

UDC 546.284-31:612.111

INTERACTION OF RED BLOOD CELLS WITH FUMED SiO_2 , $\text{Al}_2\text{O}_3/\text{SiO}_2$ AND $\text{TiO}_2/\text{SiO}_2$ BY LIGHT SCATTERING MEASUREMENTS

I.I. Gerashchenko^{1*}, V.M. Gun'ko¹, V.I. Zarko¹, B.I. Gerashchenko²,
L.S. Andriyko¹, V.F. Gorchev³

¹*Chuiko Institute of Surface Chemistry of National Academy of Sciences of Ukraine
17 General Naumov Street, Kyiv 03164, Ukraine*

²*Kavetsky Institute of Experimental Pathology, Oncology, and Radiobiology of NAS of Ukraine
45 Vasilkovskaya Street, Kyiv 03022, Ukraine*

³*Palladin Institute of Biochemistry of National Academy of Sciences of Ukraine,
9 Leontovych Street, Kiev 01601, Ukraine*

The interaction of human red blood cells (RBCs) with fumed silica and fumed X/SiO_2 ($X = \text{Al}_2\text{O}_3$, TiO_2) at different concentrations of X oxide was studied by flow cytometry and photon correlation spectroscopy. The light scattering of RBCs affected by oxides in conjunction with the hemolysis degree showed that mixed oxides (X/SiO_2), in general, had less membranotoxic effect than pure silica. The interaction of RBCs with fumed silica and other mixed oxides is a convenient model to examine membranotoxicity and biocompatibility of disperse materials. Flow cytometry and photon correlation spectroscopy are informative methods to study the mechanism of hemolysis induced by solid micro- or nanoparticles.

INTRODUCTION

The interaction of highly disperse mineral particles with human cells is of interest from practical and theoretical points of view in terms of health protection and environmental chemistry and biochemistry [1, 2]. Elucidation of the interaction mechanism between cells and inorganic membranophilic substances such as silica micro- and nanoparticles is one of the problems of membrane biology. According to Paoletti [3], certain disperse mineral particles can reach the alveoli and remain there for years without losing their chemical properties frequently being very hazardous for human health. The chemical properties of surfaces of both solid particles and cell membranes can predetermine the fate of particle-cell interaction that in some circumstances (depending upon physical chemistry and timing of this interaction, as well as type of cell response) can trigger disease. For instance, disperse crystalline silica particles can contribute to the development of pulmonary disease (silicosis). Amorphous silica is much less hazardous than crystalline silica [2, 4].

On the other hand, mineral micro- and nanoparticles can be used as medicinal adsorbents, drug fillers and carriers [5]. Controlled changes in the surface properties of solid particles of adsorbents, e.g. due to preparation of mixed oxides such as fumed alumina/silica or titania/silica (instead of pure silica), is an attractive approach to improve biocompatibility of aforementioned materials. A test of damage of RBCs (hemolysis) upon interaction with disperse solids can be used as a simple model to study the biocompatibility of these materials [6].

The fact that silica induces hemolysis shows that it can change permeability of biomembranes. Although the precise mechanism of membranolysis remains uncertain, the electrostatic factor is thought to play an important role in the interaction of silica particles with biomembranes. The surface of a colloidal silica particle is known to be negatively charged at the physiological pH due to dissociation of silanol groups [7]. The total surface charge of human RBCs is also negative due to the negative charge of the carboxyl groups of N-acetylneuraminic acid (sialic acid) residues covalently attached to membrane glycoproteins [8, 9]. In order to contact with the cell membrane,

* Corresponding author igorgera@mail.ru

a silica particle has to overcome the electrostatic repulsion barrier. There is evidence that the sensitivity of the cell membrane to silica depends upon the sialic acid content [10–12]. Silica particles have been found to interact with the erythrocyte membrane through the positively charged tetraalkylammonium-containing phospholipids [13]. The interaction of membrane proteins with silica particles is also essential in the mechanism of hemolysis [14]. Taking the electrostatic mechanism of silica–membrane interaction as a basis (including hydrogen bonding with the main contribution of electrostatic component), it has been hypothesized that colloidal silica can selectively eliminate RBCs due to differences in the architecture of the cell surface groups [15].

For confirmation of the main role of the electrostatic factor in the mechanism of membranolysis it is interesting to track as hemolytic properties of silica/alumina (SA) or silica/titania (ST) will vary in comparison with those of fumed silica. Isoelectric point (IEP), point of zero charge (PZC), and density of surface charge of mixed oxides differ from those of silica [16–20]. Additionally, mixed oxides are characterized by significant nonuniformity of the surface which depends upon the second phase content and its distribution on the surface [16–19, 21]. New active surface sites such as Brønsted and Lewis acid sites can also appear on the surface of mixed oxides [16–19].

