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The Creation of an Antimicrobial Coating on Contact Lenses by the use of Nanocopper

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***Abstract* — The aim of the research was to creation an antimicrobial coating on contact lens and examine the ability of copper nanoparticles to decrease microbial adhesion and prevent the growth of bacteria. The creation was based on the immersion method in solution with dispersed nanoparticles of copper at concentration 200 pm. There were evaluated follows factors: time of immersion and base to dispersed nanoparticles. Three solutions: ethyl alcohol 99.8%, lens liquid, and olive were tested and two variants of immersion, i.e.: double dipping for five seconds and immersion for a period of five minutes. The quality of the obtained coatings was controlled by using a biological microscope and a scanning electron microscope. Its effectiveness was tested by placing samples in the incubation medium of Staphylococcus aureus for 30 days.**

***Index Terms* — contact lenses, nanoparticles, nanocopper, antimicrobial coating, eye infection, bacterial biofilm.**

I. Introduction

The use of contact lenses may be one of a predisposing factor for infection of the eye's area. The precursors of these ocular infections are: bacteria, viruses or fungi [1]-[3]. It's assumed, that significant impact on the initiation of this disease is the accumulation of protein deposits on the surface of contact lenses [3], [4]. Additionally, it's noticed, that the greater susceptibility to infection are, so-called, ,,color contact lenses" (which use can modify eye's color), which contain special pigments [5], [6]. The most common ocular infection is a bacterial infection, which may be caused by bacterial strains, such as e.g. [7]-[9]:

- Staphylococcus aureus,
- Streptococcus viridans
- Pseudomonas aeruginosa
- Serratia marcescens
- Escherichia coli
- Enterococcus faecalis
- Streptococcus penumoniae.

Commonly occurring infection of the eye is conjunctivitis. It's basic symptoms include inter alia: redness eye, increased amount of tears, mucopurulent discharge, burning eyes, increased sensitivity to light, itchy eyes and blurred vision. In most cases its spontaneously cured during 1-2 weeks. However, in the case of other types of infections (e.g. viral or fungal infection) or chronic inflammation requiring the use of specialized treatment, i.e.: antibiotic therapy [8]-[10].

The main aspect in bacterial infection is their colonization in concentrated communities, called biofilm. Bacteria deposited on a surface of the lens and/or of the eye and next surrounding by a specific matrix, which consists of mainly exopolysaccharides and proteins, were excreted by them. In this particular form of the symbiotic colony, bacteria are acquiring additional properties, such as: increases their resistance to adverse environmental conditions, i.e. e.g., dryness or harmful substance. In practice makes bacteria difficult to combat by the immune system, antibiotic therapy or germicides. These communities of bacteria can be created by one strain of bacteria or more, which further complicates treatment. Bacterial biofilms produce specific endotoxin, which in the human body leads to inflammatory reactions and may increase the susceptibility to bacterial, by altering its permeability [11]-[14].

Thus far, the main way to fight with infections was their prevention. The primary means is a contact lens liquid, which has antimicrobial properties. In addition, a relatively new alternative are containers to store contact lenses, which are impregnated with silver [9]. As a result of the growing problem of microbial infections began to modify contact lenses. There were conducted numerous studies, test or modifications, such as: attempts to invent a new material, use of a suitable additive or create a coating. So far, in order to reduce the frequency of bacterial infections tested additives or coating on the basic of: antibiotics (e.g. cefazolin, vancomycin, minocycline-rifampin, metals (e.g. silver, copper) or other substances or compounds, such as e.g.: salicylic acid, melanin, chlorhexidine, ammonium compounds, cationic peptides. The main problem in the case of attempts to modify the contact lens was usually brevity and cytotoxicity [4], [15]-[17].

A recently popular alternative to combating infection or its therapy is the use of metal nanoparticles, which have antimicrobial properties and in particular bactericidal. The primary advantage is the absence of resistant bacteria, as in the case of antibiotic therapy [18], [19]. Their bioactive properties are mainly due to their high activity. It results, among others, that the nanoparticles have a substantially

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dispersed metal phase, they are in constant motion, have a high surface to volume ratio and haven't appended anionic groups. Thanks to these properties nanoparticles are able to generate positive charges, which cause their mutual repulsion. As a result, they may be able to penetrate the interior cells of bacteria, which have negative charges. The nanoparticles cause toxic effects on microbes, such as e.g.: disruption of the cell membrane, disturbances in the process of cellular respiration, interruption of basic activity enzymes (i.e. the metabolism and electron transport) and inhibiting replication processes (by binding DNA or RNA chain) [20]-[22].

