Antibacterial effect of nanoparticles on foodborne bacterial biofilms by nanosecond laser-induced forward transfer

P.N.Lebedev Physical Institute of the Russian Academy of Science

A. Nastulyavichus[1], S. Kudryashov[1] , E. Tolordava[1,2],
Yu. Yushina[3], A. Semenova[3], A. Ionin[1]

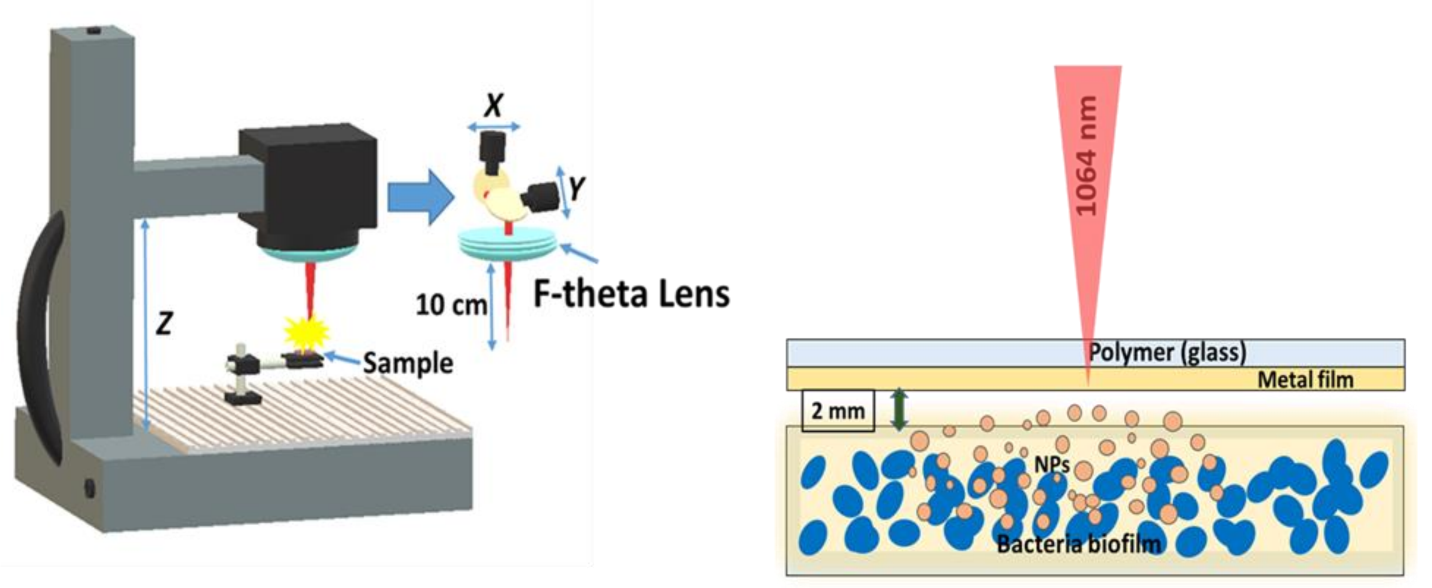
1-P. N. Lebedev Physical Institute of the Russian Academy of Sciences, Russia
2-N.F. Gamaleya Federal Research Centre of Epidemiology and Microbiology, Russia
3-Federal State Budgetary Scientific Institution "Federal Scientific Center for Food Systems named after V.M. Gorbatov "Russian Academy of Sciences, Moscow, Russia

