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Novel test procedure for testing antimicrobially active materials

Abstract: Antimicrobial coatings are typically tested for their activity and effectiveness based on an artificial procedure, the standard JIS Z 2801 (or ISO 22196). The test samples are contaminated with a predetermined concentration of bacteria and covered with a sterile film. Due to the artificial structure of this process, however, there is always the question of the validity in the practical application of these surfaces and how comparable this germ-reducing effect is to currently common disinfection measures.

Therefore, we developed a more realistic test procedure in our laboratory. The test samples are inoculated and incubated at standard room conditions. To compare the antimicrobial activity to standard cleaning procedures also disinfection and wipe controls were performed.

The results show that the antimicrobial test patterns we use have a strong efficacy. This could be shown reproducibly both with the standard method JIS Z 2801 as well as with our new test procedure.

Keywords: antimicrobial, coated samples, disinfectant, disinfectant wipe, germ-reducing, novel test procedure, sterile water
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1 Introduction

Several publications already described the risk of infection via microbially contaminated surfaces for personnel and patients in clinical settings. [1-5] In addition to the cleaning and disinfection measures, antimicrobial surfaces are becoming an additional factor for improving the safety for patient and employees.

Normally this antimicrobial activity is tested in accordance to the established standard JIS Z 2801 (ISO 22196). Due to the artificial structure of this process, however, there is always the question of the validity in the practical application of these surfaces. With this strict specification for temperature and humidity this standard procedure does not reflect the given “real-life” conditions in a OR or ICU. Also with this setting there is no possibility to directly compare this germ-reducing effect to currently common disinfection measures.

2 Material and Methods

A standard test according to JIS Z 2801 (or ISO 22196) was performed for the initial evaluation of the antimicrobial activity of our test samples. Triplicates of test samples were contaminated with a predetermined concentration of bacteria (S. aureus, approx. 5x10⁵ cfu/ml). The agar plates were incubated for 24 hours at 35°C and > 90% humidity after inoculation.

Subsequently the samples were tested using the test method we developed in our laboratory to obtain a more realistic evaluation of the antimicrobial effect. In this approach, 50 μl of a bacterial suspension (S. aureus, 1x10⁸/ml) is applied to a test sample.

After drying (about 15 min) the samples are tested at predetermined time intervals (30 sec to 60 min) by contact plates (Roti-ContiPlate TSA-Letheen; Carl Roth GmbH). The test is carried out at room temperature and at non-adjustable humidity. The agar plates were incubated for 24 hours at 37°C.

The microbial growth was documented and evaluated after the incubation. Uncoated patterns of the same kind were used as a negative control.

In order to assess the effectiveness of the germ reduction, controls were also done with common disinfectants. Therefore, the test samples were covered with a disinfectant wipe for ten seconds or immersed directly in the solution for just one second (Fig.1). As a control, the procedure was performed analogously with sterile water.
3 Results

First a standard test according to JIS Z 2801 was done to obtain preliminary information about the efficacy of our samples. All of our different antimicrobial test specimens showed a strong and reproducible reduction of the germs on the surfaces (> log 3 germ reduction).

Using our procedure, this efficacy of the test samples can also be shown. The applied contamination with gram-positive pathogens (S. aureus) was clearly reduced in some of the samples already at the first sampling immediately after drying (about 15 minutes). An enhancement of this effect could be shown with a longer exposure time. In the case of analogous evaluation criteria for the standard method JIS Z 2801, a strong antimicrobial effectiveness (> log 3 germ reduction) of the test samples can be determined in our test arrangement. The comparison between coated and uncoated test patterns can be seen in Figure 2.

A comparison with common disinfectants indicates a strong decontaminating effect when immersing the test samples in the solutions. After immersion the plates showed no detectable cfu after incubation. However, by placing the disinfectant wipes on the test samples, no significant germ reduction could be shown. In the analogous experiments with sterile water also no germ reduction can be determined (Figure 3).

4 Discussion

The results show that our antimicrobial test samples have a strong efficacy. This could be shown reproducibly both with the standard method JIS Z 2801, as well as with the new test procedure developed in our laboratory. It can be assumed that the new testing procedure reflects a more realistic “real-life” situation, because there are no defined and artificial incubation conditions. This is especially evident in test samples, which show a weak antimicrobial activity in the standard JIS procedure. This activity can no longer be detected under more realistic conditions with our method. Interestingly, we also found in our experiments that there is no germ-reducing effect after placing a disinfectant wipe without friction on contaminated surfaces. Therefore, the mechanical effect of manual cleaning by wiping is essential for the decontamination of the surfaces. Based on these findings our test method is a strong supplement for the existing standard test procedures and allows a more realistic evaluation of antimicrobial coatings in “real-life” situations.

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References


