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Short communication

Commercial metal-based nanocolloids – lack of virucidal activity against ECBO virus

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Abstract

Metallic nanoparticles, mainly silver ones, have been widely used as antibacterial agents, and some studies shown they also exert direct antiviral activity against both enveloped and non-enveloped viruses. The objective of this study has been to evaluate the virucidal activity of commercial silver, gold, copper and platinum nanocolloids, recommended by the manufacturer as antimicrobials, against the ECBO virus, according to Polish Standard PN-EN 14675:2006. The highest experimentally observed decrease in the viral load was 0.875 log, which – when contrasted with the reduction in virus titre of at least 4 log expected from disinfectants – indicates that none of the analyzed nanocolloids had a disinfectant power towards the ECBO virus under the conditions defined by the standard.

Key words: commercial metal-based nanocolloids, virucidal activity, ECBO virus

Introduction

Nanomaterials, especially metal-based nanoparticles, have recently been assigned a broader range of applications in medicine, including the role of antimicrobial agents. In vitro research shows that silver nanoparticles enter into direct interactions with particles of such viruses as H1N1 influenza A virus, herpes simplex virus (HSV), respiratory syncytial virus (RSV), human immunodeficiency virus (HIV), hepatitis B virus (HBV), monkeypox virus and adenovirus type 3, thus inhibiting virus adsorption and entry into the host cells (Elechiguerra et al. 2005, Lu et al. 2008, Rogers et al. 2008, Sun et al. 2008, Baram-Pinto et al. 2009, Xiang et al. 2011, Chen et al. 2013). Such a mode of action is typical for virucidal agents. In view of the above, we decided to evaluate the virucidal activity of commercial silver, gold, copper and platinum nanocolloids, recommended by the manufacturer as antimicrobials, against the bovine enterovirus type 1 (ECBO virus).

Materials and Methods

Nanocolloids, test virus and cell culture

Four commercial colloidal nonionic solutions of silver, gold, copper and platinum metallic nanoparticles (Silver water, Gold water, Copper water and Platinum water; Nano-Tech Poland), suspended in demineralised water, at a concentration of 50 ppm (Ag, Au, Cu) or 20 ppm (Pt), were tested for their virucidal activities against the test virus.

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Bovine enterovirus type 1 (ECBO virus, ATCC VR-248), propagated in the MDBK cell line (the Madin Darby bovine kidney, ATCC CCL-22) served as the test virus.

Evaluation of virucidal activity of nanocolloids

The virucidal activity of nanocolloids was evaluated by a quantitative suspension test according to Polish Standard PN-EN 14675:2006 (PKN 2006). Briefly, the tested nanocolloids were put in contact with the ECBO virus under the required experimental conditions (contact time 30 min, temperature 10°C, interfering substances: 3.0 g/l bovine albumin for low level soiling or 10 g/l bovine albumin plus 10 g/l yeast extract for high level soiling), as well as additional experimental conditions (contact time 60 min, test without addition of interfering substances). Afterwards, both the control virus and experimental virus suspensions were titrated in a cell culture. Viral multiplication induced microscopically visible CPE. Calculations of the viral titre was made using the Spearman-Kärber method and expressed as a negative log of $TCID_{50}$/ml. A log reduction of 4 of the viral titre should be achieved for a disinfectant to pass the test successfully.

Results and Discussion

The highest decrease in the experimental viral titre caused by the nanocolloids was 0.875 log (Table 1). For comparison, a disinfectant is required to reduce the viral load by at least 4 log. Thus, none of the tested nanocolloids produced a disinfecting effect on the ECBO virus, neither under the required experimental conditions defined by the PN-EN 14675 nor under additionally set conditions.

