Prof. Dr. Brigitte König on behalf of DHZ

Silgranit

Hygienic assessment

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30.4.2022

Contents

Questions

In order to assess the SILGRANIT material, which is used for kitchen sinks, from a hygienic point of view, three precise questions were formulated:

1. Is there a sufficient reduction in bacteria after cleaning/disinfecting the SILGRANIT material?

2. Does the SILGRANIT material inhibit microbial biofilm formation?

3. Are the growth of microorganisms on the test material (SILGRANIT) inhibited or are the microorganisms killed over time?

Question 1

Method

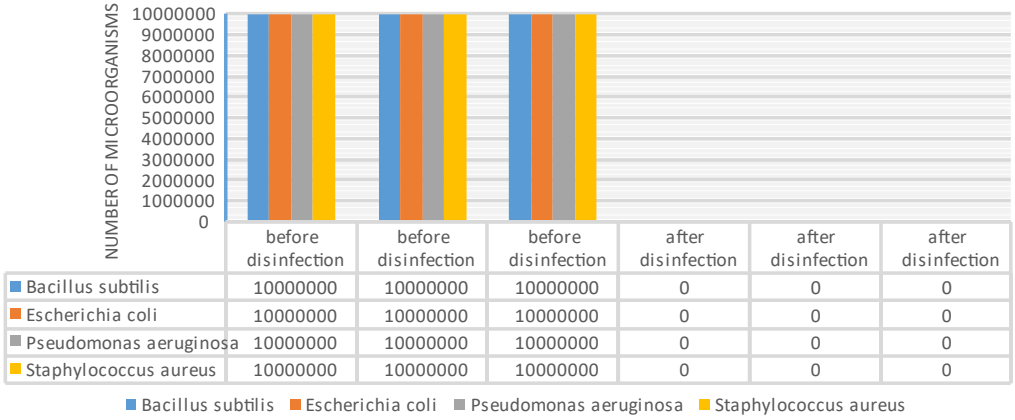
The material was wetted with artificially prepared solutions (in H_2O) containing different concentrations (n=3) of relevant bacteria. The four microorganisms *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* were selected for this purpose. The selected microorganisms are representative of microorganisms with human pathogenic potential. In addition, the selected microorganisms differ in their sensitivity to environmental conditions, disinfectants and growth conditions. The various microorganisms were checked for an optical density (600 nm) of 0.8; 0.4 and 0.2, corresponding to a bacterial concentration of about $1x10^8$/ml; $5x10^7$/ml and $2.5x10^7$/ml.

At the time point 0 (the application of the microorganisms), in two parallel batches (batch A and batch B), 100 µl each of the respective bacterial suspensions were applied with a cotton swab to a SILGRANIT area of about 5x5 cm each. After the drying process the SILGRANIT surface of approach B was treated with an alcohol-free disinfectant (RKI listed) according to the manufacturer's instructions. The SILGRANIT surface of batch A remained untreated (control). The bacteria that remained on the SILGRANIT surface of batch A and batch B were washed away with H_2O and, after making dilution series, plated out quantitatively on agar plates (Brain Heart Infusion Agar [BHI] with 10% Sheep Blood) and after a 24-hour incubation at 36°C counted. The results for the three different starting concentrations are shown in Figures 1-3 (n=3).

Result

The tests have clearly shown that even high concentrations of medically relevant microorganisms can be removed from the SILGRANIT surface using a commercially available alcohol-free disinfectant (RKI-listed).

