CRYSTALLOGRAPHIC METHODS IN HUMANITARIAN SCIENCES

Electron Microscopy Studies of Manuscripts from Russian National Library

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Abstract—Inks of historical documents from the collection of the Russian National Library and model samples on paper and polymer were studied by transmission electron microscopy and energy-dispersive X-ray microanalysis. The results of substantially nondestructive analysis of inks make transmission electron microscopy an efficient tool for studying large arrays of manuscripts.

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INTRODUCTION

The information potential of historical manuscripts is fairly versatile: they may be not only physical records of texts and images (historical, linguistic, literature etc. sources), but also are of great value for the history of material culture, in particular, history of technologies. Each manuscript is a result of the activity of people involved in its creation, as well as a product of a complex of technologies (sheet formation, ink production, etc.). In order to achieve full implementation of this part of historical document's information potential it is necessary to perform nondestructive analysis using modern scientific methods.

Historical and technological study of writing dyes (primarily, inks) begins to play a special role in modern science about manuscripts. Specifically, the objects of study are the composition of historical colorants, specific features of their formulation, and stability and variability of their properties in time. Different methods of optical spectroscopy (multispectral visualization, Raman spectroscopy, etc.), X-ray fluorescence analysis, and gas chromatography [1–5] are used for these purposes.

In our opinion, methods of electron microscopy (EM) and microanalysis can hold a particular position in the historical, technological, and expert study of various colorants (including manuscript inks) due to the possibilities of local structural analysis in combination with the study of elemental composition. Using

low-vacuum conditions and low accelerating voltage in scanning electron microscopy (SEM), one can investigate nonconducting samples without any additional preparation procedures and with radiative damage reduced to minimum.

The low penetration depth of electrons in matter makes an advantage of EM methods in comparison with X-ray techniques, especially when studying surface layers, for example, ink or colorant layer on paper or parchment. Undoubtedly, there are some limitations, related mainly to the fact that a large part of manuscript cannot be placed in an SEM chamber because of its limited size and/or likely damage of manuscript in vacuum. Therefore, the EM studies of historical documents, described in scientific literature, were generally performed on fragments or specially selected samples. In particular, the SEM method combined with energy-dispersive X-ray microanalysis (EDX) was applied in [6] to determine the elemental composition of parchment surface and the ink layer on it. A complex of SEM/EDX methods was used in [7] to study microscopic samples of black and red colorants, classified as carbon-containing ink and cinnabar, respectively. There were difficulties in determining the black ink composition because of the low intensity of Fe fluorescence signal. Another representative study is the determination of the composition of color ink of the tenth-century Quran manuscript [8]. The EDX of the parchment revealed, along with carbon and oxygen, the presence of Si, Na, K,

Table 1. Recipes of replicated inks

Material (% of dry material weight)		<i>i1, i3</i> Recipe 1 Collection 67/24 [9]	<i>i2</i> Recipe 2 Collection 39/133 [9]
Nutmalls	g	34	13.6
	%	51.52	58.62
FeSO ₄	g	23.8	4.8
	%	36.06	20.69
Gum	g	8.2	4.8
	%	12.42	20.69
Water	mL	400	350

Ca, Mg, Cl, and S in its composition; the researchers believed these impurities to be due to the treatment of parchment aimed at imparting a certain hue to it. The manuscript text was written using inks of three colors: black, red, and green. The analysis revealed that some samples of red ink contained mercury and were prepared based on vermilion or cinnabar (HgS). At the same time, some of red colorant samples contained Fe and O, which indicates that some part of red ink could be prepared based on ochre: mixture of iron oxide with clay, possibly, with organic pigment added. The green ink contained Cu salts, and the black ink consisted mainly of carbon (soot component), with a low content of Fe impurity.

Note that the SEM/EDX technique is applied relatively rarely in the analysis of ink in view of the aforementioned limitations: electrical insulating properties of paper and parchment and the necessity of selecting material from historical documents (samples with a thin ink layer); hence, the potential of this method for studying manuscripts is limited. A possible way out is the use of transmission microscopy, in which sample sizes can be very small: from several micrometers to 0.1 mm; this circumstance may be a significant advantage for the analysis of ink and pigment samples. However, we could not find any data in the literature on studying historical inks of manuscripts by methods of transmission electron microscopy (TEM) and scanning transmission electron microscopy (STEM).