In the present study, we demonstrate that RBCs are likely to be a sensitive for detecting changes in the surface properties of oxide particles. Interactions of RBCs with highly disperse mixed fumed oxides X/SiO₂ (X = Al₂O₃, TiO₂) at different X concentrations in the aqueous suspension were studied analyzing the light scattering properties of RBCs together with their lysis level chosen as a criterion to evaluate the oxide-induced effects. The light scatter of RBCs interacting with fumed silica was previously analyzed by means of flow cytometry [22–25]. The size of oxide particles and changes in the size of RBCs affected by fumed oxides could be studied using photon correlation spectroscopy (PCS).

MATERIALS AND METHODS

Highly disperse fumed silica (Aerosil A300), fumed X/SiO₂ (X = Al₂O₃ and TiO₂) at different concentrations of X oxide (Table 1) were synthesized by hydrolysis of SiCl₄ or SiCl₄/MCl_n (M = Al or Ti) mixtures in oxy-

gen/nitrogen/hydrogen flame at 1300–1700 K (pilot plant of the Institute of Surface Chemistry, Kalush, Ukraine).

Table 1. The examined samples

Al ₂ O ₃ / SiO ₂ system		TiO ₂ / SiO ₂ system	
oxide	S _{BET} , m ² /g	oxide	S _{BET} , m ² /g
A-300 (SiO ₂)	297	ST ₁	313
SA ₁	203	ST ₂	104
SA ₃	185	ST ₉	235
SA ₈	303	ST ₁₄	156
SA ₂₃	347	ST ₂₀	84
SA ₃₀	239	ST ₄₀	148
SA ₇₅	118	ST ₆₃	83
Al ₂ O ₃	125	ST ₉₄	30
		TiO ₂	42

Content of alumina (wt.%) in SA and titania (wt.%) in ST is shown as a subscript

To prepare fumed oxide suspensions 0.25 g of dry oxide powder was added to 50 ml of distilled water and stirred. To break up agglomerates, the suspensions were sonicated using an ultrasonic disperser (power 500 W and frequency 22 kHz) for 5 min. Prior to the reaction with RBCs, 40 μl aliquot of the oxide suspension was diluted in 2.0 ml of phosphate-buffered saline, PBS (137 mM NaCl, 2.7 mM KCl, 1.5 mM KH₂PO₄, 8.0 mM Na₂HPO₄, pH 7.3) to final concentration of oxide of 0.01%.

Erythrocyte mass from a healthy volunteer was washed three times in PBS at 300 g for 10 min at room temperature, and 40 μl aliquot of the RBC suspension containing approximately 10⁶ cells was added to the fumed oxide suspension and gently mixed. As a control, the RBCs were added to 2 ml of PBS. After 10 and 30 min of incubation at room temperature, the fumed oxides-RBC reaction mixtures were centrifuged at 300 g for 5 min at room temperature. The quantity of released hemoglobin was determined photometrically in the supernatant at wavelength of 405 nm using a SF-26 (LOMO, Russia) spectrophotometer. The degree of hemolysis in these samples was calculated as the relation of measured optical density to that of a sample with 100% hemolysis. The degree of hemolysis in each sample was averaged on the basis of results of two independent measurements.

The forward light scatter (FSC parameter) of cells interacting with disperse oxides was recorded after 1, 5, 10, 20, and 30 min of the reaction using a FACStar Plus flow cytometer (Becton Dickinson Immunocytometry Systems, San

Jose, CA) equipped with a 15 mW argon-ion laser (488 nm). The FSC signals were collected in a linear mode. Analysis of the data was performed with Consort 40 software (Becton Dickinson).

Particle size distributions (PSDs) were studied using a Zetasizer 3 (Malvern Instruments) apparatus ($\lambda = 633$ nm, $\Theta = 90^\circ$). Malvern Instruments software was used to compute the PSDs and D_{ef} (average hydrodynamic diameter, i.e. the particle diameter plus the double shear layer thickness), assuming that particles had a roughly spherical shape.

