INFLUENCE OF SILVER PREPARATIONS ON LIVING ORGANISMS

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The fight against resistant microorganisms has recently become increasingly important. The majority of antimicrobial drugs, including antiseptics, have successfully developed multidrug resistance in many microorganisms and viruses. Among the drugs to which the microorganisms have not yet developed resistance remain silver preparations. In this review, we offer a summary of research on the silver preparations effect not only in microorganisms, but also in the human body, including the molecular-biological aspects of such effect.

Action of silver preparation on microorganisms

The main type of silver used in the clinic is its antimicrobial effect relation to bacteria, fungi, viruses and protozoa (about 700 species of microorganisms), including strains that cause diseases in humans.

The antimicrobial activity of silver preparations is determined by the presence of its ionic form — Ag + ions, since they interact with the cellular membranes of the microorganism. The antimicrobial effect is proportional to the degree of ions release from metallic silver preparations. Biological activity of silver is increases through its ionization in aqueous solutions. The microorganisms growth-decreasing rate is depends on the concentration of silver ions in the solution. Thus, E. coli dies after 3 min at a concentration of 1 mg /L, after 20 min - at 0.5 mg / L, after 50 min - at 0.2 mg / L, after 2 h - at 0.05 mg / L [1].

The sensitivity of bacteria and fungi to silver is genetically determined and is determined by the degree of its capture by the cells and the ability to interact with key enzymes (to denature them) [22]. Silver ions react with the amino acid residues of the proteins from bacterial cell membranes, fungi and protozoa, that resulted to denaturation and inactivation of proteins, vital enzymes, RNA-ase and DNA-ase [11].

There are many different theories explaining the mechanism of silver action. They reflect its multifaceted effect on the microorganisms cells.

According adsorption theory, a cell loses viability as a result of electrostatic forces arising between negative charge bacteria cells and positively charged silver ions when the latter are adsorbed by a bacterial cell. Upon contact with the pathogen, silver attaches an electron, which causes rapid disruption in the membrane. Such universal mechanism action determines a wide range of antimicrobial activity (bacteria, viruses, fungi, protozoa) and eliminates the antimicrobial resistance development. The antimicrobial effect of silver was also attributed to the inhibition of enzymes containing SH- and COOH-groups, or a violation of the osmotic equilibrium in the bacterial cell. It is also assumed that inhibition of the transmembrane transport of Na + and Ca ++, caused by silver is one of the reasons for the broad spectrum of the antimicrobial action of silver ions. There is also an opinion that silver does not directly affect the cell's DNA, but acts indirectly by increasing the number of intracellular free radicals, which reduce the concentration of active oxygen compounds. Some researchers attach particular importance to physical-chemical processes. In particular, the oxidation of bacterial protoplasm and its destruction by oxygen dissolved in water, while silver plays the role of a catalyst [2].

Silver reacts with the bacterial cell membrane, combines with external peptidoglycans, blocking their ability to transfer oxygen into the bacterium cells, which leads to hypoxia in the microorganism and resulted to its death. At the same time, the specificity of silver action with respect to microbial cells and the resistance of mammalian cells to it are manifested. The bacterial cell wall, unlike mammals, consists predominantly of peptidoglycans with fewer teichoic acids and a small amount of polysaccharides, proteins, and lipids. Since mammalian cells have a completely different type of membrane (not containing peptidoglycans), silver has virtually no effect on them. Extracellular viruses, like organisms without a stable cell wall, are also affected by silver. There is evidence of the formation of nucleic acids complexes with heavy metals (including silver), as a result of which the DNA stability and, accordingly, the viability of bacteria are disturbed. Currently, the mechanism of silver action on a microbial cell is as follows. Silver ions are sorbed by the cell membrane and violate its protective function. The cell remains viable, but its functions, such as division (bacteriostatic effect), are disturbed. Further, silver penetrates into the cell and inhibits the enzymes of the respiratory chain, and also separates the processes of oxidation and oxidative phosphorylation in microbial cells, causing the cell to die [1].

