the basis for the present work.

In this work, the adsorption of three HA preparations (two of them were isolated from chernozem and chestnut soils, and the third was a commercial Aldrich preparation) on two clay minerals with fundamentally different structures and physicochemical properties (montmorillonite and palygorskite) was studied. The main aim of the work was to compare the chemical structure of the OM from the obtained OMCs with that of the original HA preparations. The studies were performed using direct methods without the OM's destruction using solid-state ^{13}C NMR and IR spectroscopy techniques. To elucidate the OM chemical structure of the natural OMC compared to the complexes prepared experimentally, the ^{13}C NMR spectra of the bulk sample and the clay fraction from the A horizon of the chestnut soil were also studied. Smectite and illite are the main clay minerals in the clay fraction from this soil.

OBJECTS AND METHODS

The experiments were performed with bentonite from the Wyoming field (USA), which represents almost pure Na-montmorillonite; it was converted to the Ca form by saturation with a 1 N $CaCl_2$ solution. The specific surface of the sample was about 800 m^2/g; its cation exchange capacity was 94 meq/100 g. The second tested material was palygorskite (magnesium-rich chain silicate). The palygorskite clay was sampled from a quarry near the town of Kolomna (the Vas'kino series of the Podol'sk horizon, late Carboniferous) and used without preliminary purification. Its external specific surface reaches 200 m^2/g, and its internal specific surface reaches 600 m^2/g due to the presence of channels in the zeolite-type structure. The cation exchange capacity is no higher than 20 meq/100 g; the anion exchange capacity reaches 70 meq/100 g [17]. The original clays contain traces of C_{org} (<0.1%), which is below the detection limits of the methods used in the work. The mineralogy of the original clays and OMCs was studied using X-ray diffraction by scanning with a step of 0.1° and an exposure time of 10 s (DRON-3 X-ray diffractometer, Cu radiation).

The preparations of HAs isolated from the samples of the humus-accumulative horizons of the leached chernozem (Voronezh) (the 1HA) and the chestnut soil (Volgograd) (the 2HA) were prepared by the procedure of alkaline extraction used in Russia [11] with some modifications recommended by the IHHS [16, 43]. In particular, after the separation from the FAs, the HAs were dissolved in a minimum volume of 0.1 M KOH, and KCl was added to a final K^+ concentration of 0.3 M for coagulating the finely dispersed mineral particles, which were separated by centrifugation. Next, the HAs were reprecipitated and treated with a 0.1 M HCl/0.3 M HF solution to remove the Si/Fe-containing impurities. The preparation was then washed with distilled water and centrifuged; the suspension was dried at 60°C. The content of ash was 30.2% in the 1HA preparation and 34.2% in the 2HA preparation. The third studied preparation (3HA) was a commercial HA preparation (sodium humate from Aldrich). This preparation was not purified. Its ash content was 42.2%. All three preparations were composed of humates. It is known that HAs are present in soils and interact with minerals as salts. Thus, the experiments described in this paper well simulate the natural conditions. In our previous publication [4], these preparations were referred to as HAs; to avoid confusion, this term is also used in this paper.

The adsorption isotherms were obtained under thermostated conditions (at 25°C); the duration of the experiments was 48 h. The highest concentration was 200 mg/L for the 1HA and 2HA solutions and 400 mg/L for the 3HA solution. All the HA preparations were dissolved in water; to stimulate the dissolution the solutions of 1HA and 2HA were first alkalized, and their pH was then adjusted to 7. The 3HA prepa-
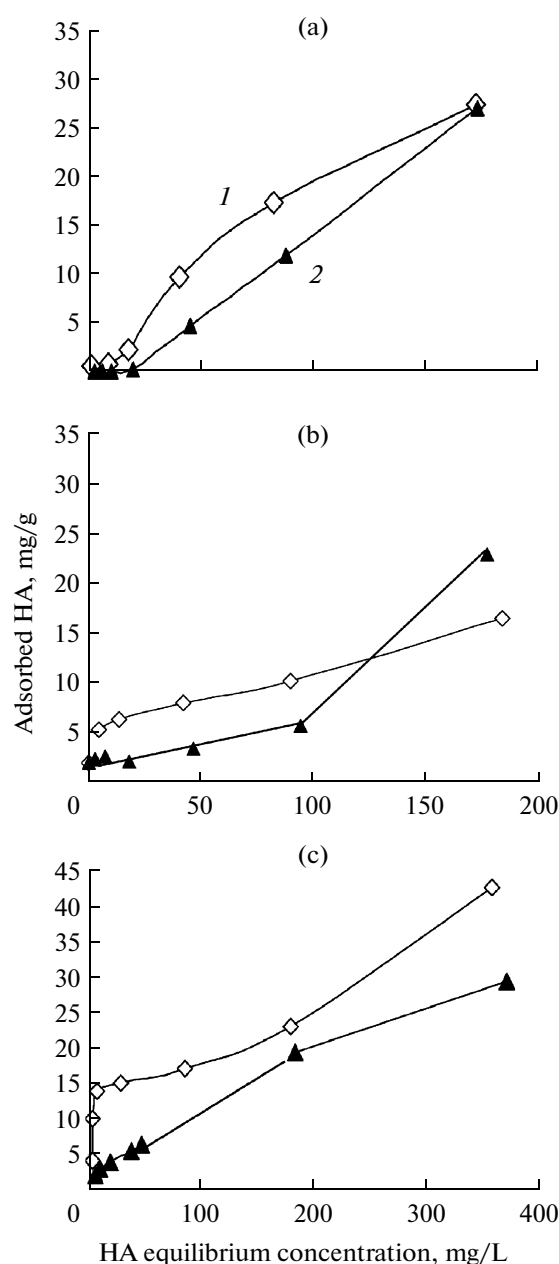
Fig. 1. Isotherms of the HA adsorption on the (*1*) palygorskite and (*2*) montmorillonite: (a) 1HA (Voronezh); (b) 2HA (Volgograd); (c) 3HA (Aldrich).

ration was easily dissolved in water without alkalization. Its pH was also adjusted to 7. The solid/liquid phase ratio was 50 mg of the mineral/50 mL of the HA solution. The pH values of the equilibrium solutions remained in the range of 6.8–7 during the entire experiment. After the end of the experiments, the solutions were centrifuged at 5000 rpm for 30 min. Then, their HA concentrations were determined from the absorption at 280 nm (on a Thermo Nicolet UV-VIS spectrophotometer). The content of the adsorbed HA was calculated from the difference between its concen-

trations in the original and equilibrium (after the end of the experiment) solutions. The precipitates on the tubes' bottoms were twice washed with water and dried at 40°C.