It has been conducted a number of studies regarding the effectiveness of the nanoparticles (especially nanosilver), which proved that has a toxic impact on bacteria, by weakening the adhesion to the lens, prevents the formation of biofilm and as a result reduces the risk of bacterial infection [16], [19], [22]. Moreover, another example of the studies was nanocopper. It confirmed its potency to inhibiting the growth of bacteria E. Coli and Staphylococcus aureus [21].

Furthermore, it should also pay attention to the possibility of the use of contact lenses in the treatment of infections, as they have a significant advantage over conventional treatment. Currently used ophthalmic drops or ophthalmic solution generate temporary high concentration, during their application and in this period its effective treatment. However, it has a decreasing trend, as a result of blinking are fuzzy and diluted in tears – by this can lead to treatment failure. It's suspected that the use of additive or coating based on nanoparticles would provide a permanent and effective therapeutic management [23].

The aim of the work was to create an antimicrobial coating on the contact lenses with nanocopper particles.

II. Materials and Methods

A. *Contact Lenses*

In the research soft contact lenses „Air Optix Night&Day" by CIBAVISION in the composition: 76% lotrafilcon A and 24% water were used. They were pre-packaged in blisters and stored in a buffered isotonic saline containing 1% Copolymer 845.

Their basic parameters are [24]:

- The oxygen permeability Dk – 140 Dk/t,
- The thickness of the central – 0.08 mm,
- Base curve – 8.60 mm,
- Diameter – 13.80 mm,
- Power – 1.50 D [-].

B. *Creation of the coating*

The coating was prepared using the immersion method. Contact lenses are immersed in a prepared solution with dispersed nanoparticles of copper. Selected two variants of immersion, i.e.: double dipping for five seconds and immersion for a period of five minutes.

C. *Bases for mixing nanoparticles*

Three different solutions, in which nanoparticles were dispersed are used: 99.8% ethyl alcohol, the contact lens liquid - „Opti-Free PureMoist" (Alcon) and olive called „Bambino". The composition of the contact lens liquid is as follows: matrix moisturizing HydraGlyde – 99.9% and preservatives: Polyquad (polyquaternium-1) – 0.001% and ALDOS – 0.0006%. However, in the olives are: Glycine Soja Oil, Paraffinum Liquidum, Parfum, Ethyl linoleate, Ethyl Oleate, Ethyl linolenate, Tocopherol, Propylene Glycol, BHA, Propyl gallate, Citric Acid.

The concentration of the solution with nanoparticles, was chosen on the basis of results obtained at work [19], it was observed apparent tendency to decrease the percentage of bacterial viability, with increasing number of nanoparticles. Satisfactory results, i.e. 60% loss of viability of bacterial flora has been obtained at a concentration of 80 ppm. Hence, in order to ensure the effectiveness of selected concentration of the solution with dispersed nanoparticles was about 200 ppm.

D. *Nanoparticles*

The copper nanoparticles produced by company MkNano were used. Their size was 50 nm. The amount of dispersed nanoparticles was selected in order to achieve the appropriate concentration of the solution and was 0.04 grams.

E. *Incubation of bacteria*

In order to prepare the substrate for the incubation of bacteria was used following work. First, it was prepared typical medium for bacteria by mixed together in one liter of distilled water the following ingredients: casein peptone (17 g), peptone S (3 g), sodium chloride (5 g), dipotassium phosphate (2.5 g) and glucose (2.5 g). This solution was maintained at a pH of approximately 7.3 [25]. Next, it was added Staphylococcus aureus and thoroughly mixed. In a next step, the solution was placed in the oven at a temperature of 38° C and atmospheric pressure conditions. After a time of one hour, the solution was again mixed thoroughly and poured into special flasks, in which the samples were placed. These prepared flasks are placed back in the incubator for 30 days (the Patent number P 409082).

F. *Measuring equipments*

To control the results of the research two microscopes were used. The first is biological microscope ZEISS with the highest magnification of 1000x, and the second is a scanning electron microscope JEOL JSM-7800F with a maximum resolution of 0.8 nm and the largest magnification close 1000000x.

