Formation of soil aggregates via clay flocculation with organic polyelectrolytes

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ABSTRACT

Soil organic matter (SOM) plays a leading role in the formation of the soil structure. However, the mechanisms of that formation are still debatable. We proposed that the combined action of two types of SOM - labile polyelectrolytes released by soil biota (flocculant) and highly transformed polyelectrolytes - humic substances (modifiers of soil mineral particles) has a synergetic effect and may be a key process that leads to the aggregation of mineral particles in soils. Humic substances adsorbing on mineral surfaces play a role of anchors that link clay particles with a flocculant and facilitate the aggregation. Different mechanisms of SOM interaction with minerals are considered. Our hypothesis was confirmed by rheological studies of model clay and sand systems with/without addition of humic acids and chitosan (flocculant). Comparison of model systems with real soils indicated the role of OM as well as other factors (clay content, humidity) in the soil aggregates formation.
Keywords: *soil structure, flocculation, clay, humic substances, chitosan.*

1. INTRODUCTION

Soil structure is defined as arrangements of organo-mineral particles and cations forming aggregates of different size and stability [1,2]. The stability of micro- and macroaggregates depends on the contacts formed inside the aggregate as well as interaction between the aggregates. That is determined by various factors (the amount and type of SOM, soil minerals, inorganic cations, oxides, the type and size of the microbial population, the specificity of environmental conditions and management practices [3-8].

Nowadays soils are susceptible to degradation which manifests itself in the breakdown of aggregates and an increase in the amount of fine grained fractions. The use of polyelectrolytes may be applied as an alternative remediation for disturbed soil [9]. However, some of these polyelectrolytes can inhibit the activity of soil microorganisms, therefore, the study of effective eco-friendly natural substances as well as factors affecting the binding of soil particles are topical issues nowadays [10-11].

One of the key factors that affects formation of soil structure is SOM, a large part of which is provided by humic substances (HS) [12-15]. Different strategies have been used to analyze the mechanism of HS influence on the formation of the aggregates: from the Gedroits' coagulation theory to an approach in which soil is considered as a Pickering emulsion where the water represents the internal phase stabilized by clay particles, modified by HS [16].

There is a new approach in which HS is considered as parts incorporated into a periodic colloidal structure reinforced by clay particles [17]. There is a hypothesis of soil structure formation based on the amphiphilic properties of soil organic matter, hydrophilic components of which bind with mineral particles and whose hydrophobic components are bound with one another to generate water-stable organo-mineral aggregates in aqueous environment [18-21]. A large number of studies are devoted to the understanding of the role of microorganisms in aggregate formation processes [22-25]. Organic substances released by soil biota, so called "young" or "fresh" SOM (polysaccharides, amino acids, lipids) at the very beginning of OM transformation, as a rule, are labile, multifunctional and able to the interaction with surfaces of soil mineral particles and other organic molecules. Under the influence of biogeochemical factors labile compounds transform into highly condensed organic complexes of humic substances Milanovskiy and Shein [26].

All these theories indicate the role of SOM in the formation of soil structure, but up to now, there has been no commonly accepted theory of the role of the mobile components of SOM. Our approach is based on the different behavior of SOM depending on their nature. We have found that a synergetic effect takes place when "young" and transformed SOM act together binding soil mineral particles. Rheological studies were applied to examine the impact of SOM on the soil aggregation.

The purpose of our work was to study the soil formation mechanism via flocculation of soil minerals due to the combined action of different types of SOM: labile polyelectrolyte released by soil biota (flocculant) and recalcitrant humic substances (modifiers of mineral particles).