Some reports describe direct interactions of nanoparticles (NPs), mainly silver ones, with virus particles, leading to some interference with the virus attachment to a host cell and inhibiting viral replication. The mechanism of these interactions is varied, depending on the virus. In most cases (HSV-1, RSV, H1N1 influenza A virus, HIV-1, Tacaribe virus) nanoparticles blocked sites binding the virus with the cell, although other developments have been recorded, e.g. nanoparticle interactions with viral genome (HBV) or even strong damage of viral particles (adenovirus type 3) in response to NPs (Elechiguerra et al. 2005, Lu et al. 2008, Sun et al. 2008, Baram-Pinto et al. 2009, Speshock et al. 2010, Xiang et al. 2011, Chen et al. 2013). Such interactions do not always lead to a complete loss of viral infectivity, but sometimes a notable effect was achieved, e.g. a decrease in the HA titre of H1N1 influenza A virus in response to AgNPs from 1:1024 to 1:2, a 7-log reduction of feline calicivirus titre under the influence of copper iodide NPs, or complete inhibition of Tacaribe virus replication in the presence of AgNPs (Speshock et al. 2010, Xiang et al. 2011, Shionoiri et al. 2012). The decline of the ECBO virus titre observed in this experiment from 0.5-0.875 log seems to imply some interactions of the analyzed nanocolloids with the test virus, but insufficent to claim they are virucidal agents useful in the veterinary area.

References

Baram-Pinto D, Shukla S, Perkas N, Gedanken A, Sarid R (**2009**) Inhibition of herpes simplex virus type 1 infection by silver nanoparticles capped with mercaptoethane sulfonate. Bioconjug Chem 20: 1497-1502.

Chen N, Zheng Y, Yin J, Li X, Zheng C (**2013**) Inhibitory effects of silver nanoparticles against adenovirus type 3 in vitro. J Virol Methods 193: 470-477.

Table 1. Results of the evaluation of virucidal activity of commercial nanocolloids.

Contact time (min)	Interfering substances	Virus titres (-log $TCID_{50}$)				
		control virus	nanocolloid/concentration (ppm)*			
			Ag/40	Au/40	Cu/40	Pt/16
30	–	7.5	6.75	7.0	6.75	6.75
	BSA	7.5	6.75	7.0	6.75	6.75
	BSA+YE	7.5	7.0	7.125	7.25	6.875
60	–	7.5	6.75	6.625	7.25	6.75
	BSA	7.5	6.75	6.75	7.25	6.75
	BSA+YE	7.5	7.25	6.875	7.5	7.0

Explanations:
* – nanocolloids were diluted to 80% of their baseline concentration during the test, BSA – 3.0 g/l bovine albumin (low level soiling), BSA+YE – 10 g/l bovine albumin plus 10 g/l yeast extract (high level soiling).

Elechiguerra JL, Burt JL, Morones JR, Camacho-Bragado A, Gao X, Lara HH, Yacaman MJ (**2005**) Interaction of silver nanoparticles with HIV-1. J Nanobiotechnology 3: 6.

Lu L, Sun RW, Chen R, Hui CK, Ho CM, Luk JM, Lau GK, Che CM (**2008**) Silver nanoparticles inhibit hepatitis B virus replication. Antivir Ther 13: 253-262.

PKN (**2006**) PN-EN 14675:2006 Chemical disinfectants and antiseptics – quantitative suspension test for the evaluation of virucidal activity of chemical disinfectants and antiseptics used in the veterinary area – test method and requirements (phase 2, step 1), Polish Committee for Standardization, Warszawa, p 28.

Rogers JV, Parkinson CV, Choi YW, Speshock JL, Hussain SM (**2008**) A preliminary assessment of silver nanoparticle inhibition of monkeypox virus plaque formation. Nanoscale Res Lett 3: 129-133.

Shionoiri N, Sato T, Fujimori Y, Nakayama T, Nemoto M, Matsunaga T, Tanaka T (**2012**) Investigation of the antiviral properties of copper iodide nanoparticles against feline calicivirus. J Biosci Bioeng 113: 580-586.

Speshock JL, Murdock RC, Braydich-Stolle LK, Schrand AM, Hussain SM (**2010**) Interaction of silver nanoparticles with Tacaribe virus. J Nanobiotechnology 8: 19.

Sun L, Singh AK, Vig K, Pillai SR, Singh SR (**2008**) Silver nanoparticles inhibit replication of respiratory syncytial virus. J Biomed Nanotechnol 4: 149-158.

Xiang DX, Chen Q, Pang L, Zheng CL (**2011**) Inhibitory effects of silver nanoparticles on H1N1 influenza A virus *in vitro*. J Virol Methods 178: 137-142.