Here, we proposed to use the TEM and STEM methods in combination with EDX for studying several historical documents. Model ink samples fabricated according to ancient Russian recipes under laboratory conditions were analyzed for comparison. The results of the analysis by the STEM/TEM/EDX methods are considered below.

EXPERIMENTAL

The objects of study were model samples of replicated (i.e., fabricated according to ancient Russian recipes under laboratory conditions) ink [9] and text colorants of historical documents.

The model objects of study were experimental samples of writing with replicated historical ink. Replicated ink samples were deposited on different surfaces for testing in order to optimize the sampling technique and fit experimental conditions: samples *i1* (recipe 1) and *i2* (recipe 2) were deposited on a paper base, while sample *i3* (recipe 1) was dried on a polymer substrate (to exclude the influence of paper on the TEM/STEM data and obtain information about the composition). The replicated inks were made according to the recipes from the “Collection of Written Sources...” (Table 1, recipe code: “source number in Collection”/“paper number in source” [9]).

Historical ink samples were taken from two manuscripts. The first was the Report to the Governing Senate from the Tula Chamber of the Civil Court on April 14, 1808 (paper document from the experimental materials of the Laboratory of Codicological Studies and Scientific and Technical Examination of Documents of the Department of Manuscripts, which is not stored in the funds of the Department of Manuscripts of the Russian National Library) (samples *i5* and *i6*, see Fig. 1). The second manuscript was a parchment Synodicon dated to the end of the 1570s—beginning of the 1580s (from the funds of the Department of Manuscripts of the Russian National Library, the Main collection of manuscripts F.no.IV.1), sheet 55, row 2, letter 3 and sheet 6, row 4, letter 41 (samples *i7* and *i8*, respectively).

The key tasks of our methodological research was to determine the minimally necessary volume of ink layer samples for TEM/STEM analysis and, proceeding from the result, to search for and optimize the sampling method minimally affecting historical manuscripts.

To select model samples of colored surface layers on paper (samples *i1* and *i2*) and polymer substrate (sample *i3*), ink was scraped off by a scalpel from an area of no more than 2×2 mm² in size, with subsequent sample transfer on an electron microscopy grid.

Further EM study showed that this sample volume is excessive; therefore, microscopic samples of historical documents were obtained using an injection needle with $d = 0.5$ mm (monitoring tools were a light microscope or a lamp loupe $\times 10$) from regions with an area of 1 mm² or less, characterized by the highest prime density. The powder obtained as a result of loosening the prime (letter or symbol) surface or scraping was captured by a copper electron microscopy grid with a perforated carbon film Lacey or a perforated film based on SiO₂. Neither organic nor inorganic solvents were used: powder particles stuck to the carbon film due to the electrostatic or van-der-Waals attraction forces.

TEM/STEM analysis was performed on a transmission electron microscope Osiris (ThermoFisher Scientific) equipped with an energy-dispersive X-ray spectrometer Super-X (Bruker) and a high-angle

Fig. 1. Report to the Governing Senate from the Tula Chamber of Civil Court on April 14, 1808 (samples i5, i6).

annular dark-field detector (Fischione); the accelerating voltage was 200 keV. No less than five particles were investigated in each sample; the results were averaged.

RESULTS

A TEM study of the model samples showed them to be agglomerates consisting of a matrix (appearing amorphous in TEM images), relatively large aggregates (several micrometers in size), and small (up to 200 nm) particles.

Figure 2 presents TEM images of a fragment of sample i1. Some large aggregates (1–3 μm in size) are indicated by white arrows, and a group of small particles is indicated by a black arrow. The character of contrast from large aggregates is similar to that of massive samples; hence, one can suggest that they are amorphous aggregates of different thicknesses.

The EDX spectrum of model samples i1–i3 shows the presence of C, O, Na, Si, S, K, and Fe in their composition (Table 2). Note that quantitative EDX was performed on particles located in free gaps rather than on the carbon grid Lacey; however, the latter may also contribute to the carbon fluorescence intensity. The fairly strong Cu signal, which is due to the fluo-

rescence from copper EM grid, was excluded from consideration when analyzing the model-sample composition. Sample i2 contains Ca. The composition of sample i3, which did not contact paper, was

Fig. 2. Bright-field TEM image of a fragment of model sample i1.