e-mail:ganuary_moon@mail.ru

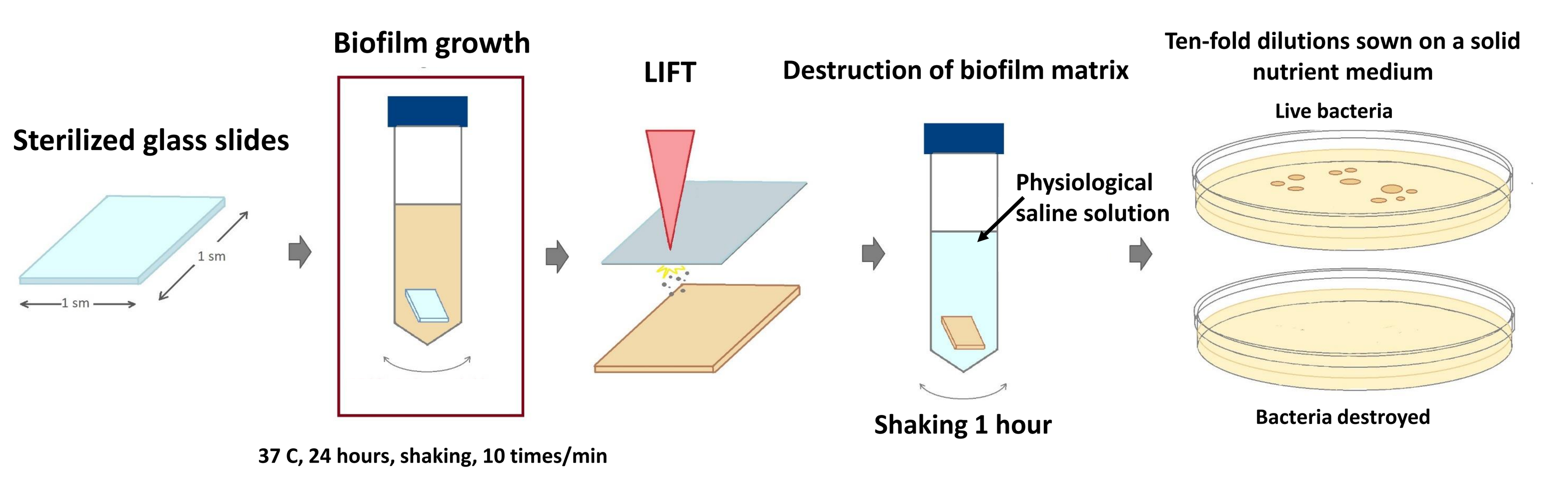
Introduction

The most bacteria lives in the form of the biofilm and costs humanity lots of infections and diseases. For the food industry it is vital to have bacteria under control, and it's important to not only maintain existing ways but also research for new ones, since well-known pathogens may adapt, and the emerging pathogens may be already resistant to what's currently in use. Laser-induced forward transfer (LIFT) of metal bactericidal nanoparticles from a polymer substrate directly onto food bacterial biofilms demonstrated its unprecedented efficiency in com-bating pathogenic microorganisms. Here, comprehensive study of laser fluence, metal (gold, silver and copper) film thickness and transfer distance effects on the antibacterial activity regarding biofilms of Gram-negative and Gram-positive food bacteria (Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli, Listeria monocytogenes, Salmonella spp.) indicates optimal operation regimes of the versatile modality. LIFT-induced nanoparticle penetration into a bio-film was studied by energy-dispersion x-ray spectroscopy.

Experiment

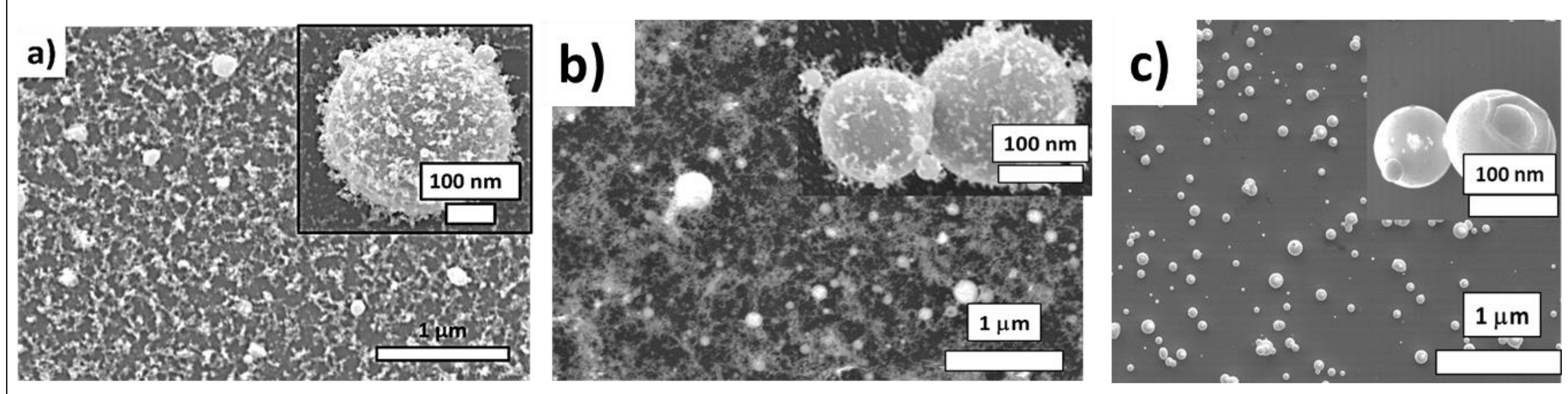


Yb-doped fiber nanosecond laser (wavelength - 1064 nm, pulsewidth - 120 ns, maximal pulse energy - 1 mJ, maximal pulse repetition rate - 80 kHz)