To elucidate the chemical OM structure of the natural OMC, the bulk sample and the clay fraction (<2 μm) from the A horizon of the chestnut soil prepared by sedimentation in distilled water were also studied.

The elemental composition of the HA preparations and the prepared OMCs corresponding to the maximum initial HA concentration was studied on an Elementar Vario EL III CNS analyzer. Their structural and group composition were studied by diffused reflection Fourier transform and near infrared (DR NIR FTIR) spectroscopy (a Nicolet 6700 spectrometer) and solid-state ^{13}C NMR spectroscopy (a Bruker Avance-II NMR 400 MHz instrument). Solid-state NMR spectroscopy is presently the best technique for studying organic–mineral interactions. It has some significant advantages over other methods, including IR spectroscopy, the main of which is the presence of only peaks from C-containing structural fragments in the ^{13}C-NMR spectra. The limitations of the method are related to the low content of C_{org} in the preparation and the small portion of the ^{13}C isotope (1% of the total C). The treatment of the samples with a 10% HF solution to remove the paramagnetic impurities increases their concentration of C_{org} [39]. In our case, the threefold treatment of the samples increased the concentration of C_{org} by three times on the average.

The chemical shifts are given relatively to tetramethylsilane. The distribution of the C atoms among the structural fragments was determined by the integration of the corresponding spectral regions. The following spectral assignments were made according to Kovalevskii [9]: 0–48 ppm, aliphatic carbon atoms; 48–100 ppm, aliphatic carbon atoms (mainly of carbohydrate fragments) bound to a heteroatom (oxygen or nitrogen) by a simple bond; 100–110 ppm, acetal carbon atoms bound to two oxygen atoms by simple bonds, mainly in cyclic polysaccharides; 110–160 ppm, C-, H-, and O-substituted aromatic carbon atoms; 160–190 ppm, carbon atoms of carboxyl, ester, and amide groups; and 190–230 ppm, carbon atoms of ketone and quinone groups. From the aromaticity index calculated as $[AR/(AR + AL)] \times 100\%$ (where AR and AL denote the aromatic and aliphatic structures, respectively) and the alkyl/O-alkyl and aryl/O-aryl C ratios [37], the degree of the OM's humification in the preparations was calculated.

RESULTS AND DISCUSSION

Adsorption of HA preparations on clay minerals. The adsorption isotherms of the HAs on the minerals are given in Fig. 1. For the 1HA preparation (chernozem, Voronezh), the adsorption isotherm on the palygorskite is a combination of the S + L isotherms, and that of the montmorillonite is of the S type [13, 23]. A dif-

Table 1. HA content sorbed on minerals at the maximum initial HA concentration (200 mg/L for 1HA and 2HA and 400 mg/L for 3HA) and the elemental composition of the HA preparations (% of the ashfree substance) and the HAs in the studied OMCs

| Sample	Sorbed HA, mg/g mineral (mg/m^2 surface area)	C	N	S	C/N
		%			
1HA (chernozem)		47.1	3.7	6.00	14.91
1HA + montmorillonite	27(0.12)	1.20	0.12	0.36	11.80
1HA + palygorskite	27(2.17)	0.56	0.11	0.26	5.96
2HA (chestnut soil)		35.7	4.4	6.83	9.55
2HA + montmorillonite	23(2.14)	0.86	0.12	0.25	8.39
2HA + palygorskite	17(6.42)	1.21	0.18	0.37	7.87
3HA (Aldrich)		59.2	1.00	4.58	67.84
3HA + montmorillonite	29(5.42)	3.29	0.07	0.40	54.99
3HA + palygorskite	43(14.96)	3.92	0.12	0.49	38.22

ferent situation was observed in the experiments with the two other HA preparations, for which the adsorption isotherms on the palygorskite are of the H type (high affinity) and those on the montmorillonite are of the S and L types. It is noteworthy that all the adsorption isotherms are clearly divided into two parts: the initial part, where the active sites of the minerals with the highest affinity for HAs are occupied, and the final part (the three last points), where the HAs are adsorbed on the mineral or organic sites with lower affinities. On the initial fragments of all three isotherms, the palygorskite has an appreciably higher adsorption capacity than the montmorillonite. The amounts of HAs sorbed on the minerals for the maximum initial concentration of HAs are given in Table 1. The estimation of the content of the sorbed HAs per unit of external mineral surface area for the initial fragment of the experimental isotherms showed that the adsorption capacity of palygorskite exceeds that of montmorillonite by 72 times for 1HA and by 11–12 times for the 2HA and 3HA. The calculations were performed per unit of the external mineral surface area, because the experimental data showed no HA adsorption on the internal surfaces of the minerals (see below and Fig. 2).

It should be emphasized that the adsorption of the HA preparations did not reach saturation (a plateau) and was limited by the solubility of the HA preparations in all six experiments. For comparison, in the experiments of Singer and Huang [40], the adsorption isotherm of the humate on palygorskite is of the L type and reaches a plateau almost at the beginning of the experiment. The adsorption capacity of palygorskite obtained in these experiments was comparable to that determined in our experiments an the adsorption of

the 3HA preparation on this mineral (41–46 mg of HA/g). The sorption capacity of the montmorillonite from the Askania field, which was experimentally determined by Zavarzina and Demin [7], is comparable with our values, as well as the predominant isotherm type (L).

Our results showed that all three HA preparations have a higher affinity for palygorskite (predominant H type isotherms) and that the adsorption of HAs on palygorskite is generally higher than on montmorillonite. In our opinion, the observed difference is due to the fundamental differences between the surface properties of the studied minerals, namely, the high anion exchange capacity of palygorskite and the related capacity for sorbing not only cations but also anions and neutral molecules. Another structural feature of palygorskite, which can explain the observed results, is the presence of numerous OH groups occupying different positions in the mineral structure and characterized by different accessibilities and reactivities [17, 24].