RESULTS AND DISCUSSION

Fumed oxides are characterized by a stepwise structural hierarchy: primary particles (their average size d and specific surface area S_{BET} are linked by the relationship as $S_{BET} \sim A/(\rho d)$ where A is a constant and ρ is the specific density of the material) form aggregates (50–500 nm in size, bulk density ρ_b corresponds to approximately $\rho/3$) which then form agglomerates (>1 μm , $\rho_b \approx 0.05$ – 0.1ρ). The bulk density of fumed silica or alumina/silica powder is 0.03–0.06 g/cm³ what corresponds to a large value of empty volume in the powder ($V_{emp} = 15$ – 30 cm³/g). Fumed silica/titania has larger ρ_b and smaller V_{emp} values due to higher specific density of titania than silica [16–19]. The adsorption capacity of fumed oxides with respect to low molecular compounds (nonpolar such as nitrogen, benzene, hexane, etc., or polar such as water, methanol, ethanol, etc.) is relatively low [0.1–1.0 cm³ (depending upon aggregation degree of primary particles of liquid adsorbate per gram of the adsorbent)], since adsorbate molecules mainly fill contact zones between primary particles in aggregates and form a thin layer on the outer surface of the aggregates. The adsorption capacity of fumed oxides with respect to polymers and biomacromolecules corresponds to similar values, i.e. 0.1–1.0 gram of adsorbate per gram of adsorbent [26–30]. However, the structure of the interfacial layer in the case of adsorption of low and high molecular compounds significantly differs [16, 19, 26–30]. The main adsorption sites at a surface of silica and mixed oxides are hydroxyl groups such as terminal MOH and bridging M_1 –O(H)– M_2 ones. Lewis (L) acid sites (incompletely O-coordinated metal atoms) can play an important role in the adsorption at the surface of mixed oxides as well as titania and alumina. However, the L sites (as well as bridging hydroxyls) are absent at

silica surface. Additionally, the second phase (alumina or titania) may be nonuniformly distributed on the surface of primary particles [21]. For instance, in the case of SA samples the maximal value of the surface content of Al atoms at total $C_{Al_2O_3} \leq 30$ wt.% corresponds to $C_{Al_2O_3} = 8$ wt.% (Fig. 1). It is obvious that the aforementioned morphological and structural features of silica and mixed oxides can affect their interaction with biomacromolecules and cell (e.g. RBC) membranes [26–30].

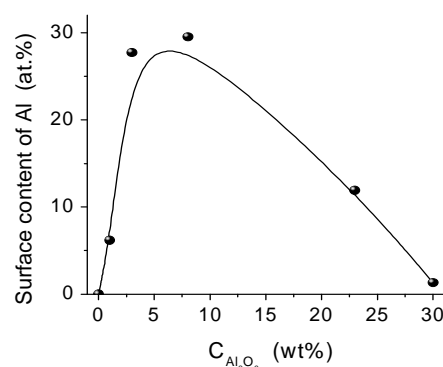
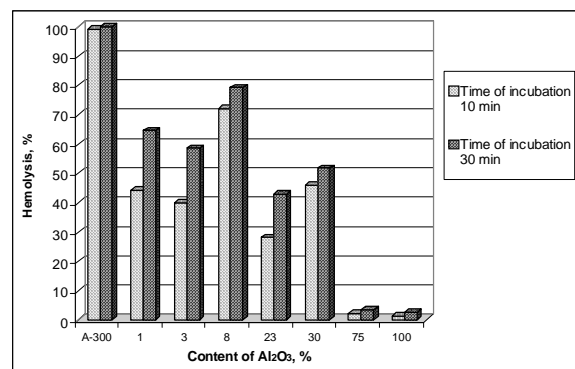
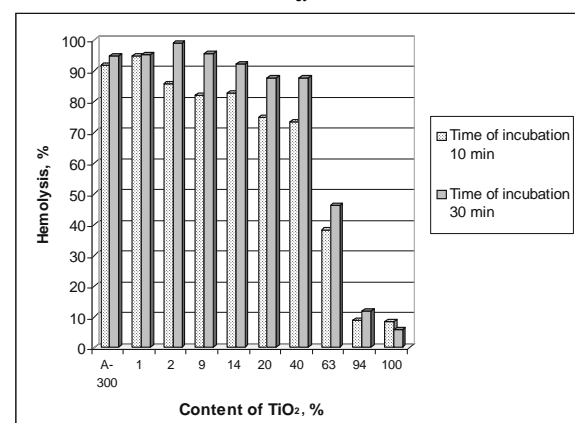


Fig. 1. Surface content of Al atoms in Al₂O₃/SiO₂ systems



a



b

Fig. 2. Rate of hemolysis (in %) as a function of Al₂O₃/SiO₂ (a) and TiO₂/SiO₂ (b) ratio in the mixed oxide

Fig. 2a, b presents the hemolysis degree estimated from photometric measurements of the levels of hemoglobin that was released after 10 and 30 min incubation of 10^6 RBCs in the aqueous suspensions (0.01%) of alumina/silica and titania/silica.

