

THE INFLUENCE OF SILVER NANOPARTICLES ON DIFFERENT STAGES OF REPRODUCTION OF INFLUENZA A VIRUS (H1N1)

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Since the 19th century, the bactericidal properties of silver have been widely used in medicine, and in recent years the nanotechnology industry increasingly pays attention to them. Studies of the effect of silver nanoparticles (AgNPs) on various microorganisms, including viruses, are constantly being conducted. After all, the problem of the lack of antiviral drugs remains urgent, especially for RNA-containing viruses, which very quickly acquire resistance. Therefore, the aim of this work was to study the effect of AgNPs mixtures on different stages of reproduction of influenza A virus (H1N1). The following methods were used – the MTT-test and determination of antiviral activity according to three schemes: prophylactic (before infection), effect on adsorption (during infection), and post-exposure (effect on infection). Experiments were performed using MDCK cell culture and influenza virus strain A/FM/1/47. The studied AgNP were represented by three samples: Ag60C (a mixture of nanoparticles from 4 to 30 nm in diameter, mostly 8-10 nm), Ag40C (a mixture of 5 to 45 nm, mostly 10-20 nm), and Ag25C (a mixture of 5 to 100 nm, mostly 40-50 nm). Oseltamivir phosphate was used as a reference drug.

To assess the cytotoxicity, the CC50 index was calculated based on the results of the MTT-test. In the concentration range from 1×10^{-5} M to 1×10^{-9} M, nanoparticles did not have a significant effect on cell viability, so it was impossible to calculate the CC50 index for them. Oseltamivir phosphate demonstrated a classical dose-dependent effect, therefore, its CC50 was $2.6 \times 10^{-3} \pm 0.1 \times 10^{-3}$ M.

According to the prophylactic scheme of the determination of antiviral activity, Ag60C showed the highest efficiency among the three types of AgNPs - from $74.53 \pm 3.72\%$ to $92.37 \pm 4.61\%$ inhibition of virus reproduction. Ag40C also demonstrated relatively high efficiency - from $64.67 \pm 3.23\%$ to $75.36 \pm 3.76\%$ inhibition of virus reproduction. Ag25C nanoparticles, the largest in diameter, inhibited virus reproduction only at the highest tested concentration of 1×10^{-5} M by $4.34 \pm 0.08\%$, which is lower than that of the reference drug ($7.18 \pm 0.35\%$). These results well illustrate the dependence of the size of AgNPs on their activity since small nanoparticles quickly release Ag^+ ions, which are the active form of silver.

The next scheme of determination - the influence on the adsorption of virions - demonstrated the absence of an effect of AgNPs on this process: the highest percentage of inhibition of virus reproduction did not exceed 6% for Ag25C, oseltamivir phosphate also showed no activity. In the post-exposure scheme, the reference drug best inhibited the reproduction of the virus - by $27.19 \pm 1.35\%$. For the studied AgNPs, the inhibition efficiency of virus reproduction ranged from 7 to 11%, with the highest rate of $11.03 \pm 0.55\%$ for Ag25C.

Thus, taking into account the high prophylactic antiviral effect of AgNPs mixtures and their low cytotoxicity for MDCK cell culture, the studied nanoparticles can be considered potential candidates for further experiments as antiviral agents.