X-ray diffraction analysis of the minerals and OMCs. The X-ray diffraction patterns of the studied clay minerals and OMCs corresponding to the maximum initial concentration of HAs (200 mg/L for 1HA and 2HA and 400 mg/L for 3HA) are given in Fig. 2. The X-ray diffraction patters of the original palygorskite and its complexes with HAs for the air-dry and ethylene glycol-saturated specimens are absolutely identical. The channels in the palygorskite's structure are 6.4 × 3.7 Å in size; along with the exchangeable cations, zeolite water molecules occur there [24]. Therefore, it is hardly expectable that the HA molecules are able to occupy nanochannel structural units; they are most probably adsorbed on the external surface of the crystallites (probably, at channel openings). The X-ray
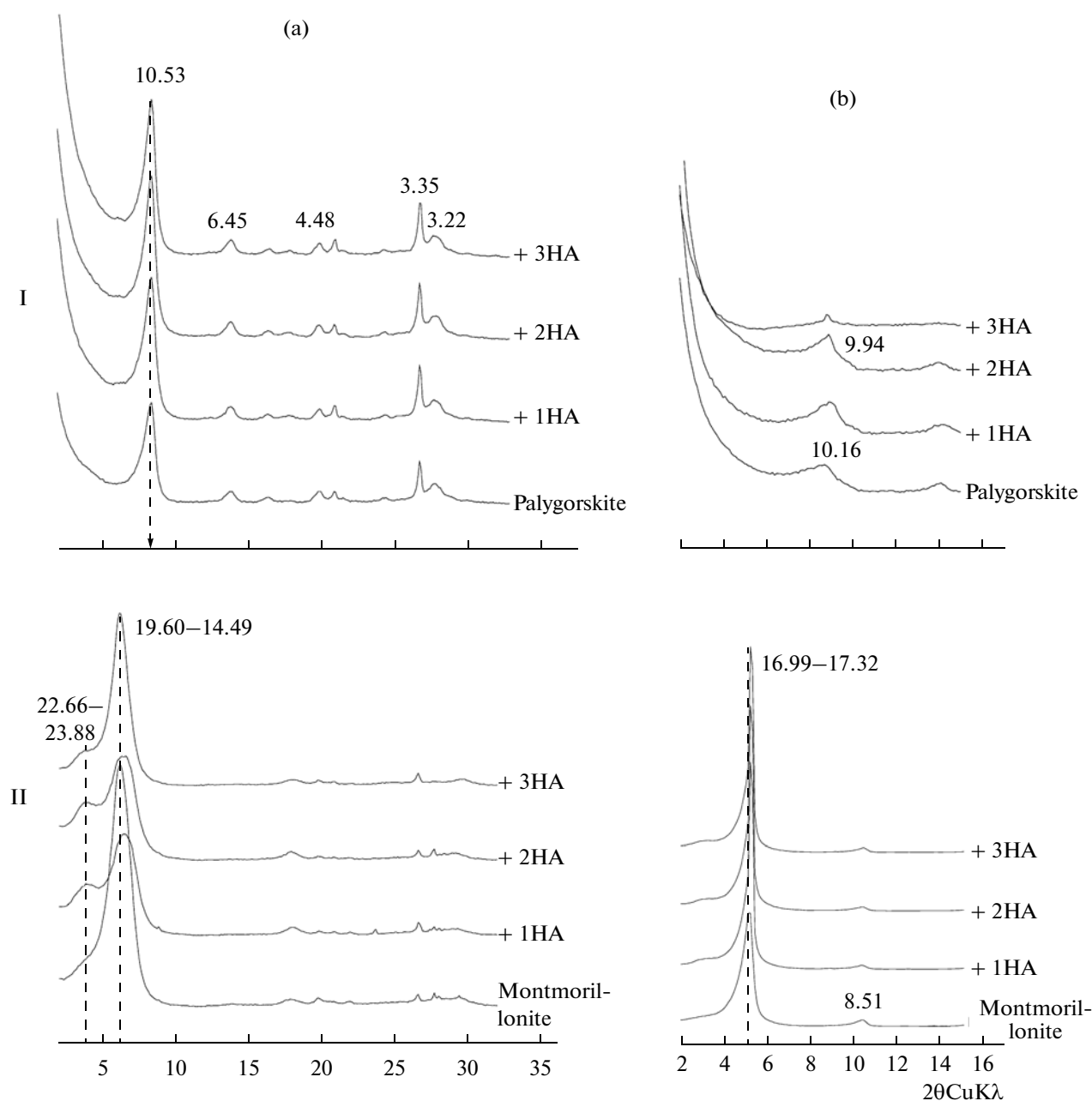
Fig. 2. X-ray diffraction patterns of the (I) palygorskite clay, the (II) Ca-montmorillonite, and the obtained OMCs corresponding to the maximum initial concentrations of the HA preparations (200 mg/L for 1HA and 2HA and 400 mg/L for 3HA): (a) the initial preparations; (b, part I) after calcination at 550°C; (b, part II) after saturation with ethylene glycol.

diffraction patters of the OMCs calcined at 550°C are characterized by a shift of the reflections from 10.16 to 9.94 Å for all the preparations and a significant decrease in the reflection intensity for the palygorskite + 3HA preparation. The obtained data can be indicative of the decreased thermal stability of the OMCs compared to the original palygorskite.