Fumed mixed oxides showed a gradual decrease in the hemolytic activity with increasing Al_2O_3 or TiO_2 content (excepting SA_8 and SA_{30} , Fig. 2a). The electrical behavior of the surface of mixed oxides depends upon their composition but nonlinearly because the presence of the second oxide results in the formation of new sites (such as Si-O(H)-M at $\text{M}=\text{Al}$ or Ti) which are absent at surface of individual oxides. Additionally, the distribution of alumina or titania at a surface of primary particles is nonuniform [16–19, 21]. Therefore, IEP and PZC of mixed oxides can be at lower pH values than that of silica (pH 2.2) despite $\text{pH}_{\text{IEP}} \approx 9.8$ (alumina) and ≈ 6 (titania) [16]. Only in the case of a low content of silica in mixed oxides IEP and PZC shift towards higher pH values (Fig. 3).

Changes in the nature of the surface of mixed oxides cause an increase in the adsorption of proteins in gram per sq. m of the adsorbent [26–30]. It should be noted that the oxides used in this study are non-ionic. Therefore, the relative number of the surface deprotonated hydroxyls as well as the surface charge density is not high (Fig. 3). This can lead to preferable contribution of the hydrogen bonds to the interaction of proteins and other biostructures with oxide nanoparticles. The sizes of primary particles (or specific surface area) can play an important role in the interaction with cell membranes. In this contest it should be pointed out the nonuniform distribution of charge density on membranes and the aggregation character of primary particles (larger the size of particles, less their aggregation [16, 30]).

The RBCs that were interacted with fumed silica (A-300) for up to 1 min, compared to the intact RBCs, showed a verifiable difference in the distribution of FSC signals. This difference increases with time (Fig. 4a). An increase of FSC from affected RBCs is likely due to increase of their average size accompanied by transformation of cell shape from discoid to spheroid. Appearance of events with low FSC can be explained by accumulation of collapsed cells (cell debris) due to the active destruction of RBCs. Significant changes of FSC were also observed for RBCs that

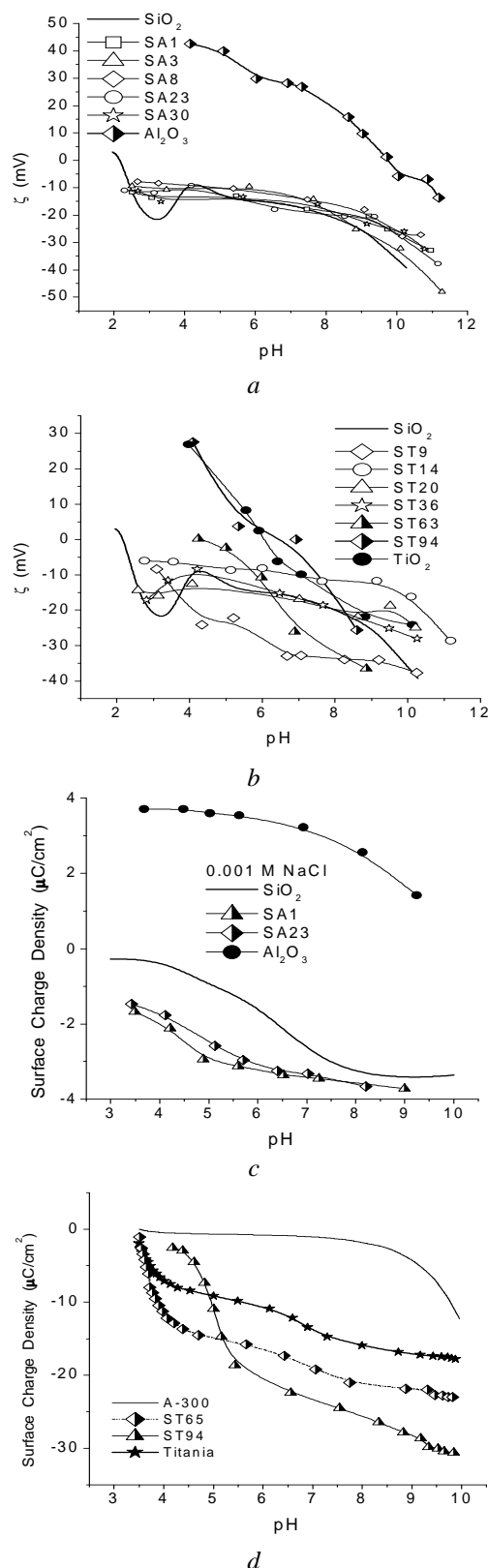


Fig. 3. Electrokinetic potential (ζ) of (a) silica, alumina and SA, and (b) titania and ST; surface charge density of (c) silica, alumina and SA, and (d) titania and ST