Scheme of the antibacterial investigations

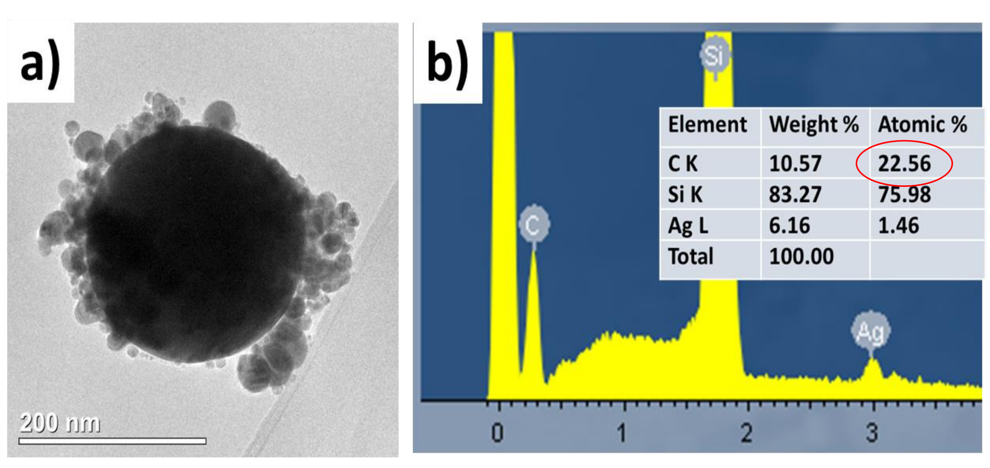
Nanoparticle characterization



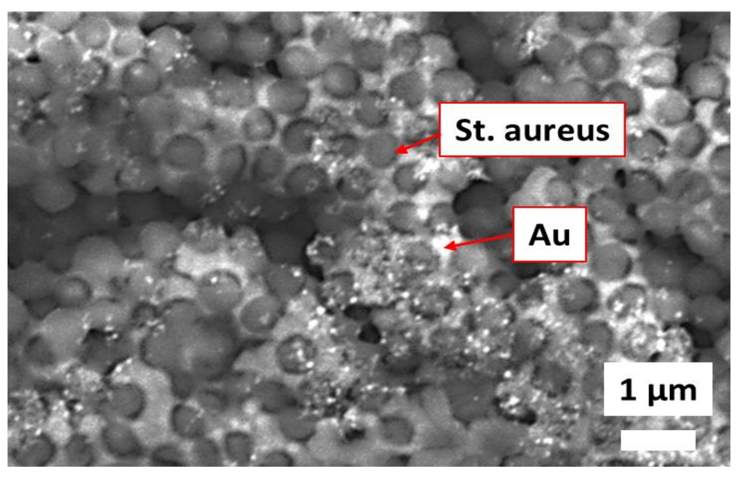
SEM visualization of Ag NPs (a), Cu NPs (b), Au NPs (c) laser-transferred onto the acceptor Si substrate from the donor PET