III. Results

A. *Immersion in alcohol*

First, the contact lens was immersed in an alcohol solution with dispersed nanoparticles of copper for five minutes. The results show Fig.1. A uniformly deposited coating on the entire surface of the lens was observed.

Fig. 1. *The contact lens with the obtained coating /immersion for five minutes/*

The quality of the obtained coating was tested using a biological microscope (Fig. 2). There was a trend towards the formation of nanoparticles agglomerations. The larger conglomerates were marked with a red circle. Moreover, using a scanning electron microscope, it was found that nanoparticles formed a specific coating (Fig. 3) – is composed of uniformly deposited nanoparticle conglomerates.

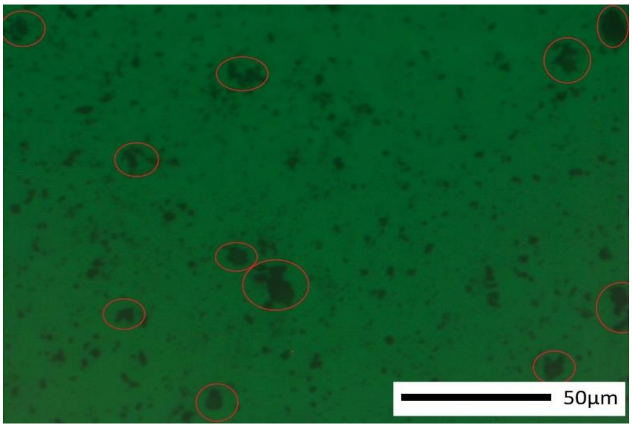

Fig. 2. *The distribution of nanocopper agglomeration on the surface of the contact lens /immersion for five minutes/ [500x]*

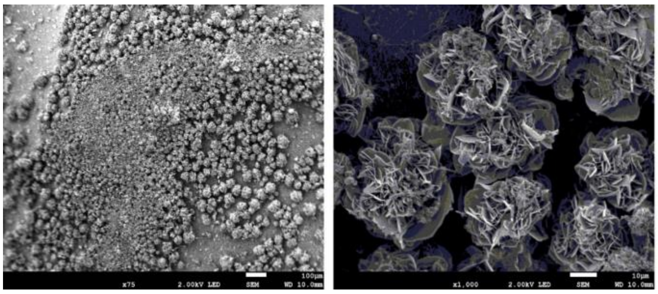

Fig. 3. *The distribution of nanocopper agglomeration on the surface of the contact lens /SEM/*

B. Immersion in contact lenses liquid

Next, the contact lenses were immersed in a contact lenses liquid solution with dispersed nanoparticles of copper for five minutes. The obtained coating was also tested by using a biological microscope (Fig. 4). It has been found that there are much larger conglomerates than in the case of the previous solution.

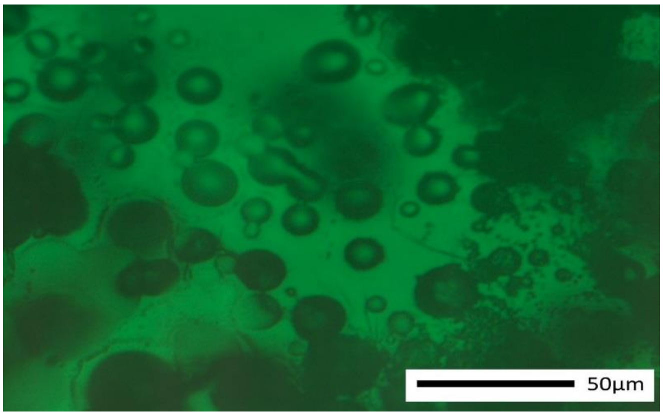

Fig. 4. *The distribution of nanocopper agglomeration on the surface of the contact lens /immersion for five minutes/ [500x]*

C. Immersion in olive

Then, the contact lenses were immersed in an olive solution with dispersed nanoparticles of copper twice for five seconds. The obtained coating was also tested by using a biological microscope (Fig. 5). For this method, the most satisfactory results were obtained. There was a uniform distribution of conglomerates of relatively similar size.