Another metabolic effect of silver ions on the bacterial cell is inhibition of phosphate uptake and efflux of accumulated phosphate, mannitol, glutamine and proline, which was shown in an in vitro study on *Escherichia coli*. These silver ion effects were inhibited by thiols and, to a lesser extent, by bromide. In the presence of N-ethylenediamine and a number of respiratory phosphorylation uncouplers, Ag + ions lost their ability to cause efflux of phosphates, but nevertheless inhibited the exchange of intra- and extracellular phosphates. Therefore, silver inhibits the efflux of metabolites, acting as an uncoupler, as an inhibitor of the respiratory chain or as a thiol reagent [3]. The fact that silver ions affect the enzymes of the microorganism's respiratory chain was shown in another experiment on *E. coli* [4]. It is noted that silver violates the function of oxygen-metabolizing enzymes of various pathogens - bacteria, fungi, viruses. As a result, microorganisms die and are removed from the body by immunocompetent cells of the lymphatic system [5].

Electron-microscopic evidence of the antibacterial action of silver nitrate is presented. The study was conducted using a representative of gram-negative bacteria - *Escherichia coli*, and gram-positive bacteria - *Staphylococcus aureus*. Used electron microscopy and radiation microanalysis. Under the action of silver ions in

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both bacteria species, similar morphological changes were revealed - separation of the cytoplasmic membrane from the cell wall. The damage was especially large in the central part of the cells, which contained DNA molecules. Granules are also identified that surround the cell wall or are located inside the cell. Silver ions were detected in these granules. The main mechanism of the silver antibacterial action, the authors considered the loss of DNA's ability to replicate and proteins inactivate. The morphological changes of *E. coli* were more significant than *S. aureus* [6].

In one of the last (2015) studies, a previously unknown mechanism of the prolonged antimicrobial action of silver nitrate was shown, which consists in the bactericidal properties of microorganisms previously killed by exposure to silver ("zombie effect"). This has been shown in the culture of *Pseudomonas aeruginosa*, which was subjected to the bactericidal action of silver nitrate. This culture also had a bactericidal effect on the living culture of this bacterial strain [7].

The references describes the features of the colloidal silver mechanism action. Colloidal silver particles have a certain size (about 10 nm), pH, electronic configuration, give a certain magnetic resonance and electromagnetic frequencies, specifically interact with water molecules. Therefore, the mechanism of colloidal silver action includes various types of effects, including electrical, magnetic, electromagnetic, chemical and resonant. Silver can exhibit bipolar and semiconductor properties. It has a resonant frequency at 910 Tera-Hz, which is in the ultraviolet region. It also contributes to its bactericidal activity [8].

Ultrastructural dynamics studies have shown that colloidal silver nanoparticles within 15 minutes attached to the outer cell membrane of *Escherichia coli* (ATCC 43888-O157: k-: H7) and *Vibrio cholerae* (O1). At the same time, the morphology of cell membranes was disturbed, their permeability increased. Then, after 30 minutes, the smallest particles penetrated into the cells. At the same time, a violation of the cytoplasm structure was noted. After penetration into the cells, silver particles interacted with DNA, disrupting the cell's ability to divide. After 60 minutes influencing of silver cells on the cholera vibrio, cells were completely destroyed (Fig. 1) [9].

