



MIXTURES OF 3D DISPERSE SYSTEMS WITH NANO- AND MICROPARTICLES: OPTICAL CHARACTERIZATION

The multiparameter analysis of simultaneous optical data for systems of nano- and /or microparticles (3D disperse systems, dispersions, colloids, ensembles) by presentation of system characteristics as N -dimensional vectors of optical parameters (ND -vectors) can help to elucidate changes in the state of the particles in systems. In this paper, the application of ND -vector approach is shown on the examples of dispersion mixtures: mixture of influenza virus particles with albumin proteins (as a model of dispersions at the process of vaccine production); mixture of *Coli bacillus* and clay dispersions (as natural water model). This approach can serve as the on-line control platform for the management of technological processes with mixtures.

ABSORPTION, BACTERIA, CLAY, 3D DISPERSE SYSTEM, MICROPARTICLE, NANO-PARTICLE.

1. Introduction

Ensembles of nano- and / or microparticles can be considered as three-dimensional disperse systems (3DDS) with particles as a disperse phase in the dispersive medium [1]. Multiparametric analysis of optical data for 3DDS can provide further progress in detailed characterization of 3DDS with particles of different nature (biological, mineral, metallic, organic, inorganic and their mixtures). This analysis includes the following:

(i) simultaneous measurements of 3DDS by different compatible nondestructive optical methods such as refractometry, absorbance, fluorescence, light scattering (integral and differential, static and dynamic, unpolarized and polarized);

(ii) solution of inverse optical problem by different methods and technologies of data interpretation. Taking into account the optical theory [1 – 8] and some experimental results [9 – 22] served as the basis for elaboration of so-called ND -vector approach [15] as the platform for on-line control of 3DDS state.

2. Materials and methods

Our studies [9 – 22] has been focused on different 3DDS with nano- and / or microparticles (with diameter less than 10 μm) and their mixtures: proteins and nucleic acids; proteins and

polymers; liposomes and viruses; liposomes carrying various substances (X-ray contrast agents, metallic particles, enzymes, viruses, antibiotics, etc.); liquid crystals with surfactants; mixtures of *Coli bacillus* with kaolin; mixtures of anthracene with cyclodextrin [16 – 18]; samples of natural and water-supply waters; air sediments in water, etc. In this paper, the application of ND vector approach is shown through the examples of 3DDS mixtures such as:

(1) the one of biological nanoparticles of influenza viruses (strain A1-H1N1) and of albumin proteins;

(2) the one of mineral bimodal kaolin dispersions (consisted from nano- and microparticles) with biological *Coli bacillus* microparticles (strains K-802 and AB 1157 [22]).

In the previous articles [18 – 22], there was the description of main optical methods used in our studies for 3DDS characterization: spectroturbidimetry, refractometry, fluorescence, absorbency, integral light scattering, differential static and dynamic light scattering, measurements of light scattering matrix elements. The measurements of dispersions were made under the same conditions. The uncertainty was about 5–10%.

Optical particle characterization in the range of nanometers up to about ten micrometers requires sophisticated data inversion

techniques. The inverse problem can be formulated as a solution of the linear first-kind Fredholm integral equation of the finite domain (1) [4], where the measured (experimental) optical characteristic $S(x)$ is related to $f(a)$ that is the unknown particle size distribution [4]:

$$S(x) = \int_{a_{\min}}^{a_{\max}} s(x, a) f(a) da, \quad (1)$$

where a is the radius of an individual particle; a_{\min} , a_{\max} are the limiting radii of particle size distribution, $s(x, a)$ is the kernel of the equation known from the experiment or from the theory of light scattering for the individual particle with radius a .

In Eq. (1), x can be a scattering angle Θ or a wavelength λ , or a frequency ν . At $a_{\min} = 0$ and $a_{\max} = \infty$ Eq. (1) converts into the linear first-kind Fredholm integral equation of infinite domain [4]:

$$S(x) = \int_0^{\infty} s(x, a) f(a) da, \quad (2)$$

The examples of the kernel $s(x, a)$ and $S(x)$ at Eq. (2) are presented in Table 1 (based on the discussion in Ref. [4]).