Table 2. Results of studying the ink sample composition (wt %)

Element	Samples		
	<i>i1</i>	<i>i2</i>	<i>i3</i>
Fe	12	12	9
S	4.5	3	4.5
O	14	13	10
C	68	71	75
Na	<1	<1	<1
K	<1	<1	<1
Ca		<1	
P	2	<1	<1
Si	<1	<1	<1
Al	<1	<1	<1

compared with that of sample *i1* (both were prepared according to the same recipe). One of significant distinctions for *i3* is the constancy of concentration ratio $\text{Fe} : \text{S} \approx 1 : 1$ practically throughout the entire sample volume. At the same time, significant variations were observed for the oxygen content, which could differ as much as twice over the sample surface. On the whole, one can conclude that paper does not change significantly the ink composition (identified by the TEM/EDX method). Since carbon enters the composition of not only the samples under study but also the grids used as sample carriers, this element was excluded from consideration when determining the sample composition.

An analysis of the EDX elemental distribution maps for samples *i1* and *i2* (the map for *i1* is presented in Fig. 3) showed that all elements are distributed uni-

formly throughout the volume; i.e., the samples are homogeneous.

A high resolution TEM (HRTEM) study of sample *i1* confirmed the suggestion that the sample is mostly amorphous. Some areas in HRTEM images (Fig. 4a) exhibit ordering; however, these areas are too small in size (less than 3 nm) to draw any unambiguous conclusions. A Fourier analysis confirmed the presence of periodicity about 0.27 nm in size.

The structural analysis of sample *i2* by HRTEM and electron diffraction (ED) showed unambiguously that this sample is amorphous: the HRTEM image does not contain any regions with periodicity, and the ED pattern from a selected sample region demonstrates a diffuse halo (Fig. 4b). The same results were obtained on other agglomerates in sample *i2*.

According to the HRTEM data and results of Fourier analysis, the structure of sample *i3*, in contrast to that of sample *i1* (prepared using the same recipe), is amorphous and does not exhibit any orderings in the form of structured microparticles. Apparently, contact with paper in certain recipes gives rise to microparticles in the ink layer.

Historical documents demonstrate a peculiar inhomogeneity of ink color, which is related, on the one hand, to the difference in the prime density and, on the other hand, to variations in the ink composition. In addition, the long-term interaction of ink with carrier (paper or parchment), as well as the destruction during the long-term existence, change unambiguously the ink layer by causing its structural transformation.

Samples *i5* and *i6* were taken from the Report to the Tula Chamber of the Civil Court ...; the letters analyzed had different hues. It is noteworthy that the composition of particles of these samples was highly inhomogeneous. Since the diversity of spectra of dif-

Fig. 3. Dark-field STEM image of a particle of ink sample *i1* and the results of EDX elemental mapping of the sample.

Fig. 4. HRTEM images of the samples on paper: (a) a selected region of sample *i1* (the inset shows a two-dimensional Fourier spectrum of this region), with an arrow indicating the ordering region, and (b) a region in sample *i2* (the inset shows the ED pattern for this region).

Fig. 5. Result of studying a particle of ink *i5*. A dark-field STEM image and EDX elemental mapping data.

ferent particles did not allow us to draw a final conclusion about the difference in samples *i5* and *i6* based on EDX data, we present below only generalized main characteristics. The results (elemental distribution maps are shown in Fig. 5) demonstrate a significant variation in composition. Carbon is observed practically throughout the entire particle volume. Particles contain Fe, C, Mg, and K; conglomerates containing Ca, S, and O (corresponding presumably to CaO , CaCO_3 , or CaSO_4) are also present. One can observe coinci-

dence of Ca and P maps, which may be related to the presence of hydroxyapatite ($\text{CaH}(\text{PO}_4)_3$); Al and Si maps coincide in some regions, which may be related to the presence of aluminosilicates. Regions with uniform distribution of Na and Cl manifest themselves, which are most likely related to the presence of salt.

