However, the surface of a gold film modified with carboranethiol C exhibited a significantly smaller number of cells, 51600±1900cells/cm^2 (FIG. 3). This may be explained by the exposure of the CH vertices of the carborane cluster, which are more acidic than the BH vertices exposed toward the cells in either A or B. Nevertheless, the cells on all tested surfaces were able to form a confluent layer.

The cell spreading areas on day 1 after seeding were significantly larger on the bare gold sample (2700±270μm^2) than in all remaining experimental groups. In these groups, the cell spreading areas were in the range from 1650±80μm^2 (on the samples modified with derivative D) to 2140±240μm^2 (on polystyrene dishes) but these differences were not statistically significant (FIG. 3).

Conclusion

Modification of a gold surface with carboranethiol derivatives A, B and D increased the population density of rat aortic smooth muscle cells after 7-day-cultivation on these surfaces in comparison with standard polystyrene cell culture dishes. However, on derivative C, the cell population density was significantly lower. This may be associated with the orientation of the carborane cluster, in which the acidic CH vertices face upward from the surface. Carboranethiol derivative D has thiol groups attached in the opposite (i.e., para-) positions, and can be considered as a promising linker for the attachment of various biological molecules to a gold surface.

Acknowledgements

This work was supported by the Grant Agency of the Acad. Sci. CR (Grant No. KAN400480701). Mr. Robin Healey (Czech Technical University, Prague) is gratefully acknowledged for his language revision of the manuscript.

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BIOMATERIALS WITH ANTIBACTERIAL ACTIVITY

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Abstract

The sol-gel derived silica spheres with surfaces modified by silver nanoparticles were used to enhance the photodynamic effect. The silica nanoparticles were prepared by modified Stöber synthesis. The diameter of obtained silica spheres was ca. 100 nm. These silica spheres we used as a carrier for noble nanoparticles. It was shown that Ag-Au nanoparticles have an antibacterial activity against Escherichia coli. This effect depends on the nanoparticles concentration and it is stronger for higher concentrations. Laser irradiation enhances this effect, and starting from certain concentration it is possible to kill Escherichia coli, totally, when using laser light.

[Engineering of Biomaterials, 81-84, (2008), 119-121]

Introduction

Recently, many new developments in nanomedicine are observed. One of the promising application of nanomaterials is in photodynamic medicine, where the nanoparticles may enhance the photodynamic activity [1-3]. Photodynamic therapy (PDT) over the past decades was mainly exploited for treatment of tumors. It can be also suitable for the inactivation of microbes by photodynamic activity. Antimicrobial photodynamic therapy (APDT) combines a nontoxic photoactive dye - photosensitizer to generate singlet oxygen and free radicals after light exposure that kill microbial cells [4-8]. In this paper we will demonstrate that silver-doped nanoparticles have antimicrobial activity and this effect may be enhanced by adding a photosensitive agent and exposing the bacteria culture to the light.

Materials

First, the silica nanoparticles were prepared by modified Stöber synthesis [9] from ethyl alcohol (95%, Polish Chemicals), ammonium water (25%, Polish Chemicals), hydrofluoric acid (35%, Polish Chemicals) and tetraethylortosilane (TEOS from Aldrich) mixed at room temperature prepared. Next, the Tollen's method for silica silver doped nanospheres production was exploited [10]. The preparation process used in this study is described in [11]. For reduction reactions glucose was used as the reducing agent. In second reduction reaction to the solution Ag-SiO_2 0,5M of $AgNO_3$ and 25% NH_4OH were added. For cementation process "gold liquid" (K_2CO_3+$HAuCl_4$+H_2O) was added.

As a photosensitive agents Photolon (18-carboxy-20-(carboxymethyl))-8-ethenyl-13-ethyl-2,3-dihydro-3,7,12,17–tetramethyl-21H, 23H–porphin–2-propionic acid) from Haemato, Poland was used. The diameter of silica nanoparticles was ca. 100nm. The concentration of noble particles is depicted in TABS. 1 and 2.

Method

The study was performed on Escherichia coli cultures. First, the bacteria were cultured in the presence of nanoparticles. The ordinary procedure for bacteria culturing, seeding and incubation were exploited. For irradiation 410 nm wavelength from TopGan laser was used, with peak optical power 200mW. The exposure time was 4 minutes. The nanomaterials in various concentrations were added to the bacterial suspension and then the bacteria were seeded on agar plates and the samples were incubated. To 1ml of suspension 0,25, 0,5 or 1ml of supernatant was added. The CFU indices (colony forming units) were estimated.