The X-ray diffraction patterns of the montmorillonite complexes with the air-dry HAs somewhat differ from that of the initial Ca-saturated montmorillonite. A shift of the (001) reflection toward lower angles is

observed, which is related to the insignificant Ca–Na exchange during the experiments on the HA adsorption. The cation exchange is insignificant, and the montmorillonite predominantly remains in the Ca-saturated form. We attribute the decrease in the intensity of the 14 Å reflection in the X-ray diffraction patterns of the complexes and the appearance of a small peak at 23–24 Å to the dispersion of the montmorillonite crystallites and the partial conversion of the montmorillonite to the superdispersed state. This phenomenon is well explainable taking into consideration that

Table 2. Degrees of aromaticity and humification of the OM in the HA preparations, the obtained OMCs, and the chestnut soil (the bulk sample and clay fraction) from the ^{13}C NMR spectroscopy data

| Sample	OM aromaticity, %	Alkyl C/O-alkyl C	Aryl C/O-alkyl C
1HA (chernozem)	39.24	1.51	1.62
1HA + montmorillonite	31.47	1.35	1.08
1HA + palygorskite	21.06	0.46	0.39
2HA (chestnut soil)	34.89	1.25	1.21
2HA + montmorillonite	25.70	1.45	0.95
2HA + palygorskite	30.06	0.85	0.84
3HA (Aldrich)	25.40	17.17	6.19
3HA + montmorillonite	18.84	5.95	1.61
3HA + palygorskite	15.53	6.83	1.44
Chestnut soil			
A1 horizon, bulk sample	22.02	0.58	0.45
A1 horizon, fraction <2 μm	19.32	0.64	0.39

the mineral was intensively stirred in an Na-containing medium for 48 h of the experiment. The absence of crystallochemical transformations of the montmorillonite's structure, including the penetration of HAs into the interlayer space, is obvious from the behavior of the OMCs saturated with ethylene glycol: the position of the (001) reflection corresponds to that for Ca-montmorillonite.

The structural and group composition of the HA preparations and its modification during their adsorption on clay minerals based on the ^{13}C NMR spectroscopy data. The results of studying the elemental composition of the HA preparations and the OMCs at the maximum initial concentrations of the HAs are given in Table 1. It can be concluded that a general tendency for decreasing of the C : N ratio in the OMCs compared to the HA preparations was observed for both minerals, although it was more typical for the complexes with palygorskite. The obtained results can indicate a more aliphatic character of the OM in the OMCs and (or) an affinity of the N-containing groups for the surfaces of the clay minerals. Many authors noted that the OM of natural OMCs is characterized by low C : N ratios compared to the soil OM as a whole [31]. In this context, the results of characterizing the properties of the OM in the fractions <20 μm from different soils before and after their treatment with a 10% H_2O_2 solution are demonstrative [30]. It was found that the C loss was significantly higher than the N loss in this case, and the C : N ratio appreciably decreased after the treatment. These results are also indicative of the better retention of N-containing organic components by the mineral surfaces. According to the available

data, about 22% of the C_{org} in the HAs is of polypeptide nature [25]. Amino acids are known to have an extremely high affinity for mineral surfaces, which is related to the wide spectrum of properties of protein molecules: the charge varies from negative to positive values, and the molecules can be both polar (hydrophilic) and extremely hydrophobic ones [36]. From these data, a special affinity of nitrogen-rich components for the studied clay minerals can be supposed. This phenomenon can be favored by the experimental pH values (7−8), which allows the dissociation of NH_2 groups in HAs.

The ^{13}C NMR spectra of the HA preparations are given in Figs. 3a−c. The contents of ^{13}C nuclei in the structural fragments of the organic HA molecules are given in Fig. 4. The 1HA and 2HA preparations have largely similar parameters: the contents of aliphatic and aromatic structures, 28−31%; the contents of O-alkyls (48−100 ppm), 16−20%; and the content of carboxylic groups (160−190 ppm), 16−17%. The calculated degrees of humification of the HAs are given in Table 2. They indicate that the degree of humification of the OM in the 1HA preparation isolated from the chernozem is higher than that in the 2HA preparation isolated from the chestnut soil. The 3HA preparation is strongly different from the other preparations by the predominance of aliphatic structures (60%), low aromaticity (25%), and low degree of OM humification.

The ^{13}C NMR spectra of the mineral complexes with 1HA and 2HA (OMCs) are given in Figs. 3a and 3b; the content of C nuclei in the structural fragments of their organic molecules are given in Fig. 4. The results show that the ratio between the HA structural frag-
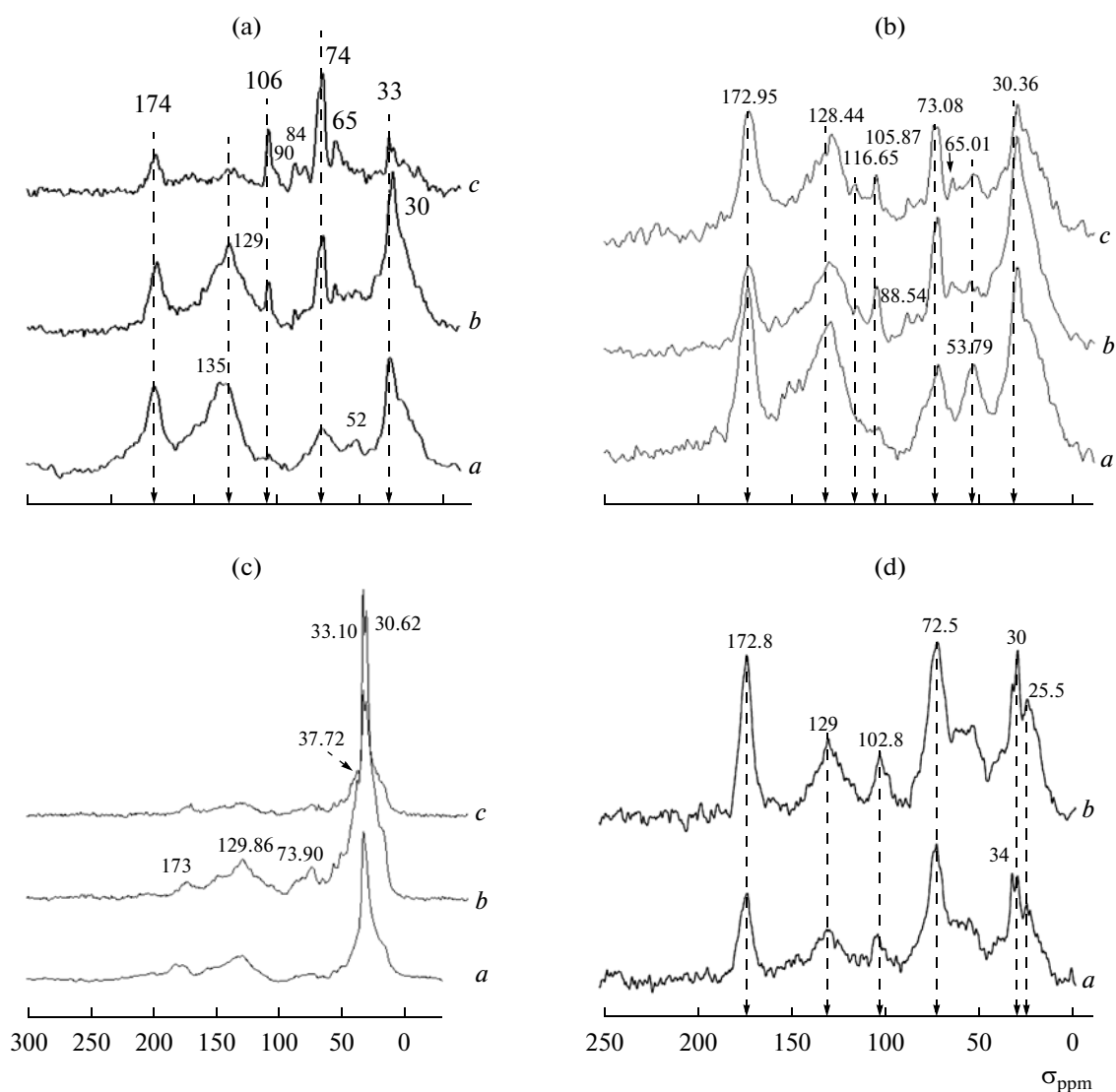
Fig. 3. ^{13}C NMR spectra of the HA preparations isolated from the (a) chernozem, 1HA; the (b) chestnut soil, 2HA; (c) the commercial preparation, 3HA: (*a*) the initial sample, (*b*) the complex with montmorillonite, and (*c*) the complex with palygorskite; and (d) the A1 horizon of the chestnut soil: (*a*) the bulk sample, (*b*) the clay fraction.