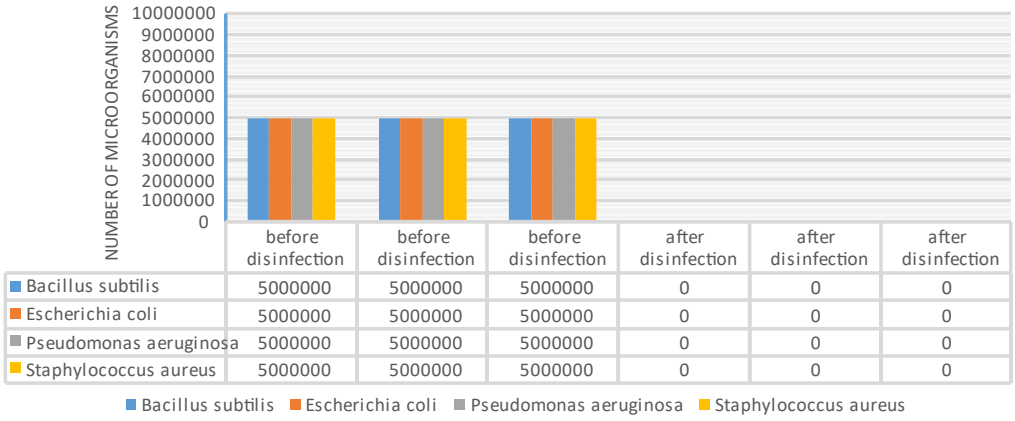
Disinfectability of SILGRANIT



	before disinfection	before disinfection	before disinfection	after disinfection	after disinfection	after disinfection
Bacillus subtilis	10000000	10000000	10000000	0	0	0
Escherichia coli	10000000	10000000	10000000	0	0	0
Pseudomonas aeruginosa	10000000	10000000	10000000	0	0	0
Staphylococcus aureus	10000000	10000000	10000000	0	0	0

Figure 1: Disinfectability of SILGRANIT. A SILGRANIT surface was contaminated with various microorganisms and then disinfected with a commercially available non-alcohol disinfectant. Shown are three experiments carried out independently of one another with an initial concentration of the microorganisms of OD 0.8.

Disinfectability of SILGRANIT



	before disinfection	before disinfection	before disinfection	after disinfection	after disinfection	after disinfection
Bacillus subtilis	5000000	5000000	5000000	0	0	0
Escherichia coli	5000000	5000000	5000000	0	0	0
Pseudomonas aeruginosa	5000000	5000000	5000000	0	0	0
Staphylococcus aureus	5000000	5000000	5000000	0	0	0

Figure 2: Disinfectability of SILGRANIT. A SILGRANIT surface was contaminated with various microorganisms and then disinfected with a commercially available non-alcohol disinfectant. Shown are three experiments carried out independently of one another with an initial concentration of the microorganisms of OD 0.4.

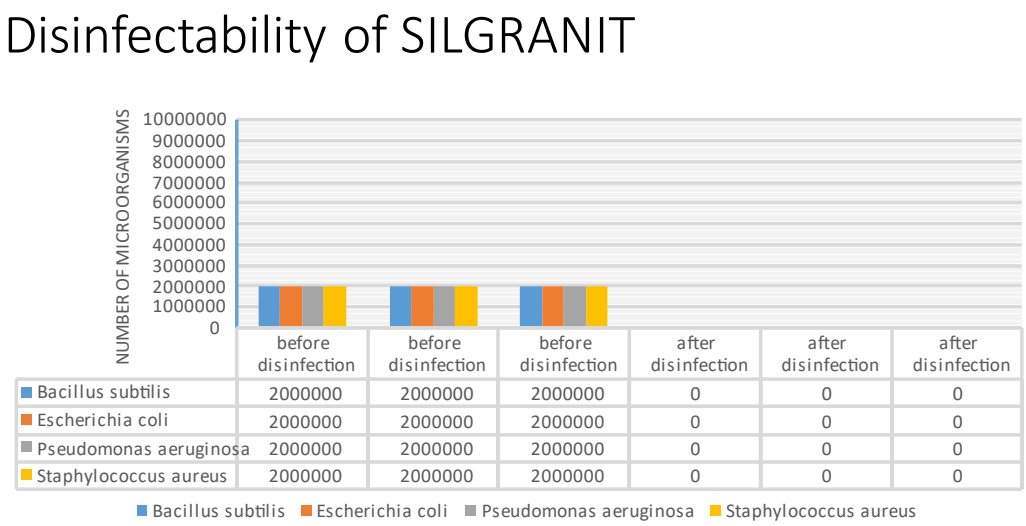


Figure 3: Disinfectability of SILGRANIT. A SILGRANIT surface was contaminated with various microorganisms and then disinfected with a commercially available non-alcohol disinfectant. Shown are three experiments carried out independently of one another with an initial concentration of the microorganisms of OD 0.2.

