FILMS VENTURESHIELD™ VS 7510 AS A WAY THE ANTIBACTERIAL PROTECTION OF SANITARY AMBULANCES

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Abstract

In the study the research findings, regarding the application of innovative solutions from the field of the material science and medicine in the interior of modern ambulances, are described. VentureShield VS 7510 E film (I and II grade) was evaluated from the point of view of high sterility level of the interior of an ambulance. The examinations were divided into two stages. Firstly, a degree of the adhesion of chosen kinds of the bacterium e.g. *Enterococcus faecalis* and *Pseudomonas aeruginosa* was evaluated. In the second stage, samples toxicity tests were conducted using the normal human dermal fibroblasts (NHDF). It has been proved that VentureShield VS 7510 E film (I and II grade) effectively protects against bacterial adhesion and it is not toxic to living cells, such as NHDF. We can assumed that the film will fulfil its protective function in sanitary ambulances.

Keywords: bacteria, fibroblasts, adhesion, ambulance, films

1. INTRODUCTION

The interior of an ambulance encourages microorganism adhesion. Many educational institutes conduct experiments in order to prevent pathogens growth and to eliminate them completely. Each unit using ambulances should obey some rules which aim at keeping a proper sanitary and epidemiological conditions.

The disinfection of the ambulance interior and the medical equipment used in it, is not always maintained on a high level. Despite the efforts and implementation of further guidelines it is still a current global issue. The conducted research has proved the presence of pathogens causing illnesses inside ambulances [1, 2]. The bacteria isolated from the interior sheathing of an ambulance may threat patients suffering from low immunity or during immunosuppressive therapy, especially *Enterococcus faecalis*, *Pseudomonas aeruginosa, Staphylococcus haemoliticus*. This fact should influence the selection of disinfectants and materials covering the interior of medical vehicles [3-6].

One method of protecting varnished surfaces against damage is using protective films. It is common in automotive industry. Films are applied on the cars elements, especially threatened by mechanical damage, i.e. bumpers, inner wheel arches, doors. Films are wear and scratch resistant, and are not damaged by chemical factors (roads salinisation). Their use allows for the increase of the functionality of protected surfaces and it aids to retain esthetical appearance for a long period of time [7].

Literature presents surveys on the behaviour of microorganisms on different surfaces, among others, on films, to begin with the commonly used aluminum foil [8], through the research on copper foil [9], which presented antibacterial properties. The application of films inside ambulances and steel surface in medical units is worth considering.

2. THE AIM AND THE SCOPE OF RESEARCH

The study is the continuation of the previous research on the selection of the material and anticorrosion coatings which would be the best to maintain antiseptic properties inside an ambulance. The study presents the results of effectiveness evaluation of VentureShield VS 7510 E film used inside an ambulance. The results which were achieved confirm that the film poses a corrosion protection of DC01 steel, in contact with physiologic solutions and ethanol based disinfectants. It helped to prove the protection effectiveness of the film when used in hospital conditions. However, it would be necessary to prove its corrosion protection properties. Owing to that, the study was divided into two parts. The first one concerned the adhesion of selected bacteria strains, i.e. *Enterococcus faecalis* and *Pseudomonas aeruginosa* on the examined surfaced, covered with the film. The second part was focused on its cytotoxic properties, in terms of mouse fibroblasts tested in direct in vitro way.

3. MATERIALS AND METHODS

3.1 Sample preparation

The DC01 steel was selected for the research in form of cold-rolled sheet-metal, commonly used for medical equipment production. The samples, size 100 x 100 mm, were cut out of accidentally chosen sheet metal areas, and then they were ground and polished. The samples prepared in this way were covered by thermoplastic polyurethane VentureShield VS 7510 E film, which size depended on the stage of the research. Two types film were selected for the research - grade I and II (the grades specification was restricted by the producer). Acrylic adhesive of high resistance was selected by the film manufacturer. Isopropyl alcohol was used as the solvent, and gelatin as a thickener. The initial thickness of the film and adhesive was 210 µm, specific weight 240 g/m^2, tensile strength and elongation 5400 N/cm^2, 460 %.

3.2 The growth of bacteria

The research consisted in the incubation of selected bacteria species - *Pseudomonas aeruginosa* ATCC 27853 and *Enterococcus faecalis* ATCC 29212 (density 0.5, 2 and 4 McF) in the 3 ml of physiological saline solution (0.9 % NaCl) on metal plates with a film of 7mm diameter.

Before culturing the bacteria, the samples were subjected to surface sterilization under exposure to UV light for 24 hours. In the next step, plate asepsis was controlled. Finally, selected bacteria species were cultured on given surfaces and incubated for 24 hours in room temperature of 25 ^{o}C. After the incubation period, the plates covered with the film were cleaned three times by a solution of physiological saline (0.9 % NaCl) and left until they dried.

Pressure-sensitive substrate, type Coun-Tact® media (bioMerieux, Fr) was applied to the dry surface of plates in order to check surface asepsis of them. The number of bacterial colonies on the contact plates was counted after 24 hours. The results are presented by number of bacterial colonies per surface square centimeter (CFU/cm^2 - colony forming unit/cm^2).

3.3 Cell culture

NHDF were isolated from plastic surgery skin sections with approval from the Ethical Committee of the University Hospital Olomouc and the patient's consent. The study was performed in accordance with the Code of Ethics of the World Medical Association. The morphology and origin of the cells were authenticated in the Histology Department, University Hospital Olomouc.

NHDF were cultured in Dulbecco's Modified Eagle Medium supplemented with 10% fetal bovine serum (FBS) and 1 % penicillin-streptomycin under standard culture conditions (5 % CO_2, 37 °C). Cells were used between the 2nd and 3rd passages.