In addition to Notes to Table, it is necessary to remark that in all the kernels the complex refractive index of the particle substance enters as parameter [4]. For homogeneous spherical

Table

Examples of the kernel $s(x, a)$ and $S(x)$ in Eq. (2) based on the discussion in Ref. [4]

N	x	$s(x, a)$	$S(x)$
1	λ	$\pi a^2 K_e$	$\sigma_e(\lambda) = \int_0^{\infty} \pi a^2 K_e(\lambda, a) f(a) da$
2	Θ	$(\lambda^2 / 4\pi^2) i_1$	$I_1 = \int_0^{\infty} (\lambda^2 / 4\pi^2) i_1(\Theta, a) f(a) da$
3	Θ	$(\lambda^2 / 4\pi^2) i_2$	$I_2 = \int_0^{\infty} (\lambda^2 / 4\pi^2) i_2(\Theta, a) f(a) da$
4	Θ	$(\lambda^2 / 4\pi^2) [(i_1 + i_2) / 2]$	$I(\Theta) = \int_0^{\infty} (\lambda^2 / 4\pi^2) \times$ $\times \{ [i_1(\Theta, a) + i_2(\Theta, a)] / 2 \} f(a) da$
5	λ $\Theta = \pi$	$(\lambda^2 / 4\pi^2) \{ [i_1(\pi, a) + i_2(\pi, a)] / 2 \}$	$\beta(\lambda) = \int_0^{\infty} (\lambda^2 / 4\pi^2) \times$ $\times \{ [i_1(\pi, a) + i_2(\pi, a)] / 2 \} f(a) da$

Notes: 1. K_e is the extinction coefficient of the individual particle with radius a ; $\sigma_e(\lambda)$ is the cross-section of light extinction by 3DDS.

2. $s(x, a)$ is the intensity of light scattered at angle Θ by individual particle at the incident light polarized perpendicular to the surface of scattered light measurements; $S(x)$ is the intensity of light scattered by 3DDS at the incident light polarized perpendicular to the surface of scattered light measurements.

3. $s(x, a)$ is the intensity of light scattered at angle Θ by individual particle at the incident light polarized parallel to the surface of scattered light measurements; $S(x)$ is the intensity of the light scattered by 3DDS at the incident light polarized parallel to the surface of scattered light measurements.

4. $s(x, a)$ is the intensity of unpolarized light scattered at angle Θ by individual particle or so-called indicatrix of the individual particle, i.e. the S_{11} element of light scattering matrix; $S(x)$ is $S_{11}(\Theta)$, i.e. the indicatrix of 3DDS.

5. $S(x)$ is $\beta(\lambda)$, wavelength dependence of indicatrix for $\Theta = \pi$, so-called the volume coefficient of back scattering.

particles, the kernel $s(x, a)$ can be calculated according to Mie theory [1 – 4]. In the previous our papers, it was discussed the 3DDS problem of polydispersity and polymodality [21] and the polarization measurements information possibility [22] for 3DDS characterization. For polymodal polydisperse 3DDS, at the solution of inverse light scattering problem the regularization technique is often used [1 – 8]. The information-statistical methodology [23, 24] also can be used for characterization of complex 3DDS.

3. Results and discussion

The experience suggests that the set of optical parameters of so-called “the second class” [15] (obtained by processing of measured values and independent on the concentration of particles) is unique for each 3DDS [15]. In other words, each 3DDS can be characterized by N -dimensional vector in the N -dimensional space of “the second class” optical parameters (ND -vector) [15]. In our previous paper [18], mixtures of anthracene with cyclodextrin were characterized by four-dimensional (4D) vectors. It was supposed in Ref. [18] that the position of mixture ND -vector on the line connecting the separate component ND -vector points could be the justification that there is no interaction between particles in the mixture.