2. MATERIALS AND METHODS
2.1. Materials.

To simulate mineral particles, the following components were used: 100% montmorillonite of the Jebel deposit, >50% of the <1 μm fraction, density 2.45 g/cm^2, pH of aqueous extract was 7.9; kaolinite of the Chelyabinsk deposit with mineralogical composition: quartz 6%, kaolin 92%, muscovite 2%, density 2.66

g/cm^3, pH of aqueous extract 6.9, average grain size ~1 μm, and sand of Lubertsy deposit with mineralogical composition: SiO_2 98.08%, Al_2O_3 0.71%, Fe_2O_3 0.36%, MgO 0.18% and losses on ignition 0.6%; quantity of grain with diameter from 1,0 to 0,5 mm – 11.2%, and with diameter 0.5 to 0.05 mm – 88.4%.

Chemical composition of montmorillonite is as follows: HCO_3^- - 0.09; Cl^- - 7.92; SO_4^{2-} - 14.41; Ca^{2+} - 2.68; Na^++K^+ - 19.91; Mg^{2+} - 1.14; dry residue,% - 1.99 ; for kaolinite: HCO_3^- - 0.13; Cl^- - 0.08; SO_4^{2-} - 0,41; Ca^{2+} - 0.01; Na^++K^+ - 0.37-0.02; Mg^{2+} - 0.10; dry residue,% - 0.04.

Natural polysaccharide chitosan (Ch) (Mw 300, 240 and 83 kDa, degree of deacetylation 85%, manufactured by Bioprogress Co., Shchelkovo, in chloride form) was used as a model flocculant. Potassium salt of humic acid (HA) separated from brown coal (leonardite) produced by Powerhumus, Humintech Ltd., Germany was used as a model HS. The main characteristics of HA measured by ^{13}C NMR spectroscopy are: elemental ratios - H/C 0.84; O/C 0.38; C/N 72.7; molecular weight characteristics - Mw, kDa 6.9; Mn, kDa 0.6; Mw/Mn 11.5; structural groups: - aliphatic 15, O-aliphatic 7; aromatic 47; O-aromatic 12; carboxylic 13; carbonylic 5 (%) [27]. The investigated soil was sod-podzolic soil (according to WRB - Albic Glossic Retisols (Loamic, Cutanic), Corg - 1.5% [28], sampled from Pushkino district of Moscow region which is situated in the temperate zone; this area belongs to the Smolensk–Moskva erosional moraine elevation in the central part of the East European Plain. Mantle (covering) silty loams serve as the parent material.

2.2. Methods.

2.2.1. Investigation of clay characteristics.

Rheological parameters of clay pastes were determined by variations in shear stress at a given strain rate using a Reotest-2 rotation viscometer (Messgeräte Medingen GmbH, Germany). A conical indenter plastometer with the controlled depth of immersion of the cone and the apex angle of 40º was used for the study of soil strength of clay-sand model systems. The moisture content of soil was determined gravimetrically as the ratio of mass of water to the mass of solids present in the soil sample.

The soil organic carbon content was determined by dry combustion with the use of the carbon express analyzer AN-7529 (Russia). Soil texture was determined by the Fritsch Laser Diffraction Particle Sizer ANALYSETTE-22 (Germany).

2.2.2. Preparation of clay pastes.

10 ml of distilled water was added to 5.0 g of clay powder and thick paste was prepared. Then another 10 ml portion of water was added and mixing was repeated again. Then the paste was stored for a day at room temperature and the last 10 ml portion of water (without or with additives) was added. The paste was mixed to homogenous state, set aside for two hours and then the soil strength was measured as described below.

2.2.3. Modification of clay samples.

Modified clay was prepared by adding HA into a clay paste followed by thorough mixing of the system. The content of clay was 14.5%, the time of swelling varied from 1 to 60 days. The concentration of HA was ranged from 0.01 g/L to 0.1 g/L. The adsorption of HA continued for three days. To estimate the effect of clay modification on flocculation, Ch was added into preliminary modified as well as non-modified clay. The concentration of Ch was ranged from 0.1 to 1.0 g/L; pH of modified paste with added chitosan was 6.0.