ments' changes (i.e., the fractionation of the HAs occurs) during the adsorption of the 1HA and 2HA preparations on both minerals. The main tendency revealed for both minerals is a decrease in the content of aromatic structures (in the spectral region of 110–160 ppm), as well as an increase in the contributions of alkyls (0–48 ppm), O-alkyls (48–100 ppm), and O-containing acetal groups (100–110 ppm). The increase in the intensity of the signal at 73 ppm (the main band assigned to carbohydrates) is accompanied by the appearance of low-intensity peaks in the regions of 63–65 and 80–90 ppm, which are also assigned to carbohydrates. The presence of lignin is insignificantly manifested in the spectra of the initial preparations (at 52–54 ppm) and appreciably decreases in the spectra of the OMCs. In the experiments with 2HA, an

enhancement of the signal at 116 ppm corresponding to protonated aromatic structures is observed (Fig. 3b).

The above-noted tendencies in the structural and group composition of the OM are typical for the complexes of both minerals with 1HA and 2HA. At the same time, the role of carbon from long-chained polymethylenes (about 30 ppm) is more enhanced in the complexes with montmorillonite, and the role of carbon from O-alkyls (carbohydrates) is higher in the complexes with palygorskite. In the experiment with 1HA + palygorskite, the portion of these compounds increases to 41% compared to 16% in the initial 1HA preparation. Another specific feature of the palygorskite complexes with 1HA and 2HA is the presence of some additional weak signals in the region of 15–25 ppm. We believe that this feature can be related to the increase in the contribution of short-chained poly-

methylenes and the appearance of methyl groups belonging to, e.g., protein or wax molecules [19, 20]. These signals are absent in the spectra of OMCs with montmorillonite. The selectivity for O-alkyls (carbohydrate) molecules is manifested at the adsorption of 1HA and 2HA on palygorskite, but it is not observed in the experiments with montmorillonite. At similar contents of O-alkyls (carbohydrates) in the initial 1HA and 2HA preparations (16 and 19%, respectively), the palygorskite complex with 1HA is enriched with carbohydrates by 25% and that with 2HA by only 3% compared to the initial preparations. We relate this fact to the differences in the qualitative composition of the carbohydrate components of these HAs and the different affinities of palygorskite for them. The selectivity of the mineral surfaces with respect to different carbohydrates was emphasized in some works, in which the predominant sorption of monosaccharides (mannose and glucose) by ferrihydrite (Fe hydroxide) [35] and the enrichment of the fine soil colloids (<0.02 μm) with monosaccharides were noted.

An increase in the portion of alkyls (0−48 ppm) is also observed at the adsorption of the commercial 3HA preparation on both minerals (Fig. 3c). A specific feature of the spectra in the region of 30 ppm is notable for the complexes of the 3HA preparation with both minerals: the appearance, along with the peak at 33 ppm, of a second intensive peak at 31 ppm. The former peak is assigned to the "crystalline" (stable) domains of long alkyl chains, and the latter peak is assigned to their "amorphous" (mobile) domains. In the spectra of the OMCs, an additional peak at 38 ppm is observed, which is assigned to the CH_2 groups of branched chains [42].

Thus, the OM in the complexes obtained by the adsorption of the HA preparations on montmorillonite and palygorskite is characterized by a lower aromaticity and degree of humification compared to the initial HA preparations and acquires features more typical for FAs.