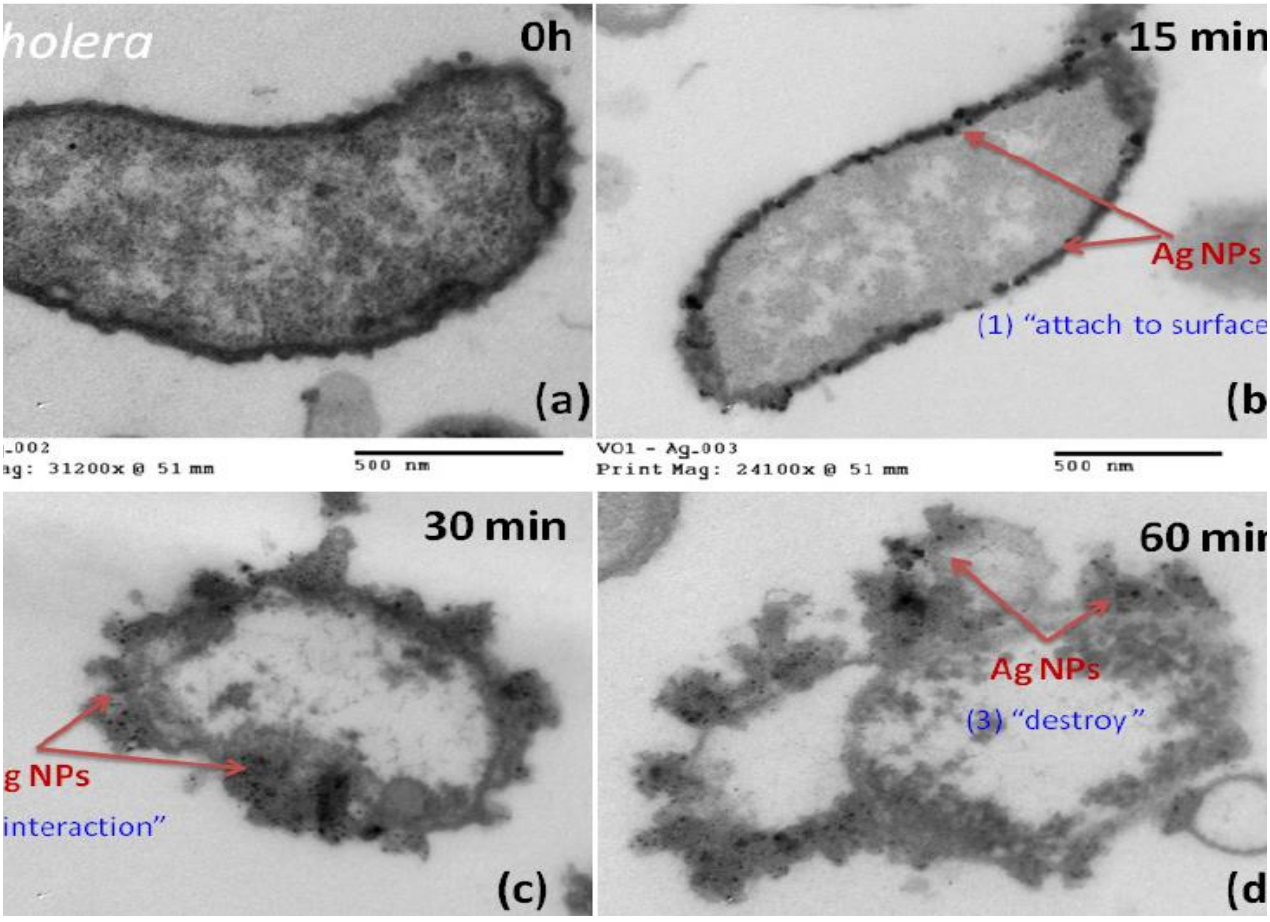

Fig. 1. Stages of the silver particles interaction with *V. cholerae*

The silver is shown a highest activity against bacteria. Bacteriostatic (at lower concentrations) and bactericidal (in large concentrations) action is manifested to the majority Gr+ and Gr-bacteria, including causing acute respiratory infection in children and adults: *Streptococcus pneumoniae, Staphylococcus aureus, Moraxella catarrhalis,* etc. Experimental studies indicate that, with a concentration of 1 μg/ml, silver ions can control most bacterial and fungal pathogens [11].

Silver ions can influence on the growth, morphology and ultrastructure of *Staphylococcus aureus* and *Escherichia coli*. Exposure of bacteria with silver ions was carried out during various periods of time. Antibacterial action was estimated by counting CFU in an agar plates and next using the flow cytometry analysis. It

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was found that CFU for some bacteria strains was reduced by more than 5 lg/mL after 90-minute silver exposure. Cytometric analysis showed a less pronounced, but also significant, decrease an *Staphylococcus aureus* and *Escherichia coli* cells number. The authors thinks, that difference in the indicators for these two methods linked with living, but nonculturable state of bacteria's after influence of silver [10].

Colloidal silver manifested bactericidal effect in relation to 5 isolates *E. coli* and 5 isolates *S. aureus*, isolated in patients with surgical wounds. The bactericidal action constant for the drug concentration of 5 μg/mL was for different strains *E. coli* 0,17-0,68 min^{-1}, for *S. aureus*-0,007-0,010 min^{-1}. At a concentration of 20 μg/mL-respectively 0,003-0,014 and 0,0001-0,0004 min^{-1}. Bactericidal action of silver had no direct dependence on concentration. High antimicrobial activity of colloidal silver determines the expediency of its use for the treatment of aseptic wounds and burns [11].

Its antimicrobial activity against standard strains of microorganisms and multiresistant clinical isolates was assessed. MIC were in the interval 0,78-6,25 μg/mL, IBC-12.5 μg/mL. Bactericidal action against Gram-negative bacteria was more pronounced-a decrease of 3 lg within 5-9 hours, for Gram+ bacteria the same effect was reached within 12 hours [12].