A quantitative analysis of spectra of individual particles of samples *i5* and *i6* showed that the samples, along with carbon and oxygen, have a high content of Fe, which may reach 15 wt %. The S content may vary

Fig. 6. HRTEM image of a particle of sample with carbon fibers (indicated by arrows).

from 0 to 15 wt %. There are also particles containing, along with Fe and S, calcium (up to 12 wt %) and potassium (up to 8.5 wt %).

Particles exhibit orderings with a characteristic size of ~ 2 nm. An analysis of two-dimensional Fourier spectra reveals peaks corresponding to interplanar spacings $d = 0.36$, 0.28 , and 0.24 nm, which can be attributed to sulfate $\text{Fe}(\text{SO}_4)_3$ with a structure described by the rhombohedral system with the sp. gr. $R3$. This compound, identified as mikasaite [10], is characterized by a wide color spectrum ranging from white to brown. The phase could not be determined more exactly on crystallites with sizes lower than 2 nm.

An analysis of sample *i6* revealed existence of SiO_2 -containing particles. In addition, HRTEM data indicated possible presence of hematite (Fe_2O_3) crystallites with a rhombohedral unit cell, sp. gr. $R\bar{3}c$. The same sample contained particles with characteristic graphite fibers and an interplanar spacing of 0.334 nm (Fig. 6).

Based on the results of complex STEM/TEM/EDX study of historical ink samples, one can conclude that their composition is closer to that given by recipe 1 (model samples *i1* and *i3*).

The EDX of microprobes of samples *i7* and *i8* with elemental distribution mapping (Fig. 7) showed, as in the case of *i5* and *i6*, high compositional inhomogeneity. Therefore, we describe below the generalized characteristics of these two samples. Along with the high content of C (up to 32 wt %) and O (up to 30 wt %), the EDX spectra demonstrate very high contents of Ca (up to 20 wt %) and Si (up to 10 wt %). The Fe and S contents are lower than in other samples: up to 6–7 wt %. The samples contain K, Mg, Na, Cl, P, and Al in small amounts, along with a high Ca content. The presence of these elements in top parchment layers was mentioned in [11]. The dark-field STEM image in Fig. 7 reveals a rounded shape of particles, which was not observed previously for such samples.

The TEM, STEM, and ED data showed that the particles containing C and Fe are amorphous, while the Ca- and Si-containing particles are crystalline. A characteristic image of a group of particles is presented in Fig. 8.

Note that the TEM/STEM/EDX study of ink samples on parchment turned out to be less informative in comparison with the analysis of paper-based historical document. The reason is the significant influence of the elemental composition of parchment surface on the sample composition. Nevertheless, a comparison of the C, Fe, and S contents suggests that the ink composition for samples *i7* and *i8* is closer to that of model sample *i2* (recipe 2).

CONCLUSIONS

The results of our study showed that developed procedure of selecting microprobes yields a sufficient amount of material for electron microscopy of ink from historical documents. This sampling technique, characterized by very low (practically insignificant)

Fig. 7. Result of studying a particle of ink *i7*. Dark-field STEM image and EDX elemental mapping data.

Fig. 8. Bright-field STEM image of particles of sample *i6*.

invasiveness, as well as high operational speed and availability for implementing directly in manuscript storage departments, makes the STEM/TEM/EDX method highly promising for studying large collections of manuscripts.

It was found that some of the ink samples under study contain iron oxides, which can be crystallized in the form of hematite nanocrystals, while a part of the matrix remains amorphous. Another ink component is graphite, which was revealed in different samples in the form of fibers several micrometers in size or as nanoparticles, no more than 30 nm in size. In addition, the ink was found to contain a number of compounds, which can be associated with $\text{Fe}(\text{SO}_4)_3$, aluminosilicates (apparently, argillaceous minerals), and hydroxyapatites. These data demonstrate a possibility of fine determination of characteristic features of medieval Russian ink and thus dividing them into recipe groups.

A comparison of model and historical inks on paper carriers showed a satisfactory coincidence between the historical ink from the paper document and the ink prepared according to recipe 1 (samples *i1*, *i3*). The ink samples taken from the parchment are closer to model recipe 2. However, it is difficult to draw any unambiguous conclusions in this case because of the high content of parchment treatment products in the samples.

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