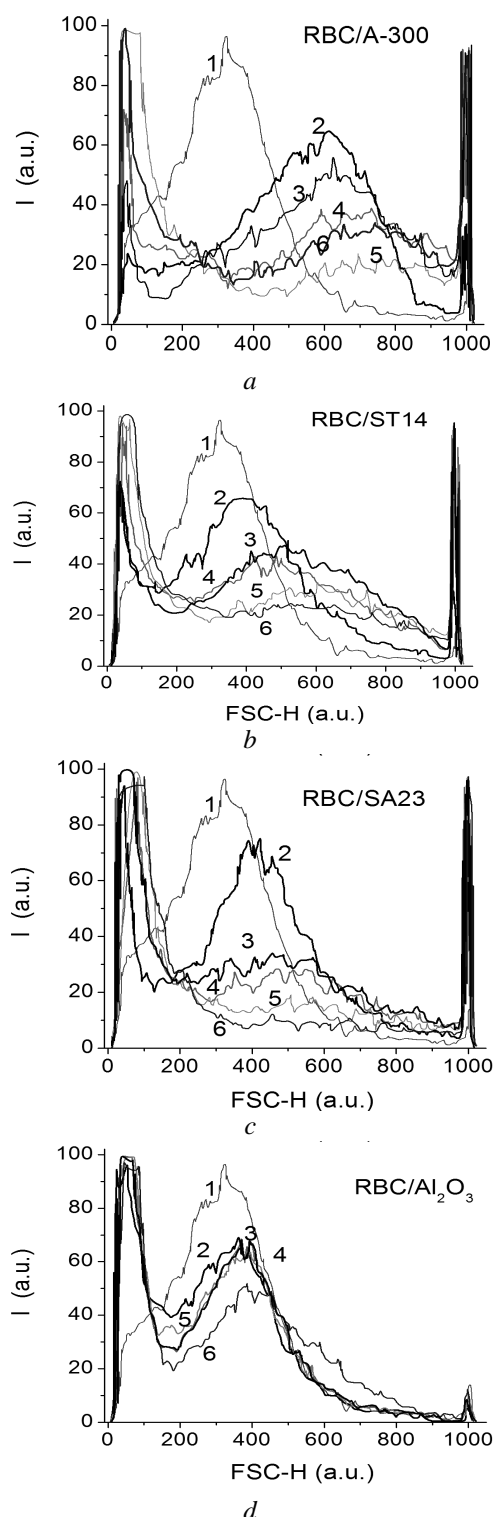


Fig. 4. FSC histogram analysis of intact RBCs (curves 1) and RBCs interacting with fumed oxides for up to 1 (2), 5 (3), 10 (4), 20 (5), and 30 (6) min after addition of 2×10^6 cells to 0.01% suspensions of oxides: (a) fumed silica A-300; (b) ST₁₄; (c) SA₂₃; (d) fumed Al_2O_3 . Abscissa corresponds to the signal intensity (a.u.). Ordinate corresponds to the signal density (a.u.)

interacted with certain alumina/silica and titania/silica oxides with moderate content of Al_2O_3 or TiO_2 (Fig. 4b, c). However, the character of distributions of FSC signals from these cells showed that they gained their size less readily than cells interacting with A-300 (Fig. 4a vs. Fig. 4b, c).

The light scatter of the RBCs after interaction with mixed oxides with high content of alumina or titania changes slightly with observation time. This effect is shown by the example of alumina (Fig. 4d). In general, changes in the light scattering by the RBCs interacting with fumed oxides are in accordance with the data on hemolysis. That is, an increase of FSC of cells interacting with oxides, which reflects an enlargement of cells prior to their collapse, is correlated with the degree of hemolysis.

Thus, the samples of both $\text{Al}_2\text{O}_3/\text{SiO}_2$ and $\text{TiO}_2/\text{SiO}_2$ systems could be divided into three groups: (i) fumed silica A-300 with the maximal damage impact on the RBC structure, and with the high hemolytic effect; (ii) 1–30 wt.% Al_2O_3 in SA and 1–65 wt.% TiO_2 in ST giving smaller structural injury but high hemolytic effect; and (iii) 75–100 wt.% Al_2O_3 and 94–100 wt.% TiO_2 weakly affecting the RBCs.

According to PCS measurements, aqueous suspensions of SA samples were characterized by mono-, bi- and trimodal distributions of the particle size (Table 2). The smallest particles (d_1 between 50 and 100 nm) that were detected in the suspensions corresponded to aggregates of primary particles.

Table 2. Characteristics of the particle size distribution in the intact $\text{Al}_2\text{O}_3/\text{SiO}_2$ suspensions

Oxide	$d_{\text{average}}, \text{nm}$	d_1, nm	d_2, nm	d_3, nm
A-300 (SiO_2)	185.4	98.1 (15%)	205.6 (76%)	498 (6%)
SA ₁	194.5	76.7 (14%)	275.9 (86%)	–
SA ₃	246.5	91.0 (16%)	278.0 (75%)	829.9 (10%)
SA ₈	246.8	99.3 (14%)	254.5 (76%)	1753.3 (10%)
SA ₂₃	240.9	81.4 (10%)	158.6 (25%)	546.6 (65%)
SA ₃₀	217.2	94.3 (14%)	254.6 (86%)	–
SA ₇₅	148.5	88.0 (11%)	148.6 (76%)	1750 (10%)
Al_2O_3	131.0	72.9 (15%)	149.9 (85%)	–

The ST samples were more polydisperse with trimodal PSDs (Table 3). A significant portion of secondary ST particles at high C_{TiO_2} values had the size larger than 1.5 μm .