The colloidal silver is shown a beneficial bactericidal action in relation to bacterial gastrointestinal infections pathogens: Escherichia coli (ATCC 43888-O157: K-: H7) and Vibrio cholerae (O1). The colloidal silver has expressed bactericidal effect at low concentrations-3 mg/L [37].

Colloidal silver effectively eliminates *P. aeruginosa*, one of the major pathogens that cause postoperative, burn and pulmonary infection, being one of the leading causes of death. As you know, *P. aeruginosa* is able to form films, which makes it resistant to the antibiotics action. In some in vitro studies have shown the efficacy of colloidal silver in relation to this microorganism. Thus, the colloidal silver shown a bactericidal effect in the concentration of 15 μg/mL after exposition beginning at 4 minutes. In a concentration more than 30 μg/mL, the effect is even faster [1].

P. aeruginosa strains were suppressed by colloidal silver. These strains separated from infected surgical wounds in eye surgery. Methods of sowing on agar and in liquid environment were used, the kinetics of bactericidal action were estimated. It was found that colloidal silver has bactericidal activity to *P. aeruginosa*. Zones of growth delay for 5 strains of *P. aeruginosa* are 27-33 mm, MIC of silver for 3 strains are 0.078 μg/mL, for 2 strains-0.313 μg/mL. Bactericidal effect linearly louder over time and has maximum in exposition at 90 min. The constant of bactericidal action was 0.084 min^{-1} [13].

Silver nanoparticles prevent the formation of films by bacteria. Silver's nanoparticles reduced ability of *Pseudomonas aeruginosa* and *Staphylococcus epidermidis* to form films on the cultures of with 24-hour contact more than 95% [14].

Various in vitro studies show the efficacy of silver in relation to resistant bacterial strains.

Colloidal silver (solution ACS 200) eliminated (100% effect) *Borrelia burdorferi* for 8 minutes, *Salmonella bongori* – for 3 min., *Candida albicans* – for 2 minutes, MRSA – for 3 min., *S. aureus* – for 15 sec., *Pseudomonas aeruginosa* – 3 min., *Legionella Pneumophila* – for 2 min. , *Rhinovirus*-for 15 minutes, *Powassan virus*-for 60 minutes. [8].

Another study also showed that the colloidal solution of silver in the form of nanoparticles is effective in relation to *P. aeruginosa* and MRSA. This solution was applied on the surface, were not colonized by these microorganisms unlike the control [1].

Silver has a long-lasting effect. At the end of the impact, his overwhelming action persisted for *P. aeruginosa* for 10.5 hours, *Staphylococcus* SRP. – 1.3 H., *Candida albicans* – 1.6 hour [12]. The effect of colloidal silver in relation to *Escherichia coli* and *Vibrio cholerae* lasted much longer than that of 5% chloramine in, and intensified with time [9].

In comparative studies it was found that silver significantly surpasses the efficiency of most known antiseptics. So, the high antiseptical ability of silver was higher, than at carbolic acid, sulema and even such strong oxidants, as chlorine, chlorine lime, hypochlorite sodium [1].

Antimicrobial action of colloidal silver in relation to *S. aureus*, *E. coli*, *P. aeruginosa*, *Salmonella typhi* studied in comparison with antibiotics of various groups: Chloramphenicol, Tobramycin, Cefaklor, Cefadroksil, Ko-Trimoxazole, erythromycin, Ciprofloxacin, levofloxacin, Norfloksacin. It was found that at least 2 of the studied strains showed resistance to each of these antibiotics. The most resistant were *E. coli* (moderate sensitivity only to Chloramphenicol) and *Salmonella typhi* (moderate sensitivity to tobramycin). *S. aureus* was moderately sensitive to Chloramphenicol and co-Trimoksazol. *P. aeruginosa* was sensitive to Tobramicin, ciprofloxacin, Levofloxacin and Norfloxacin. Only to silver were sensitive all microorganisms [15].

Colloidal silver was more effective than some antibiotics against *S. aureus* and multiresistant strains of *P. aeruginosa* [1].

Colloidal silver is also active for inactivation anaerobic bacteria of the genus Clostridium (13 strains), but inferior to such strong antiseptics as Katapol, Katamin and chlorhexidine. MIC of Katapol and Katamin made 2.25-5.0 μg/mL, chlorhexidine and Dioksidina-20-40 μg/mL, Protargol-1250 μg/mL In relation of Proteus genus (4 strains) – respectively 5-10, 40-80 and 1250 μg/mL; In relation to the Enterobacteriae family and nonfermented $Gram^-$ bacteria (5 strains)-5-10, 0.31 and 312-625 μg/mL; In relation to Gram+ cocci (5 strains) – 1.25, 1.25 and 160 μg/mL [16].

