Ershov Vadim Alekseevich

Silver hydrosol stabilized by carbonate ions: optical characteristics of nanoparticles, oxidative dissolution and antibacterial properties

Relevance of scientific research. Currently, the synthesis and application of nanoscale metal particles is one of the fastest growing areas of nanotechnology. A special place is occupied by studies of silver nanoparticles (NPs) of various shapes and sizes, which is associated with their special antibacterial and optical characteristics. The widespread use of materials with the inclusion of silver nanoparticles inevitably leads to the ingress of the latter into the environment.

One of the most common methods of obtaining silver nanoparticles is the reduction of Ag^+ silver ions in aqueous solutions with borohydride, hydrazine and other reducing agents that are toxic. To prevent the coagulation of nanoparticles, various stabilizing additives are introduced. However, these additives form a dense (commensurate with the nanoparticles themselves) "fur coat" around the nanoparticle, which leads to a decrease in the bioavailability of nanoparticles. Accordingly, attention is drawn to the creation of a method for obtaining silver hydrosol, which would include a compound contained in natural waters – carbonate ions. Such a hydrosol can be used as a model for studying the environmental impact of silver nanoparticles directly, without taking into account other impurities. An advantage would also be the possibility of spontaneous formation of silver nanoparticles under conditions similar to natural ones, for example, due to the action of light.

An urgent task is to assess the impact of silver nanoparticles on the microflora in the environment, not complicated by the influence of foreign impurities (decomposition products of reducing agents, stabilizing additives, etc.). Therefore, an important task is to develop a method for the synthesis of silver nanoparticles in which the hydrosol would not contain toxic impurities. It is also desirable that the hydrosol in its composition, if possible, contain components characteristic of natural fresh water, in particular, carbonate anions. Such a method will ensure the minimization of the anthropogenic impact on wildlife and comply with the principles of green chemistry: during synthesis, do not use toxic compounds and do not produce them as waste. The developed method of obtaining silver hydrosol by photochemical reduction of metal ions in the presence of oxalate ions meets these requirements.

The purpose of the work is to develop a method for the synthesis of silver hydrosol in accordance with the principles of green chemistry, as well as to study the transformation of the resulting hydrosol in various types of waters and its effects on the microflora.

To achieve the purpose, the following **tasks** were set:

1. Determination of the parameters and conditions of the silver ion reduction reaction at room temperature and pressure (T = 295 K and P = 98-102 kPa) in the presence of oxalate ions under the influence of UV radiation both in the presence of oxygen and in its absence, as well as the study of the mechanism of hydrosol formation.

2. Development of a method for spectrophotometric determination of the concentration of silver atoms in hydrosol nanoparticles.

3. Study of oxidative dissolution and aggregative stability of carbonate-stabilized silver nanoparticles in drinking water and natural waters of various composition and origin to assess the possible impact of silver nanoparticles on the environment.

4. Investigation of the effect of carbonate-stabilized silver nanoparticles on representatives of Gram−negative bacteria (G-) Escherichia coli and Pseudomonas putida, as well as gram-positive bacteria (G+) Paenibacillus jamilae.

5. Comparative analysis of the biocidal effect of silver ions and carbonate-stabilized silver nanoparticles of various sizes to establish the limits of tolerance of microorganisms.

Scientific novelty:

1. A method has been developed for the synthesis of silver nanoparticles by reducing Ag^+ ions with carboxyl anion radicals $CO_2^{-\bullet}$ formed from oxalate ions $C_2O_4^{2-}$ under the influence of UV radiation. At the same time, the method complies with the principles of green chemistry, the hydrosol does not contain toxic impurities. The use of this hydrosol reduces the negative impact on the environment.

2. In the absence of oxygen in the air, silver nanoparticles with an average size of 10 nm are formed. Based on the shift of the maximum of the LPPR band according to the Mi-Drude theory, it was found that during the formation of a hydrosol, an increase in the electron density occurs by ~ 10% on the surface of nanoparticles, which probably can increase the antibacterial effect. During the formation of the hydrosol, there is no increase in the size of nanoparticles, which distinguishes this method from the traditional method of chemical reduction of Ag^+. In the presence of oxygen in the air, the average size of nanoparticles is ~20 nm. In both cases, the thickness of the stabilizing layer is ~ 2 nm.

