Application masking liquid technique for SAXS study of natural bio-nanocomposites

Yu V Larichev
Boreskov Institute of Catalysis, Siberian Branch of Russian Academy of Sciences, Prospekt Akademika Lavrentieva 5, Novosibirsk, 630090, Russia
Novosibirsk State University, Pirogova Street 2, Novosibirsk, 630090, Russia
E-mail: ylarichev@gmail.com

Abstract. Natural bio-nanocomposites (rice husk, sapropel, peat and brown coal) have been studied by TEM, XRD and SAXS. Particle size distributions for the mineral matter in these systems were determined by SAXS with using full contrast technique. In the rice husk silica nanoparticles are forming compact particles with bimodal distribution and mean size is equal 4.7 nm. In the case of peat and brown coal mineral matter in organic matrix are forming more large particles with compact shapes and mean sizes 14.1 nm and 14.3 nm respectively. In the sapropel inorganic matter are forming elongated structures with lengths are equal 20-70 nm and widths are equal 2-10 nm.

1. Introduction
Natural bio-resources (such as rice husk, peat, sapropel and other) have a big perspective as a source for preparation different types of low-cost carbon and carbon-mineral adsorbents and functional materials [1-5]. The most significant hindrance for wide application of these materials in industry is unsatisfactory reproducibility of main properties due to preparation these materials. The base reasons of this problem are complex composition of natural materials, amorphous structure and respectively insufficient information about particle sizes and shapes of mineral parts of these materials. These data are needed for understanding and controlling process of formation porous structure due to calcination and activation such bio-resources. Removing of mineral matter (totally or partially) could noticeably increase specific surface and pores volume of these materials [3, 5]. Depends on the mineral matter content and their particle shape and sizes in bio-composites it is possibly to consider carbon porous structure with inorganic inclusion or vice versa porous mineral matrix covered by carbon-contain films. Nevertheless information about inorganic particle sizes and their shapes in initial bio-materials practically is absent and preparation condition of functional materials are needs to select in each case at that its depend on the batch of raw materials these conditions could be different [4, 5]. Information about sizes and shapes of mineral particles in bio-composites should help in solution such problem about reproducibility. One of the methods is suitable for obtaining data about particle sizes in bio-materials is small angle X-ray scattering (SAXS) with using special masking liquid for masking parasitic scattering signal from organic/carbon matrix of samples [6, 7]. In this work we report about results of analysis mineral components (particle size distribution and particle shapes) in rice husk, peat, sapropel and brown coal.

1 To whom any correspondence should be addressed. E-mail: ylarichev@gmail.com
2. Experimental section

Sample preparation

For study have been used four different bio-composites. Rice husk was grown in the Krasnodar region of Russia. According to [8], this rice husk contained 19–25 % lignin, 34 – 42 % cellulose, and 17–22 % hemicellulose. The ash content of the husk was 19.5%, and the SiO₂ content in the ash was above 96%. Sapropel has been mined from Gorkoe Lake (Omsk region of Russia). The ash content of the sapropel was 64.3%. Ash has a complex composition: 63.4% Si, 7.1% Al, 8.8% Ca, 5.6% Mg, 3.4% Fe and other minor elements.

Peat has been mined from Taganskoe peat field (Tomsk region of Russia). The ash content of the peat was 29.0%. Ash consists from next elements: 31.3% Si, 15.2% Al, 18.2% Fe, 25.9% Ca, 3.5% Mg and other minor elements.

Brown coal (lignite) has been mined from Kansk-Achinsk coal basin (Krasnoyarsk region of Russia). The ash content of the brown coal was 11.1%. Ash consists from next elements: 57.1% Si, 22.1% Al, 7.5% Fe, 4.2% Ca, 2.1% Mg and other minor elements.

Sample characterization

X-ray diffraction

XRD investigations were carried out at an ARLX’TRA diffractometer (Thermo, Switzerland) with a Si(Li) solid-state detector and CuKα radiation (wavelength λ is equal to 1.5418 Å). The measurements were carried out in the 2θ range of 10−90° with a step of 0.05°. Storage time in point is a 5 seconds.

True density measurement

True densities of bio-composites were measured pycnometrically. Volume of the liquid displaced by a porous sample was measured. The measurement was made at least three times for each sample, and the obtained values were averaged. The liquid used in these experiments was heptane. The measurement error was less than 4 %.