silver and copper nanoparticles are coated on top with a "fluffy" shell

Particle sizes: 10-400 nm

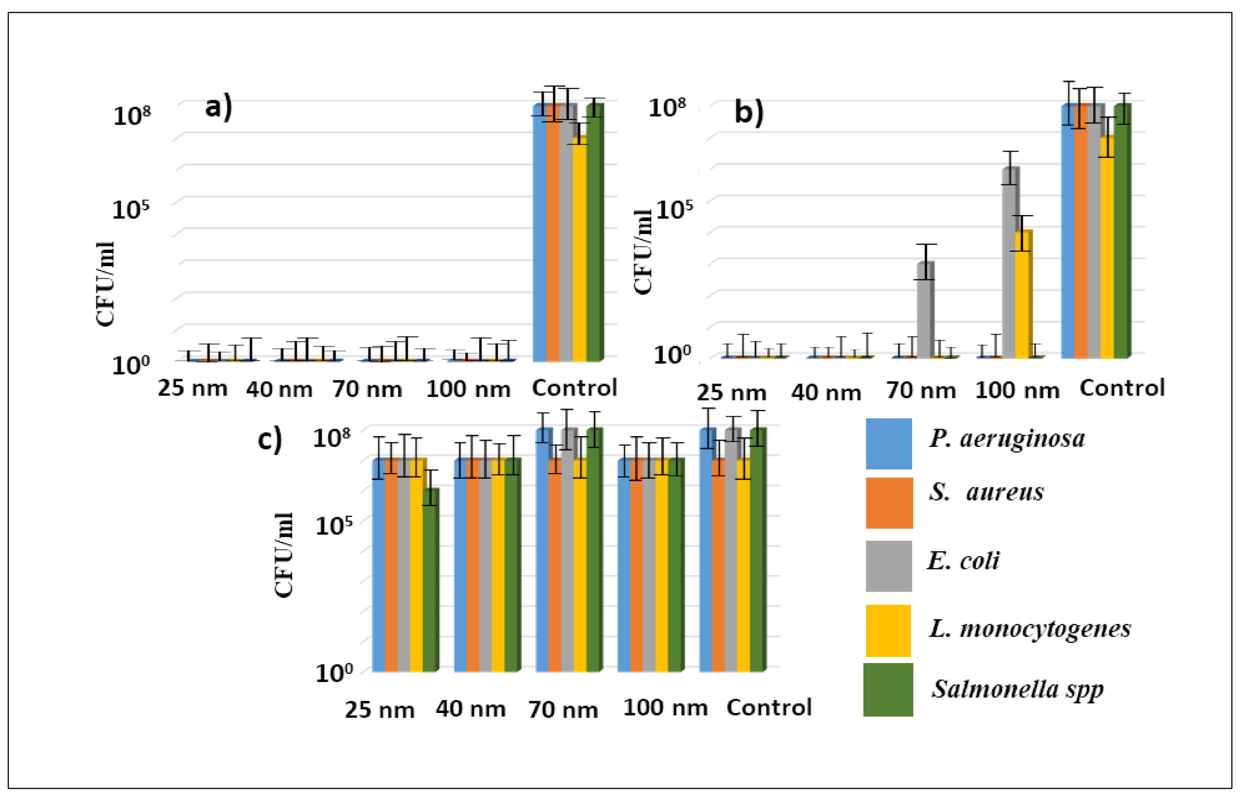


(a) TEM visualization of single Ag nanoparticle laser-transferred from the PET substrate; (b) Data (spectrum and datasheet) of its EDX analysis on the acceptor Si substrate, which also appears in the spectrum