TABLE 1. Ag concentration in sediments.

Sediment samples	Concentration Ag [mg/dm^3]	Concentration Au [mg/dm^3]
Impregnation process	10.8	-
I reduction reaction	9.4	-
II reduction reaction	35.6	-
Cementation process	8.7	16.3

TABLE 2. Ag concentration in supernatants.

Supernatant	Concentration Ag [mg/dm^3]	Concentration Au [mg/dm^3]
I reduction reaction	5.3	-
II reduction reaction	2.6	-
Cementation process	1.4	1.8

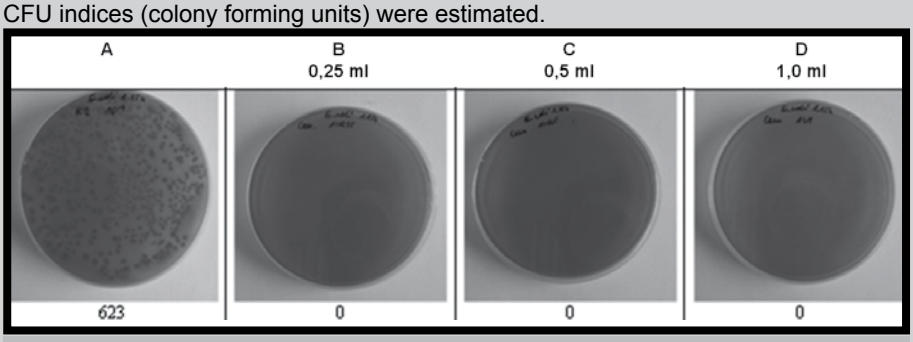

FIG. 1. Exemplary results: Ag-SiO_2-Au was added to E. coli suspension. A – control, B, C, D – samples cultured in the presence of nanoparticles.

Results

Exemplary results are shown in FIG. 1. The picture A demonstrates the E. coli in control sample, where as B, C and D are the culture incubated with nanoparticles.

The results demonstrating that the light irradiation has even more strong antibacterial effect are depicted on FIGS. 2-4.

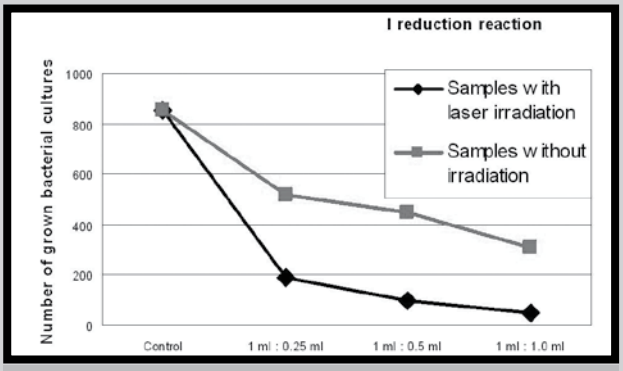

FIG. 2. Samples after I reduction. The influence of laser irradiation.

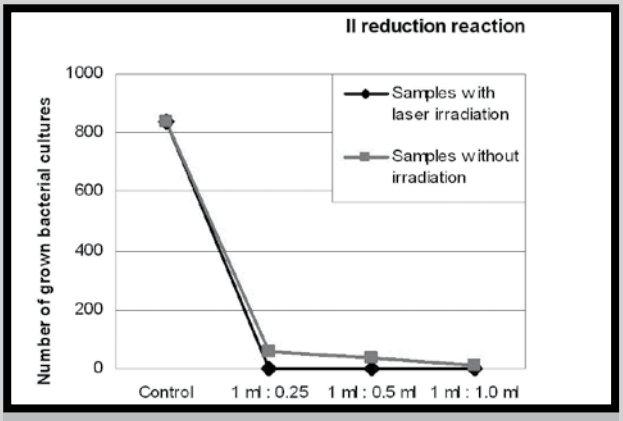

FIG. 3. Samples after II reduction. The influence of laser irradiation.