Transmission electron microscopy

Electron microscopy examination of the samples was performed using a JEM-2010 transmission electron microscope (JEOL, Japan) with accelerating voltage 200 kV and resolution 0.14 nm. The samples for the TEM study were prepared by ultrasonic dispersing in ethanol and consequent deposition of the suspension upon a "holey" carbon film supported on a copper grid. The linear sizes of not less 300 particles from mineral part of bio-composites were measured for each sample to obtain particle size distributions (Dₙ(R)). For correct comparison with SAXS data TEM histograms were recalculated as Dᵥ(R) = Dₙ(R)*R³.

Small-angle X-ray scattering

SAXS data were acquired using an S3 MICRO (HECUS) diffractometer with point collimation and Cu Kα radiation (wavelength is equal to 1.541 Å). The electrical power of generator for X-ray tube is 50W (1.0 mA at 50 kV). The diffraction patterns were measured in the vector q ranges from 0.01 to 0.6 Å⁻¹, where q = 4sinθ/λ. The samples were placed in a 1.5 mm quartz capillary. For selective extraction of the SAXS pattern of dispersed mineral phase from the total SAXS pattern from whole sample we developed an original full-contrast technique with using fluorocarbon compounds as masking liquids [6, 7]. A sample in the capillary was filled with a special liquid having a density, which roughly corresponds to the density of organic or a carbon matrix in the samples. Whereas electron density and pycnometric density of materials are close to each other the last value has been used for convenience. For amorphous or disordered samples are very difficult to measure electron density value instead pycnometric density. For this reason in practical applications it is better to use pycnometric density and make additional density corrections if it need in experiment. Processing of experimental data and calculation of the particle size distributions were performed by GNOM software from the ATSAS package [9].

3. Results and discussion
Figure 1 are shows XRD patterns from biomaterials. Reflections observed for RH can be attributed to the cellulose structure [10, 11]. No other reflections corresponding to silica are observed. At the same time there are lot reflections for a sapropel. More intensive reflections are corresponding to α-quartz (SiO$_2$) structure. Also other reflections are corresponding to albite (NaAlSi$_3$O$_8$), muscovite (KAi$_2$(AlSi$_3$O$_10$)(F,OH)$_2$), leucite (K[AlSi$_3$O$_10$]) and hematite (Fe$_2$O$_3$). In generally it is typical phase composition for the sapropels [3]. XRD patterns for peat and brown coal have also typical views for such minerals [12, 13]. In the case of peat and brown coal also are found reflections from α-quartz structure. Reflections from α-quartz structure have narrow lines and particle sizes estimated from Scherrer equation are equal approximately 100 nm. The other possible compounds are not give noticeable reflexes. Moreover XRD patterns also contain broad peaks (halo) with the maximums at 21° and 41° approximately. These broad peaks can be assigned to disordering carbon with graphite-like structure and amorphous silica too [8, 14]. According XRD data a lot part of matter in bio-composites are presented in amorphous and/or disordered states.

![Figure 1. X-ray diffraction patterns for rice husk, sapropel, peat and brown coal.](image)

Figure 2 displays TEM images from such biomaterials. According TEM data in the case of rice husk their mineral matter are forming particle with compact shape and typical sizes 2-10 nm. In the sapropel mineral phase are forming elongated structures with typical lengths are equal 20-70 nm and widths are equal 2-10 nm. The average ratio length/width for these aggregates is about 7.5. For the peat and brown coal has been found that mineral matter in both cases are forming compact particles with typical sizes about 10-30 nm. For the all TEM data (except sapropel) have been calculated volumes particle size distributions for comparing with corresponding SAXS data. The SAXS curves of bio-composites are contained complex mixture information about pores, mineral particles and organic/carbon matrix. The current q range is restricting SAXS spatial resolution up to 65 nm. Therefore huge particles of quartz with sizes about 100 nm will be unavailable for SAXS analysis. To the other hand the all amorphous or disordered inorganic matter with less sizes in bio-composites must give scattering signals in the SAXS patterns.
Figure 2. Typical TEM images for rice husk (A), sapropel (B), peat (C) and brown coal (D).

Figure 3. (Left) SAXS curves for initial rice husk (red) and rice husk after impregnation with masking liquid (blue). Black line is scattering curve from model of polydisperse spheres. (Right) Volume particle size distributions for silica particles in rice husk calculated from corresponding SAXS curve (blue line) and TEM distribution (bar chart).