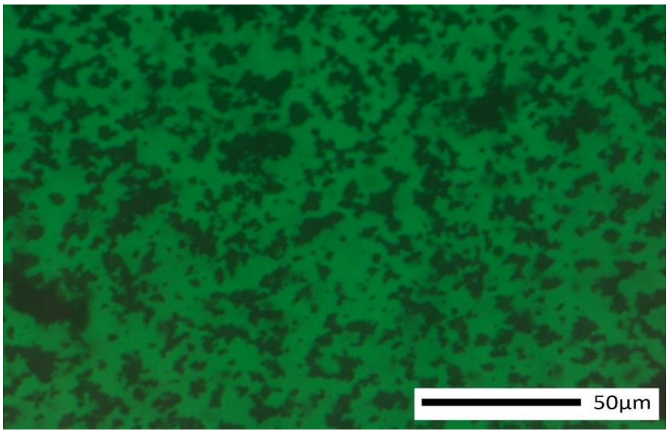

Fig. 5. *The distribution of nanocopper agglomeration on the surface of the contact lens /double dipping for five seconds/ [500x]*

IV. Discussion

Three types of bases: alcohol, contact lenses liquid and olive (Fig. 6) were studied. The choice of the base affects the distribution of nanoparticles (selected conglomerates marked with a red circle). In the case of alcohol, nanoparticles deposited relatively infrequently. It can be seen on the surface of some conglomerates. While, in the case of contact lens liquid there is a greater tendency to form larger conglomerates, but there are also small ones. However, in the case of olive, there is the greatest tendency to deposition and this deposition is comparatively evenly. Suspected important effect on the viscosity of the selection base to deposition of nanoparticles.

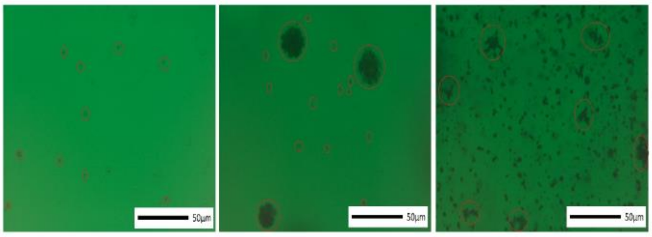

Fig. 6*. Comparison of the distribution of nanocopper agglomeration using different bases, i.e.: alcohol, contact lenses liquid, olive in double dipping for 5 seconds [500x]*

Increasing immersion time of the lenses in the solution affects the deposition of nanoparticles on their surface. In the case of five minutes, there was a significant deposition of nanoparticles in the huge conglomerates. However, in the case of double-dipping for five seconds was observed formation several smaller conglomerates (red circles Fig. 7 and Fig. 8).

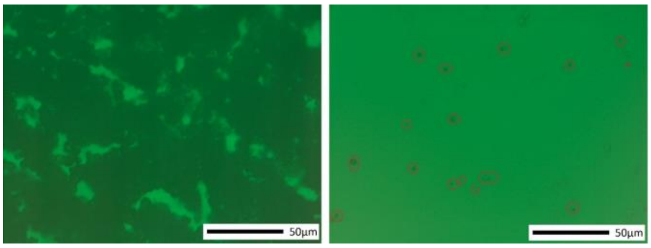

Fig. 7*. Comparison the effect of immersion time (alcohol) on the distribution of nanocopper agglomeration: 5' and 2x5'' [500x]*

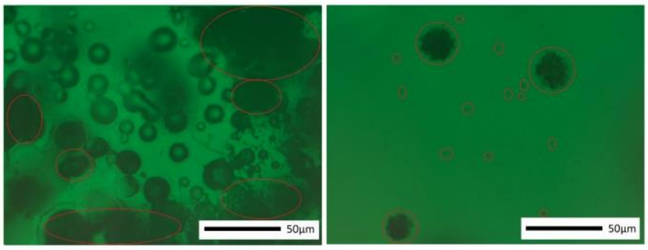

Fig. 8*. Comparison the impact of immersion time (contact lenses liquid) on the distribution of nanocopper agglomeration: 5' and 2x5'' [500x]*

The biological efficacy of the obtained coating was tested by placing the samples in the incubation medium of bacteria Staphylococcus aureus for 30 days. There were prepared control samples, that was normal unmodified contact lenses. A significant deposition of bacteria (red circle on Fig. 9) and formed bacterial biofilm (Fig. 10) were noticed on its surface.