Table 3. Characteristics of the particle size distribution in the intact $\text{TiO}_2/\text{SiO}_2$ suspensions

Oxide	d_{average} , nm	d_1 , nm	d_2 , nm	d_3 , nm
A-300 (SiO_2)	185.4	98.1 (15%)	205.6 (76%)	498 (6%)
ST ₁	194.1	109.4 (16%)	217.6 (78%)	420.8 (7%)
ST ₂	289.3	130.9 (18%)	397.7 (82%)	829.9 (10%)
ST ₉	286.0	109.4 (15%)	352.9 (74%)	2014.1 (11%)
ST ₁₄	623.0	377.9 (12%)	640.3 (74%)	1042.5 (14%)
ST ₂₀	309.6	145.2 (8%)	276.1 (65%)	911.7 (26%)
ST ₄₀	273.6	149.0 (13%)	270.0 (71%)	1141.1 (15%)
ST ₆₃	791.7	93.7 (8%)	2345.3 (92%)	–
ST ₉₄	869.0	334.8 (24%)	1711.9 (76%)	–
TiO_2	1655.4	417.8 (11%)	1009.7 (18%)	4019.7 (71%)

The interaction dynamics of RBCs with fumed silica (A-300) is shown in Fig. 5.

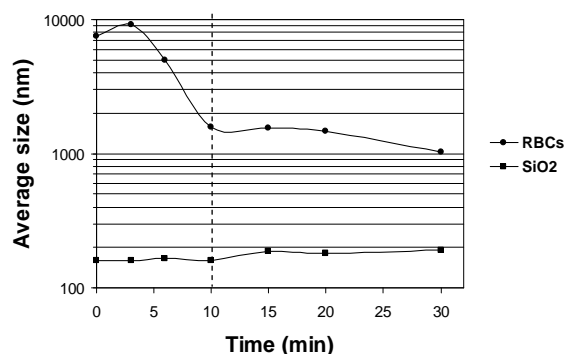


Fig. 5. Time-dependent changes in the average sizes of interacting RBCs and SiO_2 particles. Dashed line indicates the end of rapid phase of interaction ($\approx 90\%$ cell lysis)

Within 0–3 min of interaction, the average diameter of RBCs increased from 7.5 to 9.2 μm (fast stage when cells actively change their shape from discoid to spheroid). Further incubation of silica-cell mixture up to 6 min led to a considerable reduction of cell sizes ($d = 4.1\text{--}5.4 \mu\text{m}$). This effect can be explained by irreversible alterations in the cell membranes and active cell collapse. Then,

after 10 min of incubation of silica-cell mixture, the vast majority of the objects detected in the suspension were probably cell fragments. As for silica, within 0–30 min of interaction with RBCs, the average size of aggregates of silica particles slightly increased from 160 to 192 nm which can be explained by adsorption of biomolecules (released from collapsed RBCs) on silica particles.

CONCLUSION

Results obtained using three spectral methods are coherent and give clear picture of fumed oxide-induced hemolysis of the RBCs observed *in vitro*. This methodological approach can be useful for investigations of the mechanism of membranotoxicity and biocompatibility of disperse materials to be used for medicinal purposes. Obtained results reveal that the membranotoxicity of fumed oxides decreases with increasing content of alumina in silica/alumina or titania in silica/titania. It confirms the main role of the electrostatic factor in the mechanism of membranolysis because of isoelectric point and point of zero charge as well as density of surface charge of mixed oxides differ from those of silica. Other factors, such as size distribution of primary and secondary particles, content and type of active surface sites, surface content of the second oxide, etc., are likely to contribute in this mechanism.

REFERENCES

1. Seaton A., MacNee W., Donaldson K., Godden D. Particulate air pollution and acute health effects // *Lancet*. – 1995. – V. 345, N 1. – P. 176–178.
2. Fubini B. Health effects of silica // *The surface properties of silicas* / Ed. A.P. Legrand. – Chichester; New York; Toronto: J. Wiley&Sons, 1998. – P. 415–464.
3. Paoletti L. Mineral particles in bronchoalveolar lavage fluid (BALF): an attempt at designing a quantitative model // *Arch. Environ. Health*. – 1997. – V. 52. – P. 384–389.
4. Stratta P., Canavese C., Messuerotti A. et al. Silica and renal diseases: no longer a problem in the 21st century? // *J. Nephrol.* – 2001. – V. 14. – P. 228–247.
5. *Medical Chemistry and Clinical Use of Silicium Dioxide* / Ed. A.A. Chuiko. – Kiev: Naukova Dumka, 2003. – 416 p.
6. Harley J.D., Margolis J. Hemolytic activity of colloidal silica // *Nature*. – 1961. – V. 189. – P. 1010–1011.