We are present comparative data of MIC and MBC for Silver and a number of antibiotics (table) from different references [17]. This data shows that if certain antibiotics are more effective for certain microorganisms, the generally nonspecific antimicrobial action is higher in silver.

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Table. Minimum inhibiting concentration (MIC) and minimum bactericidal concentration (MBC) of silver and antibiotics against bacterial pathogens

Microorganism	MIC / MBC, μg/mL					
	Tetracycline	**Ofloxacin**	**penicillin**	**Cefaperazon**	**Erythromycin**	**Silver**
S. pyogenes	0.625/>5	1.25/2.5	>5.0	0.313/1.25	0.003/0.019	2.5/5.0
S. mutans	0.625/>5	2.5/>5.0	0.521/>5	1.25/>5	0.009/0.019	2.5/10.0
S gordonii	0.156/0.625	2.5/5.0	0.009/0.039	1.25/1.25	0.005/0.019	2:5/10.0
S. pnuemoniae	0.078/0.625	2.5/2.5	0.019/0.019	0.313/0.313	0.002/0.004	2.5/2.5
S. faecalis	0.313/>5	1.25/5.0	5.0/>5.0	>5.0	0.009/1.25	10.0/10.0
S. aureus	0.313/>5	0.417/0.625	2.5/>5.0	5.0/5.0	0.039/>5.0	5.0/5.0
P. aeruginosa	0.078/5	0.156/0.313	0.13/>5.0	2.5/5.0	2.5/>5.0	1.67/5
E. coli	1.67/>5	0.104/0.156	>5.0	0.625/>5.0	5.0/>5.0	2.5/2.5
E. aerogenes	>5	0.078/0.156	>5.0	2.92/>5.0	>5.0	2.5/2.5
E. cloacae	1.67/>5	0.156/0.156	>5.0	>5.0	>5.0	2.5/5.0
S. tiphimurium	1.25/>5	0.078/0.156	>5.0	1.25/2.5	5.0/>5.0	2.5/5.0
S arizona	0.625/>5	0.078/0.078	>5.0	0.833/>5.0	4.17/>5.0	2.5/5.0
S. boydii	1.25/>5	0.078/0.156	>5.0	0.625/0.625	5.0/>5.0	1.25/1.25
K. pneumoniae	2.5/>5	0.417/0.625	>5.0	>5.0	>5.0	2.5/2.5
K. oxytoca	1.25/>5	10.104/0.156	>5.0	1.25/>5.0	>5.0	1.25/1.25

An important aspect of Silver's action is the fact that microorganisms do not develop resistance to it [1]. There is evidence that the sensitivity to silver of pathogenic and nonpathogenic bacteria is different. Pathogenic microorganisms are more sensitive than the normoflora of the human body. On the basis of it the method of dysbiosis treatment with application of a cavity electrophoresis by silver ionic solution in concentration 500 μg/L was developed.

Silver also has good antifungal activity. Thus, at a concentration 75 μg/mL was noted 50% inhibition *Aspergillus niger*, with antifungal index 55.5%, against *Candida albicans* MIC was 25 μg/mL [12].

According to other data, silver has a pronounced fungicide effect already at a concentration 0.1 μg/mL. With microbial load of 100 000 cells per liter, the death of Candida fungi came after 30 minutes contact with silver. The effect of silver water at the same concentrations was above the action of chlorine, chlorine lime, sodium hypochloride and other strong oxidants [1].

The high antifungal activity of silver nanoparticles to 44 mushrooms strains and 6 different clinical isolates and standard strains *Trichophyton mentagrophytes* and *Candida* SPR is Revealed. The IK_{80} values were in the range of 1-7 μg/mL. The activity level of silver preparations were at the amphotericin-B level exceeded fluconazole, IK_{80} which made respectively 1-5 and 10-30 μg/mL. moreover, silver nanoparticles influenced only the mycellar form of Candida albicans [18].

The comparison of antifungal activity of Protargol, Miramistina and chlorhexidine against the MDR strain of *Candida tropicalis*, allocated in children with adenoiditis was carried out. Identified antiseptic's MIC in relation to fungus blastospore at concentration of $5 \cdot 10^7$ a CFU/ml. The values of MIC amounted to: Protargol – 0.1%; miramistin -0.005%; chlorhexidine-0.005% [19,20].