Figure 3 presents SAXS data for the rice husk. The slope of the initial scattering curve $I(q)$ for the rice husk is proportional to $\sim q^{-2.8}$. Hence, the particle form-factor cannot be determined strictly in this case, and it is difficult to find the particle size distribution. Such slope of the scattering curve can be typical,
for example, of fractal aggregates, which may consist of cellulose fibers. The application of the masking liquid technique makes it possible to suppress virtually completely the scattering signal of the organic phase. When rice husk was impregnated with the masking liquid having the density of 1.4 g/cm$^3$, the SAXS signal corresponding mainly to the silica phase was obtained for the sample. As it is shown in Figure 3, the scattering curve for the contrast matched sample has much lower intensity and a radically different slope ($I(q) \sim q^{-4.0}$). Such slope of the scattering curve is typical for particles with compact (sphere-like) shape [15]. Using spherical form-factor for modeling curve has been get volume particle size distribution for silica particles (Figure 3). This distribution has a small particle fraction with sizes from 1 to 7 nm and large particle fraction with sizes from 8 to 16 nm. Mean particle size in this distribution is equal 4.7 nm. This result has good qualitative agreement with TEM distribution (Figure 3). For quantitative comparing TEM and SAXS distributions there are need to have much more statistical data for TEM distribution. Typical quantity 300-350 particles for TEM histogram in our case is allow making only qualitative comparing with SAXS data. Moreover that wider particle size distribution than more number of particles for good statistics are needed. So SAXS with masking liquid technique is allow to high quality information about particle sizes of inorganic matter in such biomaterials.

![Image](image_url)

**Figure 4.** (Left) SAXS curves for initial sapropel (red) and sapropel after impregnation with masking liquid (blue). (Right) Such SAXS curves in coordinates $I(q)q^4$; $q$.

In the case of sapropel scattering curve $I(q)$ is proportional to $\sim q^{-3.1}$ (Figure 4). The application of the masking liquids with density of 1.35 g/cm$^3$ practically does not influence on this slope value. The slope value of SAXS curve after using masking liquids is increased up to -2.8. Such slope value (-2.8) for SAXS curve with using contrast is indicate that mineral matrix in sapropel are forming rod-like or fractal-like particles [15]. This assumption is agreed with TEM image of sapropel (Figure 2). The value of radii gyration ($R_G$) for mineral particles in sapropel is equal 16.2±0.5 nm. This value is approximately corresponding to $R_G$ values of elongated particles on the TEM image (Figure 2). For example model solid ellipsoid with such semi-axes as (a = 5 nm, b = 5 nm, c = 37 nm) has a value of radii gyration is equal 16.8 nm [16]. Indefinite form-factor of particles in this case does not allow getting particle size distribution from SAXS data. Likewise such indefinite shape of particles in sapropel is impeding to calculate of TEM distribution too. Application of masking liquid with density of 1.21 g/cm$^3$ for the peat is changed slope value of the SAXS curve from -2.9 to -4.0 (Figure 5).
Figure 5. (Left) SAXS curves for initial peat (red) and peat after impregnation with masking liquid (blue). Black line is scattering curve from model of polydisperse spheres. (Right) Volume particle size distributions for mineral part in the peat calculated from corresponding SAXS curve (blue line) and TEM distribution (bar chart).

Figure 6. (Left) SAXS curves for initial brown coal (red) and brown coal after impregnation with masking liquid (blue). (Right) Black line is scattering curve from model of polydisperse spheres.
This liquid mixture is decreasing parasitic scattering from organic matrix of peat and we observed scattering intensity from mineral particles in the peat only. Such slope (-4.0) for SAXS curve is typical for particles with compact (sphere-like) shape. Volume particle size distributions for mineral part in the brown coal calculated from corresponding SAXS curve (blue line) and TEM distribution (bar chart). The volume particle size distribution for mineral matter of peat has been get with using spherical form-factor (Figure 5). This distribution has monomodal character with mean size is equal 14.1 nm. This particle size distribution from SAXS data are agreed with similar TEM particle size distribution. Some discrepancy between SAXS and TEM data are determined insufficient TEM statistics.