The mixture of pure nanoparticles of influenza virus (strain A1-H1N1) and albumin proteins (as an example of impurity) can be considered as a model of dispersions in the technological process of vaccine production. The influenza virus particle was approximated as a homogeneous sphere with the mean diameter $d = 100$ nm. To determine the virus concentration, the bilayer sphere approximation [1] was used. In order to design the optimal scheme for dispersions’ on-line control, the 3D-vector (based on light extinction parameters [21]) was suggested for differentiation of influenza virus dispersion, albumin dispersion and their mixture (Fig. 1). In the vaccine production process, it is important to know the degree of virus dispersions purification from the protein impurities. It can be concluded from Fig. 1, that the positions of 3D-vectors $\mathbf{P} \{P_i, P_k, P_m\}$ for mixture constituent dispersions (points 1 and 2) are suitable for differentiation of these dispersions

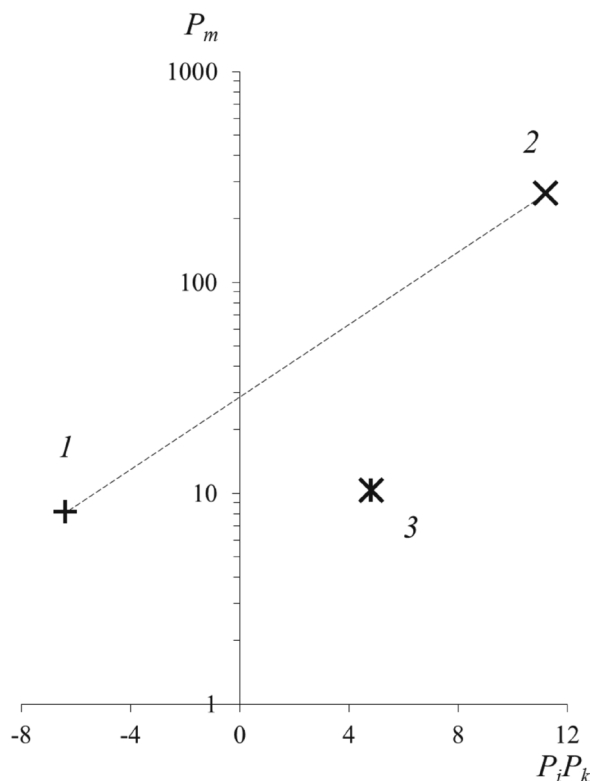


Fig. 1. An example of optical data presentation as 3D-vectors $\mathbf{P} \{P_i, P_k, P_m\}$ for dispersions of biological nanoparticles: 1 – influenza virus dispersion (strain A1-H1N1), 2 – albumin dispersion, 3 – the mixture of influenza virus and albumin dispersions in ratio 1 : 1

not only by the value but also by the sign. The preparation of vaccine will be better if the position of 3D-vector for mixture (Fig. 1, point 3) will be closer to the influenza virus 3D-vector position (Fig. 1, point 1). It is also possible to suppose from the mixture 3D-vector position (Fig. 1, point 3) apart of the line connected constituent dispersions vectors, that there are interactions between virus particles and protein molecules in mixtures.

The mixture of biological *Coli bacillus* microparticles with mineral bimodal kaolin dispersions (consisted from nano- and microparticles) could be considered as the natural water model. Mineral bimodal kaolin dispersions were characterized by different methods [21]. The justifications of mainly different forms of particles in different modes of size distribution for kaolin 3DDS were obtained at polariza-