2.2.4. Preparation of sand-clay systems.

The model sand-clay soil system was prepared by mixing of air dry clay with air dry sand in a required proportion (3, 10, 30% of clay). The samples were humidified by adding small portions of distilled water (0.3 cm^3) to the sand–clay mixture. The strength of homogenous samples was measured by a conical plastometer.

3. RESULTS

3.1. Rheological behavior of clay pastes.

To appreciate the combined action of two types of soil organic matter (recalcitrant and fresh OM), firstly we have studied the rheological behavior of clay pastes without addition of organic substances, followed by the addition of different OM types separately, and at last adding both types of OM.
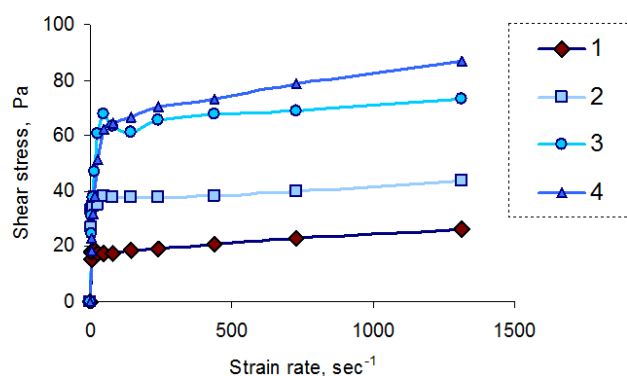
It was determined that all model pastes were thixotropic viscoplastic systems with coagulation contacts and yield limit.
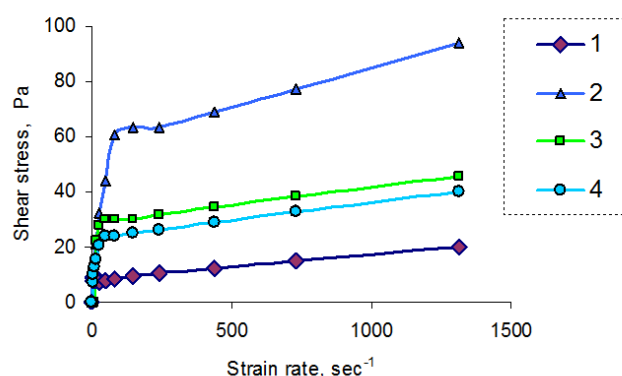

Figure 2. Influence of chitosan molecular weight on the rheological behavior of montmorillonite pastes, where: 1 – clay without Ch; 2 – clay with Ch 83 kDa; 3 – clay with Ch 240 kDa; 4 – clay with Ch 300 kDa.

The increase in the time of interaction did not lead to additional hardening of this system. We did not study the exposition of Ch paste longer than 24 h due to biodegradation of chitosan. The Ch addition to a montmorillonite paste even at a

Figure. 1. Influence of chitosan 83 kDa addition to montmorillonite pastes on their rheological behavior; Ch concentrations,: 1 – 0; 2 – 0.2; 3 – 0.5; 4 – 1 g/L. 1 – 0; 2 – 0,2; 3 – 0.3; 4 – 0,6 g/100g of soil.

concentration of 0.2 g/L increased the shear stress of the system, which was especially manifested at the concentrations of chitosan more than 0.5 g/L (Fig. 1).

The effect of chitosan of different molecular weight (83, 240 and 300 kDa) on the rheological characteristics is shown in (Fig. 2). At that, the addition of low molecular weight chitosan (83 kDa) led to the higher shear stress value compared with the addition of higher molecular weight chitosan.