Another similar study also established protargol activity in relation to the MDR strain of *Candida tropicalis*. Antifungal action protargola manifested at concentrations of 0.1-2%, miramistin-0.01-0.05% and chlorhexidin-0005-0.05% [21].

The ability of silver nanoparticles to eliminate forming films fungal pathogens is established. Thus, in the study on films forming fungi strains, causing stomatitis- *C. albicans* and *C. glabrata*, Silver nanoparticles showed a fungicide effect already in low concentrations-0,43-3.3 μg/mL Silver nanoparticles eliminated *C. glabrata* in the biofilms form even more effectively (within 2 hours of Impact) than not forming films mushrooms (within 48 Hours) [22].

Antiviral action of silver

In addition, silver can actively eliminate viral Infection. A number studies have found that silver ions have a pronounced ability to inactivate poxviruses, influenza strains A-1 and B, some Entero- and adenoviruses, as well as inhibit HIV. However silver's virulicide action is manifested at higher concentrations, than bactericidal. So, iIt is shown, that for full inactivation of bacteriophage N 163 *E. coli* and a Koksaki virus serotypes A-5, A-7, A-14 requires a higher silver concentration (500 – 5000 μg/L) than for Escherichia, Ealmonella, Shigella and other intestinal bacteria (100 – 200 μg/L.) [1].

Because of protargol solution research in various concentrations on a cells culture was shown reduction of

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viruses reproductions for viruses causing infectious rhinotracheitis and viral diarrhea at concentration 0,25-0,5% [23].

Two studies show the Protargol and Kollargol action against the smallpox Virus. In 0.05% Protargol and Kollargol added suspension of the virus pox to the final concentration of 10^5 BFU/ml. After exposure 60 minutes were selected samples for quantitative evaluation results, for which titrated on the chorion-allantois shell 11-13-day chicken embryos. As a result, a decrease virus concentration under the action of kollargol from $68*10^5$ to 60 BFU/ml, protargol -to 10^3 BFU/ml, i.e. more than 2-4 of the order [5] was revealed. In another similar study, at a concentration of the virus $6.8 \cdot 10^5$ BFU/ml, the number of microorganisms after exposure to protargol and kollargol was $1 \cdot 10^3$ and 60 BFU/ml respectively [4].

The influence of silver nanoparticles on the hepatitis B virus has been established. Human hepad38 lines liver cells that replicate hepatitis B virus, were incubated with 10 and 50 nm particles in a buffer solutions. In this case, the reduction more than 50% hepatitis B viral RNA formation by cells in comparison with the control cells, not contacts with silver. Silver has weakly influenced the viral covalently sewn ring DNA, but inhibited intracellular RNA virus formation. Particles 10 nm had good adhesive properties in relation to the viral DNA with a binding constant of $8.8 \pm 1.0 \cdot 10^5$ dm^3/mole. Particles this diameter have dimensions similar to the viruses size and can interact directly with them, which has been confirmed in electronic microscope. Thus, the silver nanoparticles are suppress the synthesis of hepatitis B viral RNA and extracellular virions formation [24].

The effect of silver 10 nm nanoparticles on HIV-1 was shown. In experiments with application of dark field method in the passing light at scanning electron microscopy has shown, that silver particles f are attached to a virus, and this connection occurs by binding with glycoprotein-120, located on a virion shell and linking to macroorganisms cells [8].

Anti-inflammatory effect of silver

Unlike other antibacterial chemotherapy drugs, protargol has not only antimicrobial, but also anti-inflammatory and immunostimulating properties. It also has some astringent effect. Being a silver containing protein compound, protargol forms a protective film from the silver and proteins molecules on the infectious process's damaged mucous membrane, which in turn leads to a decrease its sensitivity to infectious agents. Protargol has vasoconstriction effect- reduces the capillaries lumen, thereby inflammatory processes inhibiting. Vasoconstriction action is mild, which does not cause complications from the blood vessels and allows its long-term use [25].

The direct anti-inflammatory action of silver has been revealed, which is confirmed by a number of experimental studies on colitis models, inflammation of allergic nature (contact dermatitis), in experiments in vitro [26]. It also showed a decrease in nasal symptoms of allergic rhinitis in mice, inhibition of products IgE, il-4 and il-10, suppression of infiltration of inflammatory cells and hyperplasia of gobleta mucous membranes of respiratory tract [27]. Anti-inflammatory action of silver manifested in the dose dependent inhibition of marker enzymes-metalloproteinases 2 and 9 [12]. The anti-inflammatory effect of silver is also achieved by modulation of the cytokine profile. These effects, as well as the stimulation of fibrinogenic cytokines, play an important role in the silver wound healing activity.