7. Iler R.K. The chemistry of silica: solubility, polymerization, colloid and surface properties, and biochemistry. – New York: J. Wiley & Sons, 1979. – 896 p.
8. Cook G.M.W., Heard D.H., Seaman G.V.F. Sialic acids and the electrokinetic charge of the human erythrocyte // *Nature*. – 1961. – V. 191. – P. 44–47.
9. Mehrishi J.N. Molecular aspects of the mammalian cell surface // *Progr. Biophys. Mol. Biol.* – 1972. – V. 25. – P. 1–70.
10. Depasse J. Influence of the sialic acid content of membranes on their sensitivity to silica and aluminate-modified silica // *Environ. Res.* – 1978. – V. 16. – P. 88–91.
11. Depasse J., Ghysdael J., Léonis J. Effect de la charge des érythrocyte sur leur sensibilité à la silice // *Arch. Intern. Physiol. Biochem.* – 1974. – V. 82. – P. 797.
12. Cantrell A.C., Ellis P. Reaction of colloidal silica with membranes of intact mammalian cells // *Chem. Biol. Interactions*. – 1983. – V. 44. – P. 169–183.
13. Depasse J., Warlus J. Relation between the toxicity of silica and its affinity for tetraalkylammonium groups // *J. Colloid Interface Sci.* – 1976. – V. 56, N 3. – P. 618–621.
14. Diociaiuti M., Bordi F., Gataleta L. et al. Morphological and functional alterations of human erythrocytes induced by SiO₂ particles: an electron microscopy and dielectric study // *Environ. Res. Section*. – 1999. – V. 80. – P. 197–207.
15. Gerashchenko B.I., Gerashchenko I.I., Pantazis C.G. Possible selective elimination of red blood cells under the influence of colloidal silica // *Medical Hypotheses*. – 1996. – V. 47. – P. 69–70.
16. Gun'ko V.M., Zarko V.I., Leboda R., Chibowski E. Aqueous suspensions of fumed oxides: particle size distribution and zeta potential // *Adv. Colloid. Interface Sci.* – 2001. – V. 91. – P. 1–112.
17. Gun'ko V.M., Turov V.V., Zarko V.I. et al. Active site nature of pyrogenic alumina/silica and water bound to surfaces // *Langmuir*. – 1997. – V. 13. – P. 1529–1544.
18. Gun'ko V.M., Zarko V.I., Turov V.V. et al. The effect of second phase distribution in disperse X/Silica (X=Al₂O₃, TiO₂, and GeO₂) on its surface properties // *Langmuir*. – 1999. – V. 15. – P. 5694–5702.
19. Gun'ko V.M., Zarko V.I., Turov V.V. et al. Characterization of fumed alumina/silica/titania in the gas phase and aqueous suspension // *J. Colloid. Interface Sci.* – 1999. – V. 220. – P. 302–323.
20. Gun'ko V.M., Voronin E.F., Pakhlov E.M. et al. Features of fumed silica coverage with silanes having three or two groups reacting with the surface // *Colloid. Surf. A*. – 2000. – V. 166. – P. 187–201.
21. Gun'ko V.M., Zarko V.I., Mironyuk I.F. et al. Surface electric and titration behaviour of fumed oxides // *Colloids Surf. A*. – 2004. – V. 240. – P. 9–25.
22. Gerashchenko B.I., Gerashchenko I.I., Bogomaz V.I., Pantazis C.G. Adsorption of aerosil on erythrocyte surface by flow cytometry measurements // *Cytometry*. – 1994. – V. 15. – P. 80–83.
23. Gerashchenko B.I. Short note: Heterogeneous response of red blood cells to colloidal silica as a criterion for study of their membrane alterations: flow cytometric approach // *Med. Hypotheses*. – 1998. – V. 51. – P. 355–357.
24. Gerashchenko B.I., Gerashchenko I.I., Bogomaz V.I. Flow cytometric criteria for assessment of silica-cell adsorptive interaction // *Cytometry*. – 1996. – V. 23. – P. 174.
25. Gerashchenko B.I., Gun'ko V.M., Gerashchenko I.I. et al. Probing the silica surfaces by red blood cells // *Cytometry*. – 2002. – V. 49. – P. 56–61.
26. Gun'ko V.M., Turov V.V., Zarko V.I. et al. Features of aqueous suspensions of fumed silica and interaction with proteins // *J. Colloid. Interface Sci.* – 1997. – V. 192. – P. 166–178.
27. Gun'ko V.M., Vlasova N.N., Golovkova L.P. et al. Interaction of proteins and substituted aromatic drugs with highly disperse oxides in aqueous suspensions // *Colloids Surf. A*. – 2000. – V. 167. – P. 229–243.
28. Mironyuk I.F., Gun'ko V.M., Turov V.V. et al. Characterization of fumed silicas and their interaction with water and dissolved proteins // *Colloids Surf. A*. – 2001. – V. 180. – P. 87–101.
29. Gun'ko V.M., Zarko V.I., Voronin E.F. et al. Impact of some organics on structural and adsorptive characteristics of fumed silica in different media // *Langmuir*. – 2002. – V. 18. – P. 581–596.
30. Gun'ko V.M., Klyueva A.V., Levchuk Yu.N. et al. Photon correlation spectroscopy investigations of proteins // *Adv. Colloid. Interface Sci.* – 2003. – V. 105. – P. 201–328.