No considerable effect of humic acid concentration on the rheological behavior of modified clay pastes was found (Fig. 3).
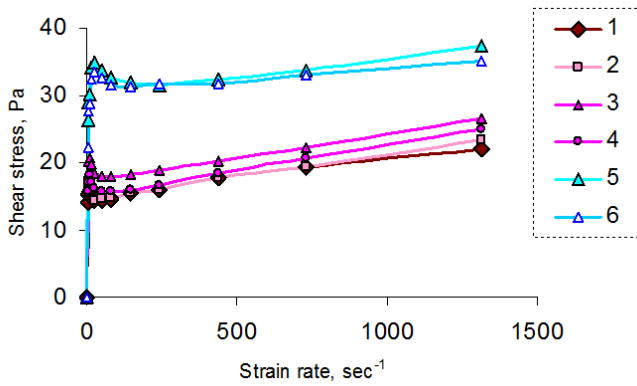

Figure 3. Influence of humic acid and chitosan addition to montmorillonite pastes on their rheological behavior, where: 1 – clay without HA; 2 – clay with HA 0.01 g/L; 3 – clay with HA 0.5 g/L; 4 – clay with HA 0.1 g/L; 5 – clay without HA, but with chitosan (one hour); 6 – clay without HA, but with chitosan (1 day).

The maximal strength of the system was observed when the concentration of HA was 0.05 g/L. The further increase in HA concentration did not lead to an increase in the shear stress value of the system. However, the chitosan addition to a modified montmorillonite paste at relatively low Ch concentration (0.1 g/L) led to a synergetic effect consisting of substantial hardening of the system, compared with such value when chitosan or HA were added separately (Fig. 4).
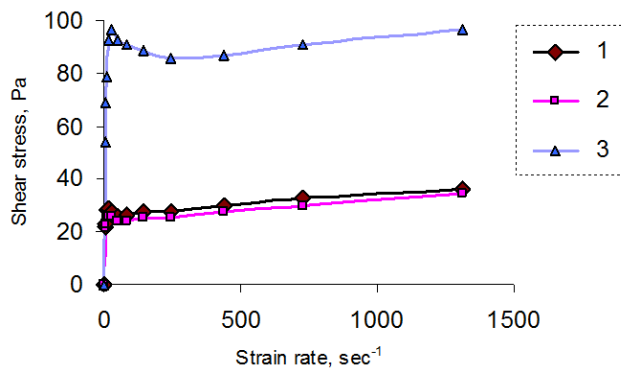

Figure 4. Influence of chitosan addition to montmorillonite pastes modified by humic acid on their rheological behavior, where 1– clay without HA and chitosan; 2 – clay modified by HA without chitosan; 3 – clay modified by HA with Ch addition.

3.2. Study of clay and sand by a conical indenter plastometer.

As far as kaolinite, in distinction from montmorillonite, is not a swelling mineral, we could not study kaolinite pastes with the same quantity of water as montmorillonite by rotation viscometer. For that reason, we used a conical indenter

plastometer. The results confirmed the data obtained with montmorillonite pastes, i.e. negligible hardening of the system modified by humic acids but a considerable hardening of the system upon the addition of a flocculant (chitosan) to kaolinite pastes modified by HS.

The same effect of synergetic action of HS and a flocculant, however, has not been confirmed in the case of sand pastes where the modification with HS leads to a decrease in the strength values in the presence of chitosan (Fig. 5). That fact points to the leading role of clay minerals in soil aggregates formation.
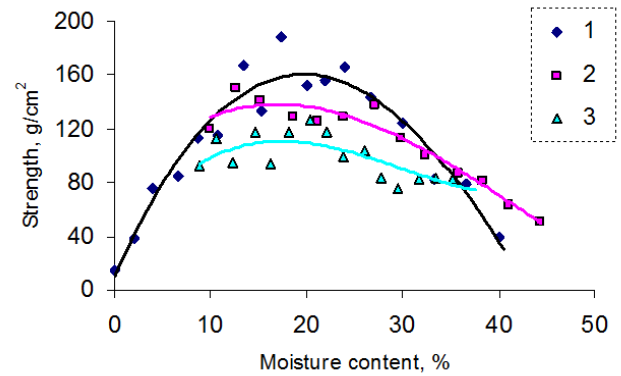

Figure 5. Influence of chitosan addition to sand and sand paste modified by HA on their rheological behavior, where: 1 – sand; 2 – sand modified by HA; 3 – sand modified by HA with added chitosan.