Silver nanoparticles have anti-inflammatory effect to nasal polyps, suppress activation of epithelial cells of nasal polyps in the person in the experiments in vitro. At a concentration of silver 10 μg/mL the polyps cells survivability has considerably decreased, its inhibitory effect on production interleukins il-6 and il-8, granulocytic – macrophage and colonial-forming factor, nuclear factor NF-kB and activator protein AR-1 is established. At a lower concentration of silver- 1 μg/mL, decreased cytokines production and factor NF-kB level [28].

In recent years, there has been evidence that silver is a powerful immunomodulator, comparable to steroid hormones. In various studies it is shown that nanoparticles of silver inhibit the production of cytokines by macrophages. Particles of small sizes (1.5 Nm) inhibited secretion interleukin IL-5, γ-interferon and TNF-α, and particles in diameter 100 nm- IL-6, IL-8 and IL-11. It is believed that the interaction of silver nanoparticles (diameter 0.1-100 Nm) with the human immune system begins with binding to the receptors PRRs, located on the surface of immunocompetent cells.

Under the influence of silver increases the number of immunoglobulin classes A, M, G, increases the percentage of the absolute number of T-lymphocytes. It is established that, depending on the dose, silver can both stimulate and suppress phagocytosis [2]. Silver nanoparticles have a cytoprotective effect on the HIV-infected T-cells.

In recent years, the antitumor potential of colloidal silver has also been established. His cytotoxic action is shown in the experiments in vitro on various human tumors cells and in the experiments in vivo on the animals tumors models -increase the duration of mice survival with lymphoma Dalton [29].

Anticancer activity of silver

In the study on tumor cells MCF-7 (human mammary glands), the impact of 0.75-17.5 ng/ml colloidal silver was spent for 5 hours at 37^0C in the CO_2. Vitality of cells was estimated by coloring trypan blue, The mechanism of cell death was determined by the presence of mono- and olygo - nucleosomes with the help of tests ELISA and TUNEL, colorimetric method determined the production NO, activity lactatdehydrohenase, peroxidase, dismutase, catalase and general antioxidant activity. It was found that colloidal silver rendered dose cytotoxic effect by induction of apoptosis. It has LD_{50} 3.5 ng/ml, LD_{100} 14 ng/ml, marked a significant decrease in lactatdehydrohenase activity and increase superoxyddismutase. No impact on peripheral mononuclear cells. The authors of the study have concluded that the use of silver in cancer therapy is possible [30].

In vitro experiments the cytotoxic action of silver 7-12 nm nanoparticles to human tumor cells-HT-1080

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fibrosarcoma and A431 epidermal Carcinomas was evaluated. After exposure to silver nanoparticles in the concentration less than 6.25 μg/mL cells did not change their morphology. At increase concentration to 6,25-50 μg/mL cancer cells of both tumors types became less polyedrical, more wrinkled. Semi-inhibiting concentrations (IK_{50}) for tumors HT-1080 and A431 amounted respectively 10.6 and 11.6 μg/mL. At processing of cages by silver particles in concentrations constituting ½ from IK_{50} (6.25 μg/mL), showed signs of oxidative stress – increase level of the restored glutathione, dismutase activity, peroxide oxidation strengthening. Also DNA fragmentation occurred, which indicates about presence of apoptosis. During the analysis of activity indicators caspase-3 it is established, that for occurrence apoptosis of cells HT-1080 and A431 are enough silver concentrations respectively 0.78 and 1.56 μg/mL, and cells necrosis of both tumor types occurred at a concentration of 12.5 μg/mL [31].

In a number of studies it is shown that cells of healthy human tissues are less susceptible to cytitoxic action of silver nanoparticles and are significantly better restored than tumor cells. Thus, in vitro research the influence of silver nanoparticles on human fibroblasts (IMR-90) and glioblastoma cells (u251) was studied. The silver toxicity in relation to these cells was evaluated on the basis of data on their morphology, viability, metabolic activity and oxidative processes. The cultivation of cells with silver caused a doses-dependent decrease in the ATP content, damage to mitochondria and increased production of reactive oxygen. In tumor cells more than in the fibroblasts, there was DNA damage, the violation cell cycle in the phase G (2)/M by coloring Annexin-V-propidium iodide showed that there was no massive apoptosis or necrosis. With the transmission electron microscopy the presence of silver in mitochondria and a nucleus is revealed, that has determined its participation in damage of DNA and mitochondria. Perhaps there was a disintegration of the mitochondrial respiratory circuit, the reactive oxygen appearance and a violation ATP synthesis, which caused DNA damage [32].