The cytotoxicity of VentureShield VS 7510 E film was observed by the examination of direct contact. The circle samples of 3.5 cm diameter and square samples (its side of 1 cm) were cut out of the film. Next, the film was applied on the bottom of the Petri dishes and sterilized in 70 % ethanol for 15 minutes and exposed to UV radiation for 15 minutes also. The cells were seeded on Petri dishes with the film and incubated for 24 h. The morphology and viability of the cells were examined by microscope.

4. ANALYSIS OF RESULTS

4.1. Adhesion test

The results of adhesion examination of selected bacteria strain on the steel covered by the film, grade I and II were presented, by showing the amount of bacteria colonies per surface square centimeter (CFU/cm^2 - colony forming unit/cm^2), in Table 1. Due to the temperature variability in a medical vehicle, the research was conducted in the temperature: 25 °C and 37 °C.

Table 1 The amount of bacterial colonies subjected to adhesion on the surface of the researched material [CFU/cm^2] in selected temperature

Density of the bacteria solution [McF]	Grade I				Grade II			
	Pseudomonas aeruginosa ATCC 27853		*Enterococcus faecalis ATCC 29212*		*Pseudomonas aeruginosa ATCC 27853*		*Enterococcus faecalis ATCC 29212*	
	25°C	37°C	25°C	37°C	25°C	37°C	25°C	37°C
0,5	0	0	3	5	0	0	0	0
2	0	0	4	10	0	0	0	20
4	0	0	10	17	0	2	0	20

The growth of the selected pathogens was low or it was not observed at all on the examined surfaces, especially in room temperature, i.e. 25 °C. The film grade II presented lower level of contamination by *Enterococcus feacali* in 25 °C. The samples covered by lacquer coatings restrained the microorganism growth (Table 2) – mainly the RAL 9003 coatings. It was proved in the previous studied [3, 4] that the increase of surface roughness influences the growth of the examined bacteria adhesion. It is also confirmed by the smoothing effect of the film applied on a surface [9].

Table 2 Bacteria colonies on the tested surfaces Amount of bacterial colony [CFU/cm^2] [3]

Density of bacterial cells [McF]	*Pseudomonas aeruginosa ATCC 27853*		*Enterococcus faecalis ATCC 29212*	
	Varnish antibacterial coating RAL 9003, gloss of 60%	Varnish standard coating RAL 9002, gloss 90%	Varnish antibacterial coating RAL 9003, gloss of 60%	Varnish standard coating RAL 9002, gloss 90%
0.5	-	0	-	0
1	0	0	0	0
3	0	-	0	-
5	0	> 150	1	> 180
7	1	-	1	-

Density of bacterial cells [McF]	Plate of plastic - smooth	Plate of plastic - plexi (organic glass)	Plate of plastic - chattered	Plate of plastic - smooth	Plate of plastic - plexi (organic glass)	Plate of plastic - chattered
0.05	> 10	>10	-	>10	>10	-
0.5	> 250	>100	0	> 150	>100	0
1	-	-	0	-	-	0
5	> 300	>150	>500	>250	>150	>600

4.2. Toxicity test

The results of the NHDF observation after the incubation period are shown in figure 1.

a) b)

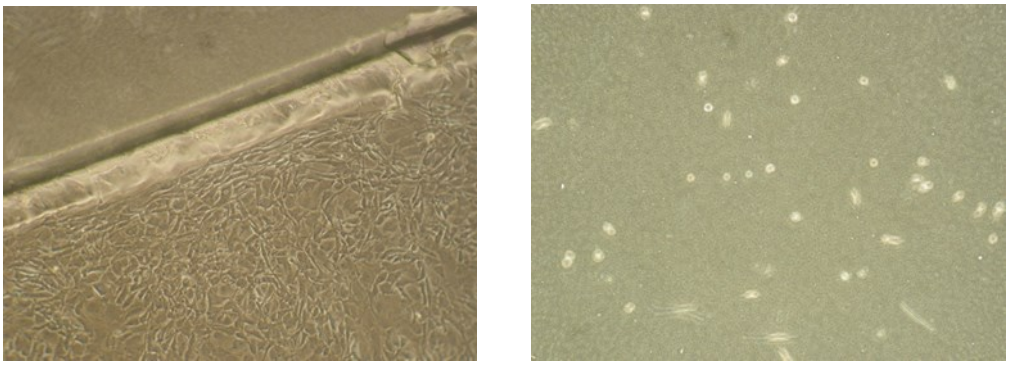

Fig. 1 The image of the edge of the sample – cell culture:
a - NHDF grown with a film of 1x1 cm size, b - NHDF grown directly on a film

The cells were cultivated with the film for 24 h and it was not detected any morphological changes. It may be assumed that the examined films are not toxic for living cells (Fig. 1 a). In order to assess if the cells may be grown on the film surface (Fig. 1b), NHDF were cultivated directly on the surface of Petri dishes cover with film. The test proved their anti-adhesive properties, which prevents growing cells on them.

CONCLUSION

On the basis of conducted laboratory examinations and results discussion the following conclusion can be formulated:

- the surface covered by the film, grade I and II is effectively protected against adhesion of *Pseudomonas aeruginosa* in the examined range of culture density,
- the film, grade II presented lower contamination level of *Enterococcus feacalis,*
- the film do not have a toxic effect on the cell growth.

Modern techniques of sterilization and disinfection of medical vehicles interior are not sufficient to eliminate the risk of infection occurrence. Our study shows new possibilities in terms of surface antibacterial protection.

The results confirm that VentureShield VS 7510 E films may be used for the protection of ambulances interior, as well as in hospital environment. Taking into consideration the previous results, it may be concluded which materials are the best for antibacterial protection, in case of possible film damage. However, some further study is necessary.

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