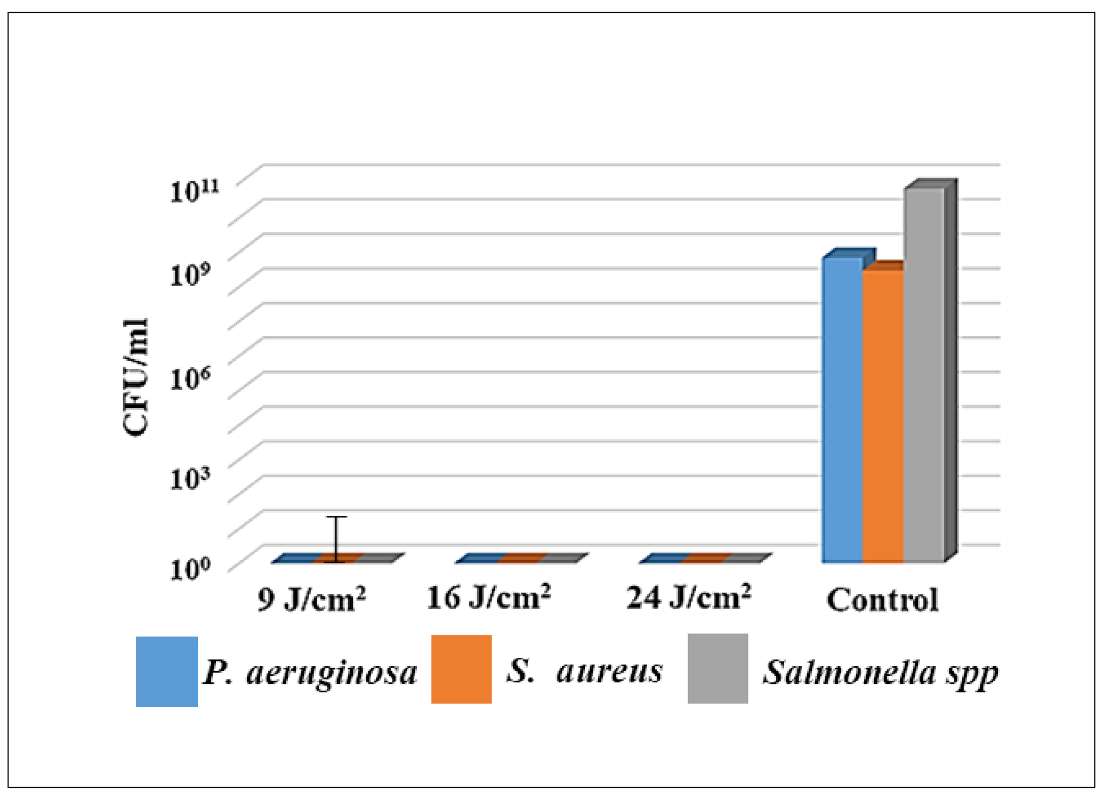


SEM visualization of Au NPs transferred on *St. aureus* bacterial biofilm

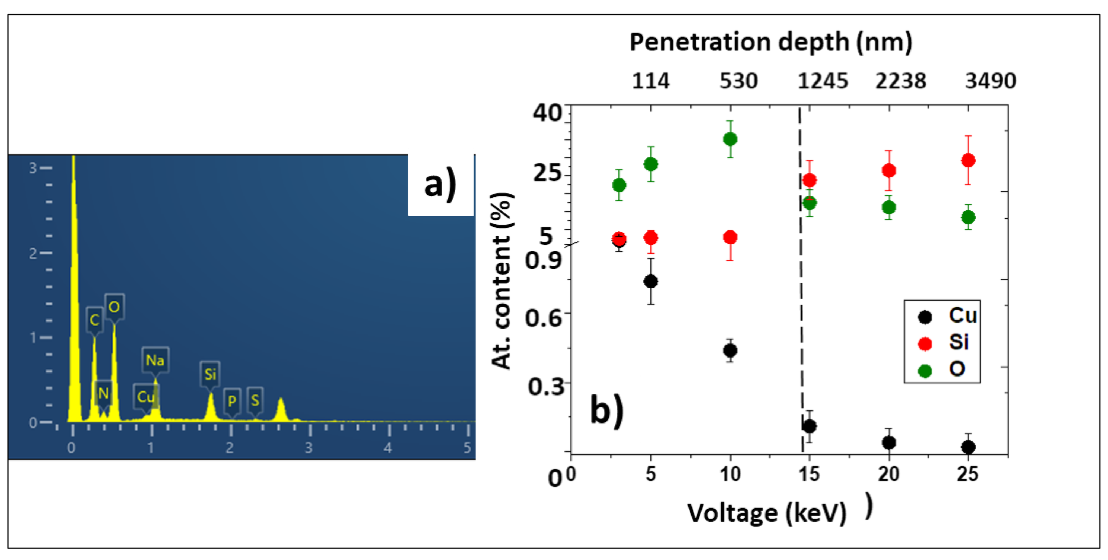
Antibacterial activity



Antibacterial effect (in CFU/ml) of Cu NPs (a); Ag NPs (b); Au NPs (c) transferred from metal films of various thicknesses on biofilms of clinical isolates.



Antibacterial effect (in CFU/ml) of Ag NPs transferred at various laser fluences from silver metal films on biofilms of clinical isolates



In-depth EDX distribution of oxygen (biofilm component), silicon (substrate component) and copper (component of transferred NPs), derived from their voltage dependences (c). The vertical dashed line indicates the Si substrate position and the rear side of the biofilm

Conclusion

In this work, laser-induced forward transfer of antibacterial nanoparticles onto biofilms of Gram-positive and Gram-negative food bacteria was in details studied in terms of basic regimes by varying laser fluence, metal type and film thickness, and nanoparticle transfer distance. Nanoparticles of silver and copper have proven their excellent antibacterial activities in comparison to chemically-inert gold, by demonstrating almost complete suppression of the food bacterial biofilms. The EDX method was used to estimate the penetration depth of LIFT-transferred nanoparticles inside the biofilm to enlighten their bactericidal effect. Thus, it was shown that nanoparticles did not penetrate into the entire biofilm and thus acted on it indirectly from outside. This method allows to quickly inactivate mature bacterial biofilms. It can be used in medicine for the treatment of wounds as a plaster and in the food industry for the treatment of food surfaces.

The research was supported by the Ministry of Science and Higher Education of the Russian Federation (Project No. 075-15-2020-775).