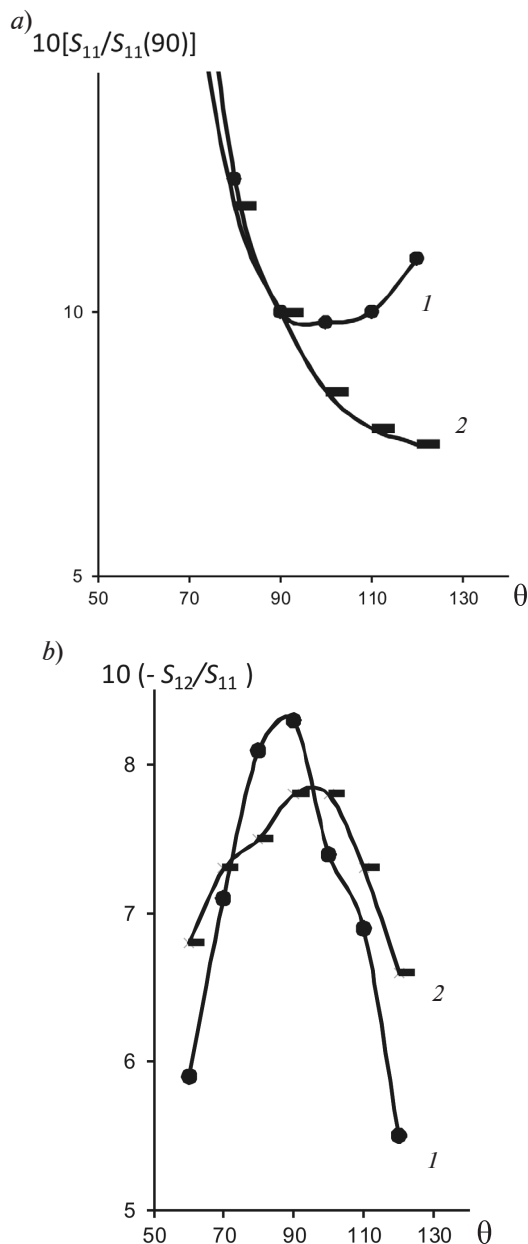


Fig. 2. Plots of the 90°-normalized S_{11} (a) and $-S_{12}/S_{11}$ (b) versus scattering angle θ in degrees (S_{11} is the first element of light scattering matrix, S_{12}/S_{11} is the polarization of scattered light [3]) for two modes (1, 2) of bimodal kaolin dispersion; $n(500) = 1.5$ (the 1st mode); $n(500) = 0.2$ (the 2nd one)

tion measurements at angles θ about 90 degrees (Fig. 2). The shifts of the positions of the $S_{11}(\theta)$ minimum in Fig. 2, a and of the maximum of scattered light polarization ($-S_{12}/S_{11}$) in

Fig. 2, b for fraction of “coarse” particles (curves 2) to $\theta > 90^\circ$ are the evidence that there are aspheric particles [3, 5, 6] in kaolin dispersions. The kaolin nanoparticles (the first mode [21]) can be approximated as the homogeneous spheres and the kaolin microparticles (the second mode and “tail” of particle mass distributions [21]) – as the homogeneous oblate (based on electronic microscopy data) ellipsoids of rotation [11].

Coli bacillus bacterial cells (*Escherichia coli*, *E. coli*, *Coli bacillus* rods) were approximated as a homogeneous prolate ellipsoids of rotation and as a volume-equivalent spheres with the mean diameter of cells, $d = 1.0 \mu\text{m}$ for strain K-802 and $d = 1.3 \mu\text{m}$ for strain AB 1157 (Figs. 3 – 6).

For the mixtures of *Coli bacillus* and mineral bimodal kaolin dispersions with nano- and microparticles (Fig. 3, based on the integral and differential static light scattering parameters

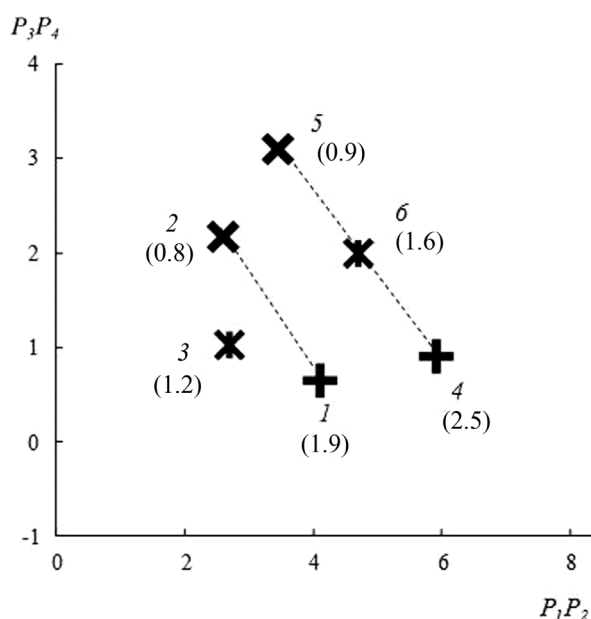


Fig. 3. Examples of optical characterization of two 3DDS mixtures (in ratio 1 : 1) and their constituents as 4D-vectors $\mathbf{P} \{P_1, P_2, P_3, P_4\}$: (i) – dispersion of *E. coli* strain AB 1157 (1), kaolin dispersion with $n(500) = 0.8$ (2) and their mixture (3); (ii) dispersions of *E. coli* strain K-802(4), kaolin dispersion with $n(500) = 0.9$ (5) and their mixture (6)