The study of the structural and group composition of the OM in the entire soil sample from the A1 horizon of the chestnut soil and its clay fraction by ^{13}C NMR spectroscopy (Fig. 3d) showed that the OM of the clay fraction (the natural OMC) contains more alkyls compared to the entire sample (27 and 25%, respectively) and is characterized by lower aromaticity (19 and 22%, respectively) and a higher content of carboxyl carbon (13.5 and 11%, respectively). The additional peak at 34 ppm in the spectrum of the clay fraction indicates an increase in the contribution from the stable domains of long alkyl chains [42]. The enrichment with alkyls was also observed in the experiments on the adsorption of HAs by clays. The analysis of the natural samples shows that the aliphatic structures of the OM have higher affinity for the surfaces of clay minerals, which are the main components of the clay fraction in soils.
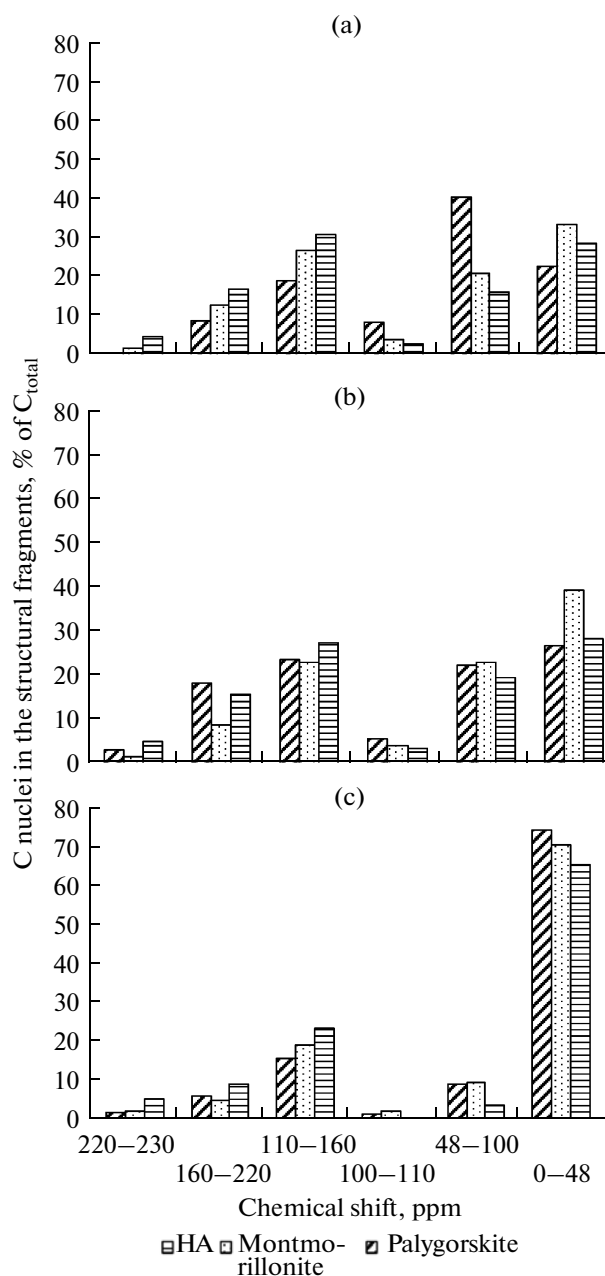
Fig. 4. Content of ^{13}C nuclei in the structural fragments of the HA molecules—(a) 1HA, (b) 2HA, (c) 3HA—and their complexes with minerals from the ^{13}C NMR spectroscopy data.

Other authors also noted an increase in the portion of aliphatic fragments in the OM adsorbed on clay minerals. Kholodov et al. [16] found for gray forest soil that the portion of aliphatic fragments and carbohydrates increased and the portion of aromatic fragments decreased in successive alkaline extracts (an approximation of the extraction of humic substances of proper OMCs). Travnikova and Titova [14] noted an increase in the content of FAs in the colloidal fraction from solonetzic horizons compared to the fine silt fraction,
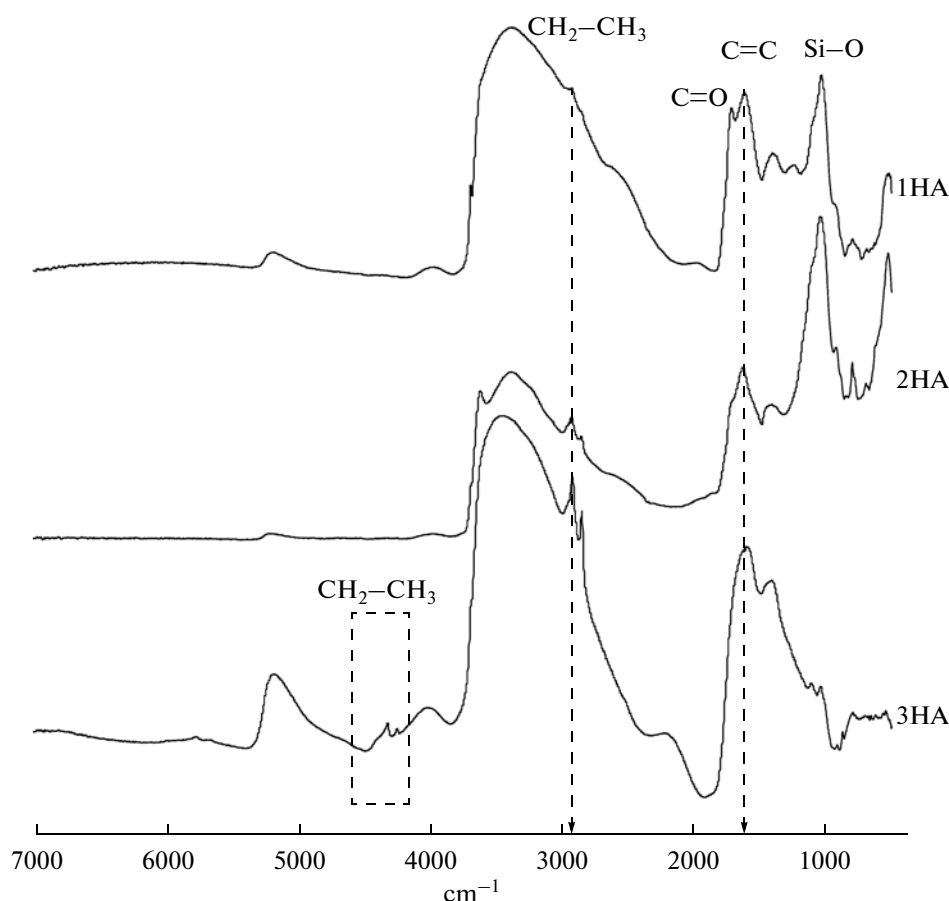
Fig. 5. IR spectra of the HA preparations.

where HAs are predominant. The authors explain this phenomenon by the predominance of montmorillonite-type minerals in colloids.