Podzolic soil with Corg 1,5% were studied to compare the strength of model clay-sand system (10% – clay, 90% – sand) as a function of water content with real soil. The results presented in the Fig. 6 show that both types of soils (real and model) demonstrate extreme character and the soil strength is higher (1000 g/cm^2) in the case of model soil when the water content was 20% compared with podzolic soil (600 g/cm^2) where the water content was nearly 40%.
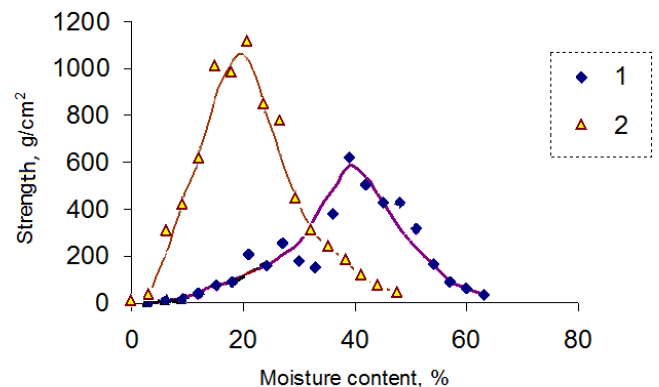

Figure 6. Influence of the water content and organic matter concentration on the strength of sod-podzolic soil and model sand-clay system, where: 1 – sod-podzolic soil with Corg 1,46%; 2 – model sand -clay (10%) system.

3.3. Discussion.

Aggregation of soil particles leads to the formation of micro-aggregates, that combine and form macro-aggregates [13]. This is determined by the relative contributions of biotic and abiotic processes or organic and inorganic binding agents [29-31]. The important role of the solid-phase reaction between clay minerals, polyvalent cations and SOM in the formation of

micro- and macro-aggregates was formulated by Edwards and Bremner [32], developed by Oades [1], Six et al. [2] and Shein et al. [33]. In our work we have largely focused on the soil aggregation via the flocculation of soil minerals with fresh SOM. One of the first scientists who in the early 1930-s emphasized the significant role of biological factors and fresh OM in the soil formation was Williams [34]. Recently published articles on soil formation are in agreement with this approach and revealed not only the key role of bacterial and fungal debris in the stable micro-aggregates formation [14, 35, 36] but also showed their synergistic effect with products of soil biota (plant roots, mycorrhizal roots and mycorrhizal hyphae) affecting aggregate stability [37]. Moreover, the soil biota releases CO_2 and forms SOM which may increase the dissolution of primary carbonates and enhance aggregation via the bridges between cations and organo-mineral particles [38]. The formation of water-stable organo-mineral aggregates on the basis of amphiphilic properties of SOM was explained by Milanovskiy. According to his view, hydrophilic components of SOM adsorb on mineral particles and their hydrophobic components are bound by hydrophobic interaction [18]. However, according to that approach, the HS is considered without their division into stable and mobile OM. A study of Elliot was published more than 30 years ago, in which he suggested that organic matter of macro-aggregates is "qualitatively more labile and less highly processed" than the organics stabilizing micro-aggregates [39].

The influence of soil HS fractions on soil formation was studied in [40]. The negative correlation was recorded between the content of organic carbon (C_{org}) in the fractions of water-resistant macro-aggregates (WRA) and the amount of extracted humic acids in coarse grained soil and fulvic acids in fine-grained soil. In other words, the formation of WRA is controlled by the more labile fraction of organic matter. These data are in agreement with our idea that different types of OM affect the formation of soil aggregates via flocculation by a different mechanism: labile hydrophilic component of SOM (fresh OM) may behave as a flocculant, and HS (recalcitrant OM) as a modifier of mineral particles. We consider that adsorption-flocculation may be one of the mechanisms that controls the soil structure formation.