Numbers in brackets are the $n(500)$ values for corresponding dispersions

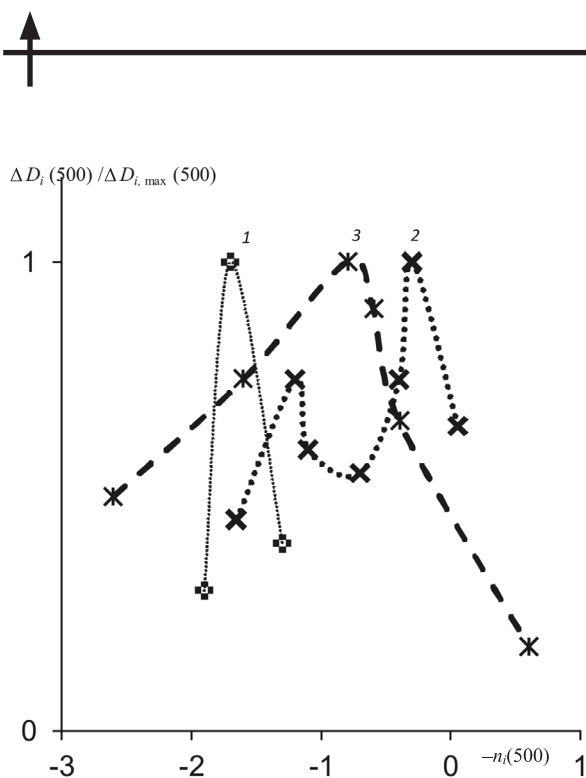


Fig. 4. Plots of $\Delta D_i(500) / \Delta D_{i,max}(500)$ versus $(-n_i(500))$ value registered at dispersions sedimentation (some sort of analog to particle size distribution); the data presented: *E. coli* strain AB 1157 (1) ($n(500) = 1.9$); kaolin dispersions (2) ($n(500) = 0.8$); their mixture (3) in ratio 1 : 1 ($n(500) = 1.2$)

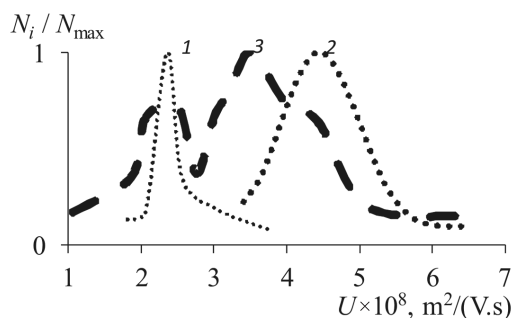


Fig. 5. Particle number distributions from particle electrophoretic mobility (U) for the same dispersions relative to those in Fig. 4. The form of mixture distribution is the evidence in favor of kaolin and bacteria heteroaggregation

[21]) according to Ref. [18] one can observe both situations, i.e. the supposed interaction between constituent dispersions for mixture with $n(500) = 1.2$ and the absence of that for mixture with $n(500) = 1.6$. The detailed analysis of the data for the mixture of kaolin with *E.coli* strain AB 1157 (Fig. 4 and 5) showed that in

this dispersion there is an interaction between the particles: the form of mixture distributions is the evidence in favor of kaolin and bacteria heteroaggregation.