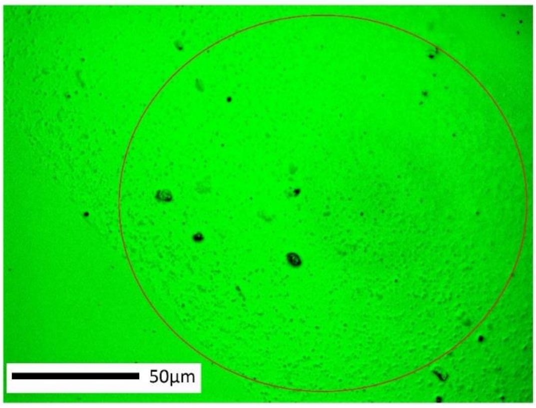

Fig. 9*. The surface of the controls ample removing from bacteria medium [500x]*

Fig. 10*. The biofilm on the surface of the contact lens*

Research results of bacteriological tests are shown in Fig.11. It was found, that alcohol can not be the base for the nanoparticles, because the samples have a higher deposition of bacterial biofilm than the control samples. It is suspected, that alcohol can disrupt the physicochemical properties of the lenses. In turn, for the contact lens liquid was observed to reduce the deposition of bacteria. In contrast, advantageous results obtained using olive and the mixture of olive and alcohol. It is stated for all the samples, that prolonged immersion time affects the greater destruction of bacteria.

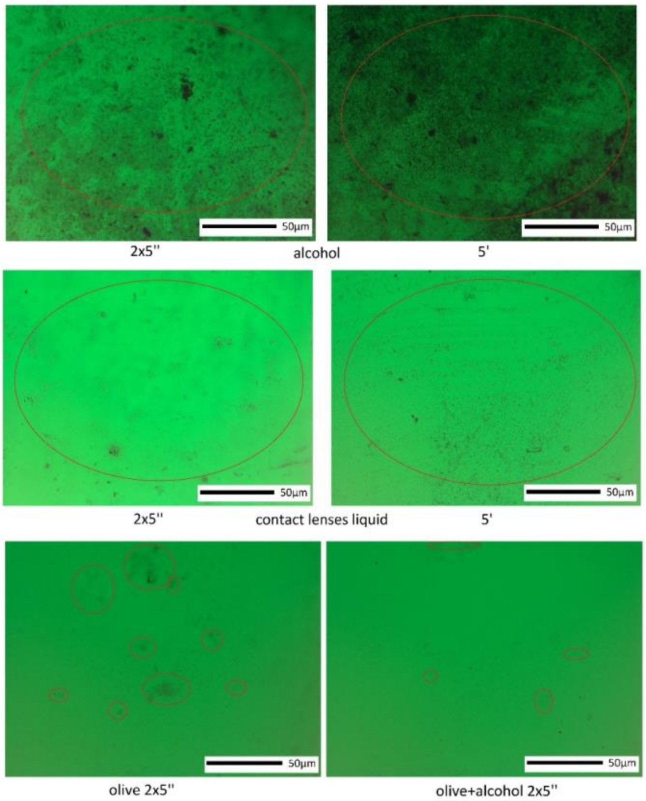

***Fig. 11.** Comparison of the distribution of bacteria: Staphylococcus aureus on the surface of the tested lens [500x]*

V. Conclusions

- A significant impact on the creation of coatings have: time of immersion and choice of base.
- It was observed, that increasing immersion time reduces the deposition of the nanoparticles and as a result affect the effectiveness of the coating.
- The main aspect of the immersion method was the selection of the base to dispersed nanoparticles. It is assumed the hypothesis that one of the important elements in the selection of the base are the density and viscosity. It is expected that the increase occurs favorable distribution of nanoparticles on the entire lens surface and a lower tendency to form conglomerates.
- The fundamental problem in the creation of coating on the contact lenses was its instability. By checking the distribution of nanoparticles was observed after one month of their significant leaching. This is due to the fact, that the lenses are kept constantly in an aqueous medium – i.e. a specialized contact lenses liquid.
- It is possible to create an effective coating by this method. It is considered in the selection of appropriate parameters and proper base of creation a coating to reduce the adhesion of bacteria to the surface of the lens. Additionally, it is confirmed the efficacy of the nanocopper in combating bacteria.

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