To confirm that approach we studied the rheological behavior of model clay systems (montmorillonite and kaolinite pastes) without or with addition of organic matter of two types: HS and a flocculant (chitosan). The minerals were chosen due to their prevalent occurrence in soils. Montmorillonite predominates in chernozem, while kaolinite is the main clay of sod-podzolic soils [41]. Both these minerals belong to aluminosilicates. Kaolinite consists of stacked sheets of two layer units, it does not swell and is susceptible to the formation of gross particles, while montmorillonite is stacked of three layer units, which weakly interact with each other, so that the ions and molecules of water or low-weight organic compounds may penetrate into the interlayer spaces [42-44].

Chitosan (Ch) is a cationic polysaccharide with the structure consisted of D-glucosamine and N-acetyl-D-glucosamine. It is widely used as a natural flocculant. This biopolymer is produced in soil by microbial transformation of chitin via the alkaline N-deacetylation which consists of

removing the acetate group and replacing it with an amide group in alkaline solutions or via hydrolysis [45]. Ch has a high affinity to the surface of silica-based minerals due to the interaction between the part of protonated amino groups of polymer and dissociated hydroxyl groups of silica, which are formed in aqueous solution [46]. Thus, the mechanism of Ch interaction with the selected minerals is due to the electrostatic interaction as well as hydrogen bonding. On the other hand, Ch may interact with hydrophobic groups of organic substances (HA, FA, components of bacterial walls) via hydrophobic interactions [6], coordinate transition metal ions: Zn(II), Cu(II), Cd(II), and Fe(III) and some other soil metals [47] and thus enhance aggregate stability.

Our study showed that the addition of a small quantity of Ch (<0.5 g/L) to montmorillonite paste had a negligible effect on the shear stress of the system. The increase in the Ch concentration to 1 g/L resulted in a more visible hardening of the system, but a decrease of the shear stress was determined at a further increase of Ch concentration to 2 g/L (that curve is not shown in Fig 1). That may be explained by the change of the role of Ch from a flocculant at the concentration <2 g/l on a stabilizer of the system at higher concentrations [48].

The decrease of the Ch molecular weight led to an increase of shear stress at the same strain rate. Probably it is caused by the penetration of more mobile low weight Ch molecules into the interlayer space and formation of more stable aggregates (Fig. 2).

The modification of clay pastes with HA acid in concentration 0.01 g/L– 0.1 g/L during three days led to negligible strengthening of model soil composites (near 20 Pa) (Fig. 3), while the addition of Ch (even in a small concentration of 0.1 g/L) to the system with modified clay led to substantial strengthening of the system (up to 95 Pa) (Fig. 4).

These results permit to propose an adsorption-flocculation mechanism of the interaction of SOM with clay mineral particles. There are two types of adsorbed polyelectrolytes on clay surfaces: anionic polyelectrolyte (HS) and cationic polyelectrolyte (Ch). HS may adsorb due to penetration of some HS chains into interlayer space of montmorillonite [43], when Ch interacts due to the electrostatic mechanism. As it was shown by Tombacz et al. [49] that the formation of mixed polyelectrolyte adlayers decreases the ζ-potential of clay particles and allows them to come closer together and interact with each other due to hydrophobic interactions between side chains of HA. It improves the flocculation efficiency considerably compared with an individual polyelectrolyte action Moreover, anionic polyelectrolytes HS, having a negative charge in water medium, may interact with the positively charged Ch molecules, That also increases the stability of the formed soil aggregates [50]. Interaction of flocculants with adsorbed HS may not only bind the mineral particles but promote further binding of microaggregates into macroaggregates, because they provide nucleating sites for the microbial activities resulting in the deposition of extracellular polysaccharides [4].