3. The optical absorption of silver hydrosols at $\lambda = 250$ nm refers to the interband electronic transitions in the metal. Absorption in this area, unlike LPPR, is not sensitive to the state of the surface of nanoparticles. The molar extinction coefficient of silver atoms in spherical nanoparticles, calculated according to the Booger–Lambert–Behr law, turned out to be $\varepsilon_{250} = 3500 \pm 100$ L mol^{-1} cm^{-1}. On this basis, a simple method for determining the concentration of silver atoms in hydrosol nanoparticles is proposed and justified.

4. In the absence of air, the silver hydrosol remains stable for several months. The presence of oxygen initiates the oxidation of the metal with the release of Ag^+ ions into the solution, and the aggregation of nanoparticles occurs at the final stage. The rate constant of oxidative dissolution of carbonate-stabilized silver nanoparticles and ion release is calculated to be $(1.6\pm0.2)\times10^{-3}$ min^{-1} in the studied concentration range $(1\text{-}4)\times10^{-4}$ mol L^{-1} Ag^0. The mechanism of oxidative dissolution of silver nanoparticles has an electrochemical nature.

5. The hydrosol of carbonate-stabilized nanoparticles is unstable in contact with natural waters. At the same time, nanoparticles show a pronounced tendency to agglomeration and aggregation, culminating in the release of metal into the precipitate. The reason is the presence of ions in the waters, primarily such as Cl^-, SO_4^{2-}, Ca^{2+}, Mg^{2+} and Na^+. Such waters have a high ionic force, which is the main reason for the compression of the double electric layer, as a result of which the stabilizing Coulomb repulsive forces acting between the particles are weakened.

6. Carbonate-stabilized silver nanoparticles have a depressing effect on gram-negative cells of the bacterium Escherichia coli and Pseudomonas putida and gram-positive Paenibacillus jamilae. Nanoparticles inhibit bacterial cell growth at concentrations of $\sim1\times10^{-6} - 1\times10^{-4}$ mol L^{-1}. In silver hydrosols obtained by photochemical reduction, when they are used, only silver directly affects bacterial cells, since there are no toxic reducing agents, stabilizers or their decay products.

7. The complex mechanism of antibacterial activity of silver nanoparticles has been confirmed and substantiated, including the indirect action of silver ions and the contact action of

the nanoparticles themselves, causing the formation of reactive oxygen species as a result of their oxidative dissolution.

The practical significance of the work lies in the development of a method for the synthesis of silver hydrosol containing silver nanoparticles and carbonate anions that are part of natural water. This circumstance makes it possible to exclude the influence of toxic reducing agents and stabilizers when evaluating the antibacterial properties of silver. Such hydrosol can be recommended as a model for studying the effects of silver in the form of nanoparticles on microorganisms. The developed method for determining the concentration of silver atoms in hydrosol nanoparticles using spectrophotometric analysis is simple and inexpensive, while additional expensive equipment and sample preparation measures are not required.

Provisions submitted for thesis presentation:

1. Method of synthesis of silver hydrosol in the presence of oxalate ions under the influence of UV radiation (in the presence and absence of oxygen in the air).

2. The mechanism of hydrosol formation and the change of its electronic and optical properties in the process of photochemical synthesis.

3. The method of spectrophotometric determination of the concentration of silver atoms in hydrosol nanoparticles.

4. Kinetics of oxidative dissolution of carbonate-stabilized silver nanoparticles in water and formation of silver ions.

5. Stability of carbonate-stabilized silver nanoparticles in natural waters.

6. Biocidal effect of carbonate-stabilized silver nanoparticles in suppressing the vital activity of cells of Gram-negative bacteria *Escherichia coli* and *Pseudomonas putida* and cells of gram-positive bacteria *Paenibacillus jamilae*.

7. Indirect (silver ions) and direct (nanoscale silver particles) mechanism of antibacterial action of silver nanoparticles