Mechanisms of the HA−mineral interaction from the IR (DR NIR FTIR) spectroscopy data. The IR spectra of the preparations are given in Fig. 5. The spectra of the 1HA and 2HA preparations are similar; they are characterized by the presence of the following basic bands (cm^{-1}): 2928 and 2852 (CH$_2$ and CH$_3$), a shoulder at 1714 (C=O), 1630 (aromatic C=C), 1388 (OH, COO−, and CH), and 1034 (Si−O and alcohol OH) [10, 12]. Taking into account the high ash content in the HA preparations, we cannot exclude the superposition of silicate and HA bands in the spectra. This is primarily the case for the regions at 1630 and 1030 cm^{-1}.

The IR spectrum of the 3HA preparation is strongly different. The aliphatic CH$_2$ and CH$_3$ groups (2920 and 2850 cm^{-1}) are more manifested and have overtones at 4340 cm^{-1}. Basic bands occur at 1582 and 1388 cm^{-1} and are assigned to Na carboxylate (COO−Na). The weak peaks in the region of 1100−1030 cm^{-1} are assigned to the OH groups of alcohols and the Si−O groups of silicates.

The spectra of the montmorillonite−HA complexes contain three additional bands in the region of 2960−2950 cm^{-1} (CH$_2$−CH$_3$ groups) compared to the spectra of the original montmorillonite (Fig. 6a). The position and intensity ratio of the bands at 2928 and 2852 cm^{-1} are identical to those in the spectra of the HA preparations. The band at 2962 cm^{-1} assigned to the CH$_3$ group is detected only in the spectra of the OMCs and is not manifested in the spectra of the HAs. No other distinctions between the spectra of the original montmorillonite and montmorillonite−HA complexes were revealed. On the basis of the revealed features of the OMC spectra, we believe that the CH$_3$ groups of the HAs have a higher affinity for the mineral surfaces compared to the other groups, including the CH$_2$. The interaction of nonpolar alkyl groups of HAs with montmorillonite is a weak (van der Waals, hydrophobic) interaction.

The spectra of the palygorskite−HA complexes are given in Fig. 6b. As in the previous case, the spectra contain three additional bands in the region of 2960−2950 cm^{-1} (CH$_2$−CH$_3$ groups), and a band at 2962 cm^{-1} assigned to the CH$_3$ group is detected only in the spectra of the OMCs. The second-derivative spectra presented in Fig. 7 show the visible differences between the spectra of the OMCs and the original palygorskite in the region of 3700 cm^{-1} (Si−OH) [22]. On the spec-
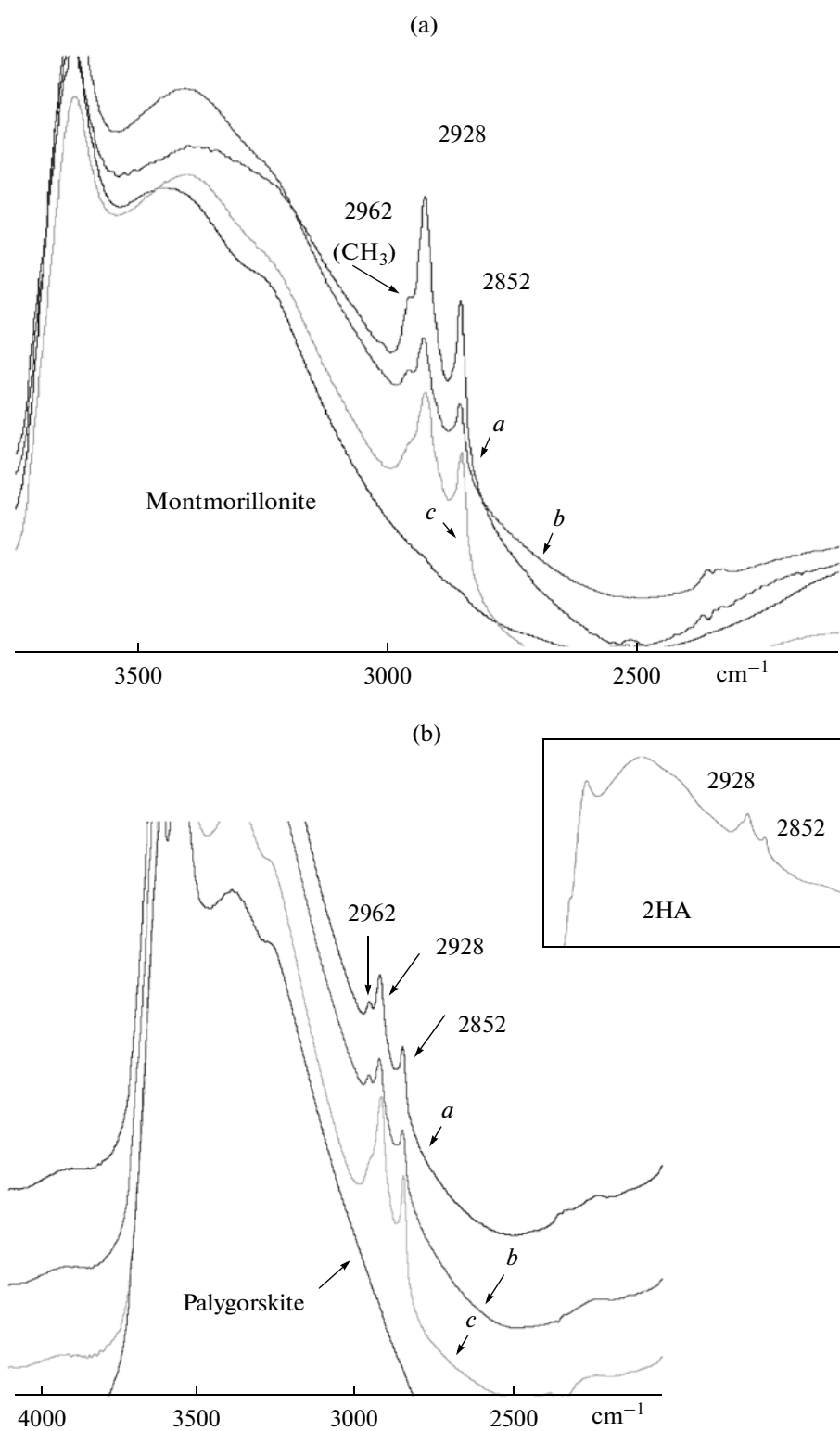
Fig. 6. The region of 2500–3500 cm^{-1} in the spectra of (a) the montmorillonite and (b) the palygorskite and their complexes with the preparations: (*a*) 1HA, (*b*) 2HA, (*c*) 3HA; (inset) the spectral region for the 2HA preparation.