We decided to confirm the synergetic effect (combined action of HA and a flocculant) found in clay systems using model sand systems. However, we obtained an opposite result. It

was found that the modification of sand by HS led to a decrease in the strength values in the presence of chitosan (Fig. 5). It may be explained by the formation of polyelectrolyte Ch-HA complexes not on the surface of sand particles that have a small surface area, but between them. Non-adsorbed HA may interact with Ch and form organic floccules which can play the role of lubrication between sand particles and decrease strength values.

Earlier it was found that the maximum specific activity of the respiration of the chitinolytic community was at a rather low redox potential with the soil moisture close to the total water capacity [51]. We decided to study the rheological behavior of model soil systems in the range of moisture at which the most intense microbial transformation of chitin was observed. It was found that the maximum strength value of model soil was 1000 g/cm^2 while as sod-podzolic soil had the maximum strength value of 600 g/cm^2 (Fig. 6). The addition of clay to sand systems led to hardening of the system up to 1900 g/cm^2 when the content of clay was 30%. At the same time, sand systems were characterized by lower values of strength compared with clay-sand systems. Thus, the stability of aggregates showed a positive correlation with the clay content, which has been shown in the studies of other researchers [52, 53].

The comparison of model systems with soils showed that rheological parameters of fallow chernozem are similar to the model systems, while the rheological characteristics of virgin chernozem exceed the results of model systems by an order of magnitude [54, 55, 56]. Intense tillage can lead to the mineralization of soil OM. The shear stress of non-treated chernozem (C_{org} 6.8%) was significantly higher compared with plowed soil (C_{org} 3.0%) The nearly two fold decrease in the soil OM content leads to the degradation of soil structure and may enhance the role of OM, especially its labile fractions, in the contact interactions in soils [57]. These data agree with the published results that SOM storage and soil aggregation were significantly reduced in the surface soils of croplands compared to those under native vegetation [58, 59]. Hence, agricultural practices that decrease soil disruption can enhance aggregation and structural development [7].

The Vernadsky's idea [60] to consider soil as bio-inert matter in which interactions between living and inert matter is a key factor regulating biogeochemical cycles is compliant with our approach in which the interaction of different types of SOM take part in soil structure formation. In our case, the newly formed polyelectrolytes behave as bio-components, and HS plays the role of inert substances.

The proposed adsorption-flocculation mechanism of soil formation has a common character. It may be applied to other fields of investigation, for instance, to the study of aggregate formation in the river-sea mixing zone (delta, estuary) in which it can initiate soil formation [61]. Water purification, preparation of sand-clay pastes of various applications and some other process of mineral-OM interaction may be considered on the basis of the described mechanism.

4. CONCLUSIONS

The role of SOM in the formation of soil aggregates is still debatable. We propose a new approach based on the flocculation of modified mineral particles by polyelectrolytes (fresh SOM) released from soil biota in situ. The modification of mineral particles is due to adsorption of another kind of polyelectrolytes - humic acids (recalcitrant OM).

Rheological studies were applied to model systems to show the synergistic effect of the combined action of these two types of OM. Thus, the modification of clay with humic acids of varied concentration did not affect the rheological behavior of the pastes. The action of a single flocculant (chitosan) only slightly affected the strength of the systems. However, the addition of flocculant to modified clay enhanced the strength of such systems considerably.

That is explained by the formation of a mixed layer of both types of polyelectrolytes on the surface of mineral particles facilitating the aggregation. Aggregation due to such adsorption-flocculation mechanism was not revealed in sand systems, which confirmed the leading role of clay particles in soil aggregation.

The comparison of the model and sod-podzolic soil systems showed the effect of humidity and content of OM on the systems strength. The proposed adsorption-flocculation mechanism can be applied not only to explain soil aggregation, but also to the study of other systems in which there are interactions between minerals and OM.

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6. ACKNOWLEDGEMENTS