In the this publication it is indicated that capture of silver particles by fibroblastama and cages of a glioblastoma happened by an endocytosis - klatrin-dependent process and a makropynocytosis that was followed by increase eventually and an exocytosis. Electronic microphotos show intracellular distribution of silver in cytoplasm and nucleus. At the same time the instability of chromosomes and violation of a mitosis is noted. However, in normal cages of fibroblast, there is an effective restoration of a mitosis, and in tumor cells proliferation wasn't restored. Particles of silver had the effect by means of the intracellular calcium transition and chromosomal aberrations. Toxic action is mediated by intracellular transport of ions of Ca2 + and considerable changes in morphology of cages. The aktin-connecting protein, filamin was suppressed, there was a stress leading to violation of genes regulation activity for metalloproteinase and gemoxydase-1. Thus, tumor cells were more subject to damage by silver, in the absence of restoration [33].

Some molecular effects of silver

Silver, when applied systemically (or when it enters the systemic circulation when applied locally), has important effects, since it is a trace element essential for the normal functioning of the endocrine glands, brain and liver. The high biological activity of trace elements-metals in the body is associated primarily with their participation in the synthesis of certain enzymes, vitamins and hormones. Silver ions take part in the body's metabolic processes. Depending on the concentration, its cations can both stimulate and inhibit the activity of a number of enzymes [2].

Under the influence of silver, the oxidative phosphorylation intensity in brain mitochondria is enhanced, and the content of nucleic acids also increases, which improves brain function. When various tissues are incubated in the presence of 0.001 μg of silver, the absorption of oxygen by the brain tissue increases by 24%, in the myocardium by 20%, in the liver by 36%, in the kidneys by 25%. An increase silver ions concentration to 0.01 μg reduced the degree of oxygen absorption by the cells of these organs, which indicates the silver participation in the regulation of energy metabolism [1].

When studying the effect of silver preparations on the human body, its stimulating effect on the blood-forming organs was observed, manifested in the disappearance of young forms of neutrophils, an increase in the number of lymphocytes and monocytes, erythrocytes and hemoglobin, and inhibition of ESR [1].

Applications of silver and conclusions

Currently, the main areas of application of silver preparations are: water disinfection, wound care, prevention of infection, oral hygiene (prevention and control of gingivitis, paradontosis), eye hygiene, including in newborns, treatment of nasal infections [34]. It has been established that silver solutions are effective in direct contact with surfaces that fester and become inflamed due to bacterial contamination. Silver is effective in treating wounds, burns, bone prostheses, used in periodontitis, in reconstructive orthopedic surgery. New studies have shown its effectiveness of silver in AIDS, pneumonia, herpes, shingles [8]. The effects of silver in disorders of the central nervous system, the digestive system, as a tonic in the elderly, in the treatment of arthritis, hemorrhoids, allergies, psoriasis and dandruff are not sufficiently proven [34]. Silver preparations are treated with wound dressings, dental materials, bone cement, catheters, hygiene items and other products [35]. Protargol is used in the treatment of complicated rhinitis of various etiologies - with prolonged purulent rhinitis, sinusitis, etimoiditis and frontal sinusitis, used for the treatment of recurrent and prolonged nasopharyngitis, as well as for the treatment of adenoiditis. Due to the high safety Protargol is widely used in pediatric practice.

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The fight against resistant microorganisms has recently become increasingly important. The majority of

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antimicrobial drugs, including antiseptics, have successfully developed multidrug resistance in many microorganisms and viruses. Among the drugs to which the microorganisms have not yet developed resistance remain silver preparations. In this review, we offer a summary of research on the silver preparations effect not only in microorganisms, but also in the human body, including the molecular-biological aspects of such effect. The article presents results of studies silver ionic forms, colloidal and metallic form influence on microorganisms, viruses and cancer cells. The effect of various silver preparations on the biochemical pathways in the human body is also shown.

Keywords: silver, microorganisms, viruses, immunity, cancer, biochemistry

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