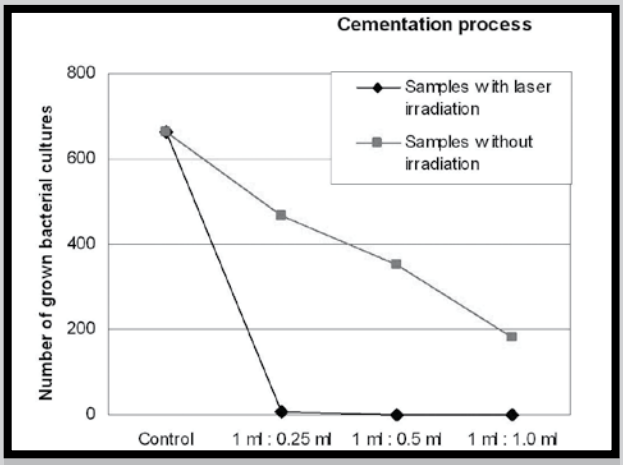

FIG. 4. Samples after cementation. The influence of laser irradiation.

Conclusions

In this work it was shown that Ag-Au nanoparticles have an antibacterial activity against Escherichia coli. This effect depends on the nanoparticles concentration and it is stronger for higher concentrations. Laser irradiation enhances this effect, and starting from certain concentration it is possible to kill Escherichia coli, totally, when using laser light.

Acknowledgements

The work was supported by the Polish Ministry of Education and Scientific Research, Grant No. N N518 327335.

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COMPARATIVE STUDY OF HYDROXYAPATITE AND HYDROXYAPATITE MIXED WITH BIOGLASS COATINGS OF METALLIC IMPLANTS, DEPOSITED BY PLD METHOD

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Abstract

Hydroxyapatite and bioglasses are important bioactive materials as they exhibit direct bonding with human bone. Therefore they are used as coatings for metallic implants. The aim of the present study was to compare two types of layers: monophase hydroxyapatite (HA) and composite-type HA+BG hydroxyapatite mixed with bioglass (BG) (BG 50% of weight) during the initial stage of their interaction with cell medium. In vitro studies were performed in order to determine the effect of the investigated layers on cell response.

After 3 and 7 days the behaviour of the cells grown on the above surfaces was estimated through determination of the cell adhesion (CV colorimetric assay). Cell morphology and properties of biomaterials surfaces were analysed by atomic force microscopy (AFM).

[Engineering of Biomaterials, 81-84, (2008), 121-123]

Introduction

Steel as well as titanium-based alloys are widely used as materials for implants because of their good mechanical properties. Unfortunately, they are not bioactive and therefore need to be coated with bioactive layers such as hydroxyapatite (HA) and/or bioactive glasses.

The chemical and structural charcteristics of HA promote chemical bonding to adjacent bone tissue [1,2], which leads to direct attachment to bone. Some glasses are also considered as bioactive materials. Ceramics in the SiO_2-CaO-P_2O_5 system are the class of biocompatible materials that can be used as coatings on metallic implants [3]. As a glassy phase, bioglasses can reveal high surface reactivity, which causes a strong attachment to surrounding bone tissue. It is possible to design bioglasses with properties useful for particular medical application [4]. In this work a novel approach of bioglass reinforced hydroxyapatite was used to increase osteoblasts response.

Among many techniques pulsed laser deposition (PLD) is commonly used for deposition HA films [5,6]. The physical as well as chemical properties of obtained coatings can be controlled by changing conditions of PLD method and a composition of target [7]. The crucial parameters of the PLD process are pressure of ambient gas, laser fluence, substrate temperature and laser wavelength [8,9].

The aim of this paper was to perform and evaluate ceramic coatings deposited by PLD method with the use of monophase HA and composite-type HA+BG targets. In this paper the topography of obtained coatings as well as initial stage of intaraction with cell culture medium is shown.

Materials and methods

Two targets of different chemical composition were used for deposition of coatings: the first one consisted of pure HA ($Ca_{10}(PO_4)_6(OH)_2$) while the second was the composition type HA mixed with 50%wt. of BG. The composition of BG was following: SiO_2–80mol%, CaO–16mol% and P_2O_5–4mol%. The target materials were deposited by use of pulsed excimer ArF laser which operated at the wavelength of 193nm with the pulse energy of ~300mJ and 20ns of pulse duration (FWHM). The laser fluence was 4±0,5J/cm^2 and the laser beam was focused of 40^O out of the normal surface of the target. All layers were deposited on 316L stainless steel substrates with nanocrystalline diamond (NCD) buffer layer which thickness was 300nm. The NCD layers were deposited by RF PACVD method described by