In the case of brown coal there is similar picture comparing to peat. After application masking liquid with density of 1.30 g/cm³, the slope value of SAXS curve is changed from -3.1 to -4.0 (Figure 6). After using spherical form-factor for fitting SAXS curve from mineral particles in brown coal matrix has been got particle sizes distribution with mean size is equal 14.3 nm and bimodal character. TEM particle size distribution has practically the same particle sizes and also there is quality agreement between SAXS and TEM data (Figure 6). The near sizes for mineral matter in the peat and brown coal could be as simple coincidence or indicated on the similar formation conditions and transformation of such bio-composites on our planet. In any case it is need more data for precise conclusion.

Finally in Table 1 are shows some properties of bio-composites and particle sizes of mineral components. The rice husk has mineral particles with minimal sizes. Peat and brown coal are containing more large particles with similar mean sizes. Although in brown coal are presenting as less small and more large particles while in peat mineral particles has more homogeneous distribution.

Table 1. Physical properties of bio-composites and particle sizes of mineral parts therein.

| Samples    | Density, g/cm³ | Ash content, wt% | R_G, nm | D_ave, nm* |
|------------|----------------|-------------------|---------|-------------|
| Rice husk  | 1.54           | 19.5              | 5.8±0.1 | 4.7         |
| Sapropel   | 2.29           | 64.3              | 16.2±0.5| 14.1        |
| Peat       | 1.62           | 29.0              | 8.4±0.2 |             |
| Brown coal | 1.47           | 11.1              | 11.0±0.3| 14.3        |

* *<D_ave> = \sum n_i * d_i / \sum n_i where d_i the diameter of i-particles on the SAXS histogram, N the number points on the SAXS histogram.

In sapropel sample mineral particles have a maximal sizes and elongated shapes. Due to high percentage of mineral phase in sapropel these particles are aggregated and formed elongated structures. Probably that for sapropels with less quantity of mineral phase their particle sizes will be close to such particles in the peat and brown coal.

4. Conclusions

The SAXS method with liquid masking technique has been used for study mineral matter in biomaterials. In the case of rice husk silica nanoparticles are formed bimodal distribution with mean sizes particles are equal 4.7 nm. In the case of sapropel mineral matter of this material are formed elongated and/or fractal-like particles with mean R_G value is equal 16.2 nm. In the case of peat mineral matter of this material are formed nanoparticles with monomodal distribution and mean particle sizes are equal 14.1 nm. In the case of brown coal mineral matter of this material are formed nanoparticles with bimodal distribution and mean particle sizes are equal 14.3 nm. There is a good agreement between TEM and SAXS results.

Acknowledgement
The author is grateful to P.M. Yeletsky, O.I. Krivonos and E.V. Maltseva for samples provision and D.A. Zyuzin, A.V. Ishchenko for assistance in the investigations of the samples by XRD and TEM. This work was supported by the Russian Science Foundation (grant no. 14-23-00037).

References
[1] Pietrzak R, Wachowska H, Nowicki P, Babel K 2007 Fuel Processing Technology 88 409
[2] Yeletsky P M et al 2009 Microporous and Mesoporous Materials 121 34
[3] Kovalenko G A, Adeeva L N 2010 Chemistry for Sustainable Development 18 181
[4] Virla L D et al 2016 Microporous and Mesoporous Materials 234 239
[5] De Gisi S et al 2016 Sustainable Materials and Technologies 9 10
[6] Larichev Yu V, Tuzikov F V 2013 J. Appl. Cryst. 46(3) 752.
[7] Larichev Yu V, Tuzikov F V 2013 Kinetics and Catalysis 54(5) 632
[8] Larichev Yu V et al 2015 Journal of Physics and Chemistry of Solids 87 58
[9] Konarev P V, Petoukhov M V, Volkov V V, Svergun D I 2006 J. Appl. Cryst. 39 277
[10] Park S, Baker J, Himmel M, Parilla P, Johnson D 2010 Biotechn. Biofuels 3:10.
[11] Lu P, Hsieh Y L 2012 Carbohydrate Polymers 87 564
[12] Asapo E S et al 2012 J. Miner. Mater. Characteriz. Engineer. 11 709
[13] Takagi H, Maruyama K, Yoshizawa N, Yamada Y, Sato Y 2004 Fuel 83 2427
[14] Larichev Yu V et al 2008 Catalysis Letters 120(3-4) 204
[15] Feigin L A, Svergun D I 1987 Structure Analysis by Small-Angle X-Ray and Neutron Scattering (New York: Plenum Press) 335
[16] online SAXS calculator: http://scattering.tripod.com/xitami/java/iq.html