(a)

Palygorskite

3708

3688

Palygorskite + 1HA

3702 cm^{-1}

(b)

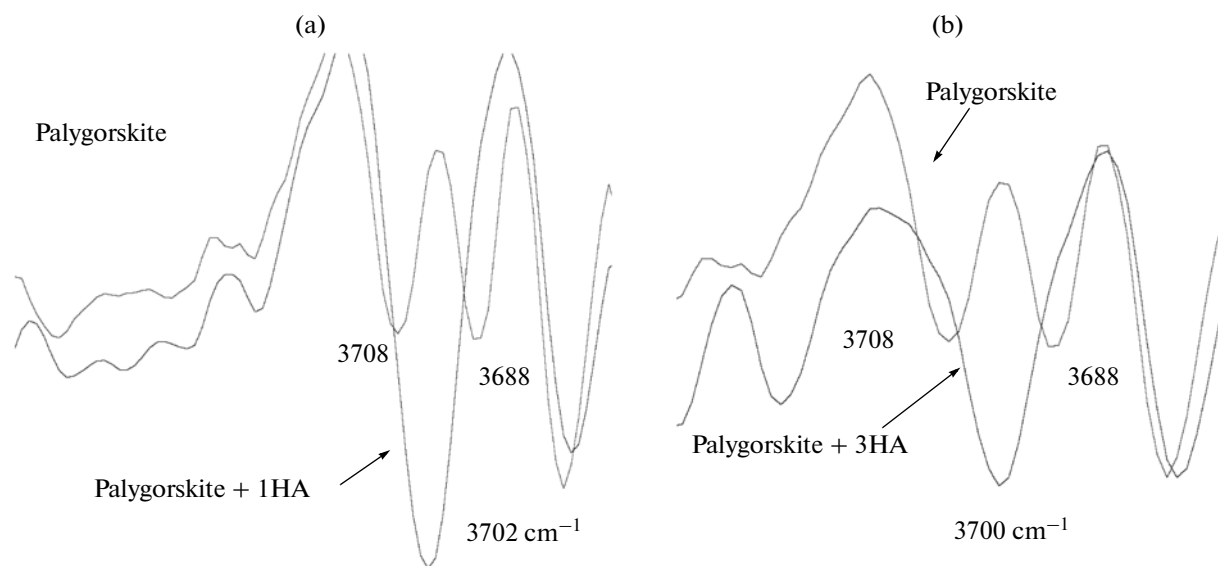Palygorskite

3708

3688

Palygorskite + 3HA

3700 cm^{-1}

Fig. 7. Second-derivative IR spectra of the palygorskite and its OMCs with (a) the 1HA and (b) the 3HA.

tra of the complexes, the intense band at 3700 cm^{-1} displaces two bands of lower intensities (at 3708 and 3688 cm^{-1}) typical for the palygorskite spectrum. The palygorskite structure is characterized by the presence of OH groups occupying different positions, including Si–OH groups. The content of the latter groups is estimated at 8% of the total Si surroundings from the ^{29}Si NMR data [28]. From our results, we believe that, in the experiments with palygorskite, along with the weak (van der Waals, hydrophobic) interactions of nonpolar alkyl groups, HAs also form strong chemical bonds with this mineral, which involve the Si–OH groups of palygorskite and, most probably, the O-alkyls of HAs.

CONCLUSIONS

Our study showed that palygorskite sorbs greater amounts of HAs than montmorillonite. The studied HA preparations have a higher affinity for palygorskite, which is confirmed by the predominant H type of adsorption isotherm. The X-ray diffraction data show that the HAs preparations are rather adsorbed on the external surface of the montmorillonite particles. In the interaction of the HA preparations with montmorillonite and palygorskite, the selective adsorption of the separate HA structural fragments was revealed. From the NMR data for the maximum initial concentration of HAs (200–400 mg/mL), an enrichment of OMCs with alkyls, O-alkyls (carbohydrates), and acetal groups, as well as a decrease in the portion of aromatic structures, was revealed. Therefore, the aromaticity of the OM in the OMCs and its degree of humification are lower compared to the initial HA preparations, which imparts features of FAs to the OM of OMCs.

The selective character of the sorption also affects the elemental composition of the humic substances in the OMCs: e.g., a decrease in the C : N ratio is noted, which reflects the more aliphatic nature of the OM in the OMCs and (or) the affinity of the N-containing groups for the mineral surfaces.

It has been found that the fractionation of the OM is controlled by the properties of the mineral surfaces. In experiments with palygorskite, a predominant sorption of O-alkyls is observed and signals of carbon from methyl groups probably of proteins and waxes are detected. Montmorillonite primarily sorbs alkyls (linear polymethylenes). The interaction of nonpolar alkyl groups of HAs with minerals is a weak (van der Waals, hydrophobic) interaction. Along with these assumed weak interactions, palygorskite can also form strong chemical bonds with HAs due to the involvement of Si–OH groups in the adsorption process, as was evidenced by IR spectra. The formation of complexes with the participation of Si–OH groups can explain the high resistance of OM to mineralization. A demonstrative example can be the unsurpassedly stable blue pigment of the ancient Mayas. The pigment includes plant indigo dye (*Indigofera suffruticosa*) and palygorskite [24].

Our previous studies [2, 4] showed the preservation of fossil fulvate-type OM in a complex with palygorskite 300 million years old.

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