Received 16.09.2010, accepted 29.09.2010

**Дослідження взаємодії еритроцитів з пірогенними оксидами
 SiO_2 , $\text{Al}_2\text{O}_3/\text{SiO}_2$ та $\text{TiO}_2/\text{SiO}_2$
шляхом вимірювання параметрів світлорозсіювання**

І.І. Геращенко, В.М. Гунько, В.І. Зарко, Б.І. Геращенко, Л.С. Андрійко, В.Ф. Горчев

*Інститут хімії поверхні ім. О.О. Чуйка Національної академії наук України
вул. Генерала Наумова 17, Київ 03164, Україна*

*Інститут експериментальної патології, онкології і радіобіології ім. Р.Є. Кавецького НАН України
вул. Васильківська, 45, Київ 03022, Україна*

*Інститут біохімії ім. О.В. Палладіна Національної академії наук України
вул. Леонтовича 9, Київ 01601, Україна*

Методами проточної цитометрії та лазерної кореляційної спектроскопії досліджено взаємодію еритроцитів людини з пірогенними оксидами – немодифікованим кремнеземом і змішаними оксидами X/SiO_2 ($X = \text{Al}_2\text{O}_3, \text{TiO}_2$) з різним вмістом X . Результати вимірювання параметрів світлорозсіювання у поєднанні з визначенням ступеня гемолізу еритроцитів свідчать, що змішані оксиди в цілому проявляють меншу мембранотоксичну дію у порівнянні з немодифікованим кремнеземом. Взаємодія еритроцитів людини з пірогенним кремнеземом та іншими змішаними оксидами є зручною моделлю для тестування мембранотоксичності/біосумісності дисперсних матеріалів. Проточна цитометрія і лазерна кореляційна спектроскопія є інформативними методами для вивчення механізму гемолізу, індукованого твердими частинками.

**Исследование взаимодействия эритроцитов с пирогенными оксидами
 SiO_2 , $\text{Al}_2\text{O}_3/\text{SiO}_2$ и $\text{TiO}_2/\text{SiO}_2$
путем измерения параметров светорассеяния**

И.И. Геращенко, В.М. Гунько, В.И. Зарко, Б.И. Геращенко, Л.С. Андрійко, В.Ф. Горчев

*Інститут хімії поверхності ім. А.А. Чуйко Національної академії наук України
ул. Генерала Наумова 17, Киев 03164, Украина*

*Інститут експериментальної патології, онкології і радіобіології ім. Р.Є. Кавецького НАН України
ул. Васильковская 45, Киев 03022, Украина*

*Інститут біохімії ім. А.В. Палладіна Національної академії наук України
ул. Леонтовича 9, Киев 01601, Украина*

Методами проточної цитометрії і лазерної кореляційної спектроскопії изучено взаємодієвие эритроцитов человека с пирогенными оксидами – немодифицированным кремнеземом и смешанными оксидами X/SiO_2 ($X = \text{Al}_2\text{O}_3, \text{TiO}_2$) с разным содержанием X . Результаты измерения параметров светорассеяния совместно с определением степени гемолиза эритроцитов свидетельствуют, что смешанные оксиды в целом обладают меньшим мембранотоксическим действием в сравнении с немодифицированным кремнеземом. Взаимодействие эритроцитов человека с пирогенным кремнеземом и другими смешанными оксидами можно рассматривать как удобную модель для испытания дисперсных материалов на мембранотоксичность/ биосовместимость. Проточная цитометрия и лазерная корреляционная спектроскопия являются информативными методами для изучения механизма гемолиза, индуцированного твердыми частицами.