In Fig. 6, the 16D-vectors (based on the light scattering matrix parameters [22]) of kaolin dispersion, *E. coli* K-802 dispersion and of their mixture are presented. It can be seen that in 16D parameter space the differentiation of dispersion vector positions is about several orders and that the “non-supposed” (according to 4D-vector approach in Fig. 3) interaction between bacterial and kaolin particles can also occur. The data of polarization measurements for kaolin 3DDS (Fig. 2) allows to predict that (the prolate bacterial – kaolin “small” spherical) particle interaction and (the prolate bacterial – “coarse” oblate) particle one can be different. In addition to the discussion in Ref. [21] about natural 3DDS polymodality taking into account the shape of the particles makes the model for solving inverse problem of mixtures more complex.

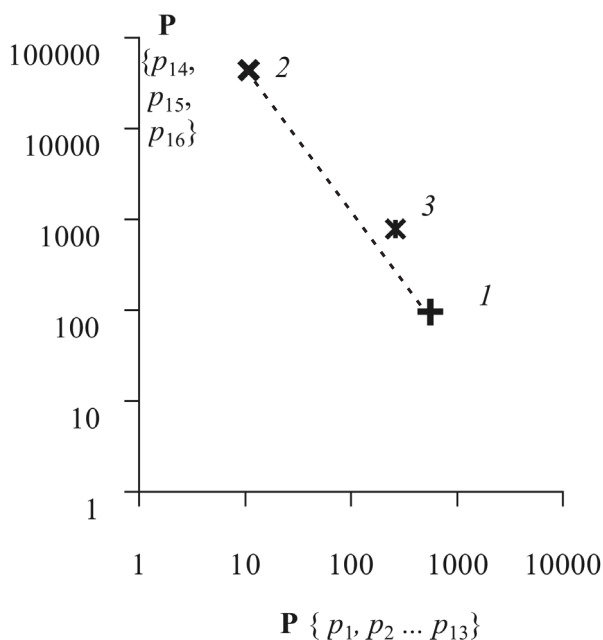


Fig. 6. The optical characteristics as 16D-vectors $\mathbf{P} \{p_1, p_2, \dots, p_{16}\}$ for dispersions with “non-supposed interaction of particles in mixture” (according to 4D-vectors approach in Fig. 3): dispersion of *E. coli* (1) strain K-802 ($n(500) = 2.5$); kaolin dispersion (2) ($n(500) = 0.9$) and their mixture (3) in ratio 1:1 ($n(500) = 1.6$)

4. Summary

For many technological processes, the information about the integral state of the 3DDS and about the changes of its state at any influence can be sufficient for management. *ND*-vectors can reflect the changes in the state of the mixtures. In this case, the polymodality of particle size distributions [21] and the difference of particle forms are no obstacle. Combination of *ND*-vector's approach with other methods of inverse problem solution can help to investigate the processes in 3DDS such as aggregation, disaggregation, coalescence, heteroaggregation, sedimentation, etc. The pro-

posed approach allows the study of any 3DDS as an intact non-destroyable unity, with the minimal interference. It can demonstrate the unique potentials of solving problems of polymer science, bio- and nanotechnology, medicine and of environmental protection.

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Безрукова А.Г., Власова О.Л. ОПТИЧЕСКАЯ ХАРАКТЕРИСТИКА СМЕСЕЙ 3D ДИСПЕРСНЫХ СИСТЕМ НАНО- И МИКРОЧАСТИЦ.

Многопараметрический анализ одновременных оптических измерений для систем нано- и/или микрочастиц (3D-дисперсные системы, дисперсии, коллоиды, ансамбли частиц) с помощью представления характеристик систем как N -мерных векторов оптических параметров (ND -векторов) может помочь выявить изменения состояния частиц в системах. В представленной статье применение ND -векторного подхода показано на примерах смесей дисперсий: смесь частиц вируса гриппа с молекулами белка альбумина (модель дисперсий в процессе производства вакцин); смесь дисперсий кишечной палочки и глины (модель природной воды). Этот подход может служить для контроля он-лайн и управления технологическими процессами с участием 3D дисперсных систем и их смесей.

БАКТЕРИЯ, ND -ВЕКТОР, ВИРУС, ГЛИНА, 3D-ДИСПЕРСНАЯ СИСТЕМА, НАНОЧАСТИЦА, ПОГЛОЩЕНИЕ, СВЕТОРАССЕЯНИЕ.

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