Question 2

Method

The material was wetted with artificially produced solutions of two different biofilm formers. A biofilm-forming *Staphylococcus epidermidis* and a biofilm-forming fungus of the species *Candida albicans* were chosen as microorganisms. Both microorganisms were used at a concentration of $1x10^7$/ml. The biofilm formation of both microorganisms on the SILGRANIT was compared with the biofilm formation on a plastic surface as a standard material for biofilm formation after 24h incubation at 36°C. The results are summarized in Figure 4.

Result

The investigations have clearly shown that the SILGRANIT surface prevents biofilm formation.

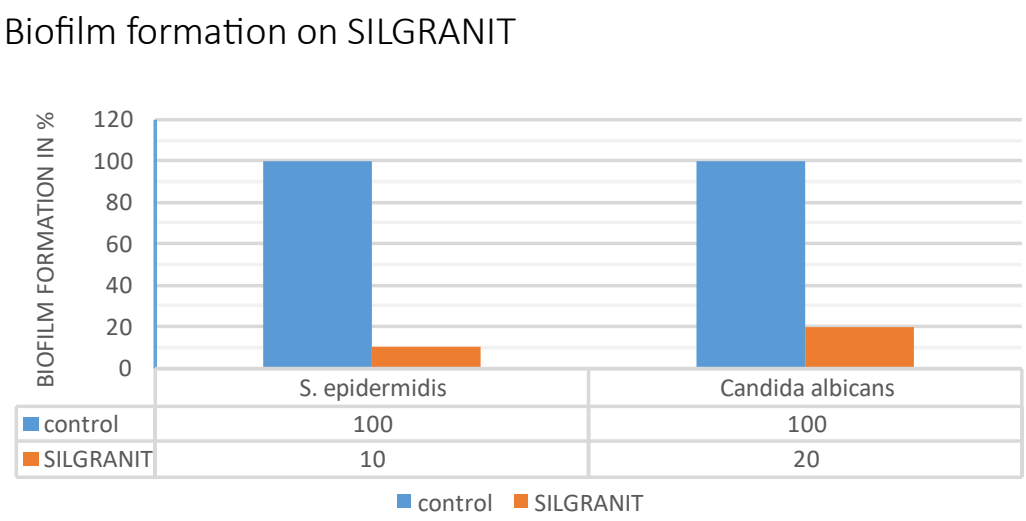


Figure 4: Biofilm formation of Staphylococcus epidermidis and Candida albicans on SILGRANIT. A SILGRANIT surface was contaminated with a) Staphylococcus epidermidis and b) Candida albicans in a concentration of $1x10^7$/ml. As a control (biofilm formation), both microorganisms were placed on a plastic surface (control). After 24 hours of incubation at 36°C, biofilm formation was determined quantitatively. The biofilm formation on the plastic surface is set to 100%. The mean value of three analyses carried out independently of one another is shown.

Question 3

Method

The material was wetted with artificially prepared solutions (in H_2O) containing different concentrations of relevant bacteria. The four microorganisms *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* were selected for this purpose. The selected microorganisms are representative of microorganisms with human pathogenic potential. In addition, the selected microorganisms differ in their sensitivity to environmental conditions, disinfectants and growth conditions. The various microorganisms were checked for an optical density (600 nm) of 0.8; 0.2 and 0.002, which corresponds to a bacterial concentration of about $1x10^8$/ml; $2.5x10^7$/ml; and corresponds to $2.5x10^6$/ml.

At the time point 0 (application of the microorganisms) and after defined time intervals (approximately 1 hour, 4 hours, 8 hours, 24 hours, 48 hours), the bacteria still alive were cultured. To do this, the remaining bacteria on the SILGRANIT surface were washed away with H_2O and, after dilution series had been prepared, plated out quantitatively on agar plates (Brain Heart Infusion Agar [BHI] with 10% sheep blood) and counted after 24 hours of incubation at 36°C. The results for the four different starting concentrations are shown in Figures 5-8 (n=3).

The growth rate of the bacteria in water and in complete medium (brain heart infusion medium) are shown in Figures 9 and 10 using *Escherichia coli* as an example.

Result

The investigations have clearly shown that there is a significant rate of death of medically relevant microorganisms on the SILGRANIT surface. The death rate depends both on the length of time and on the individual microorganism.

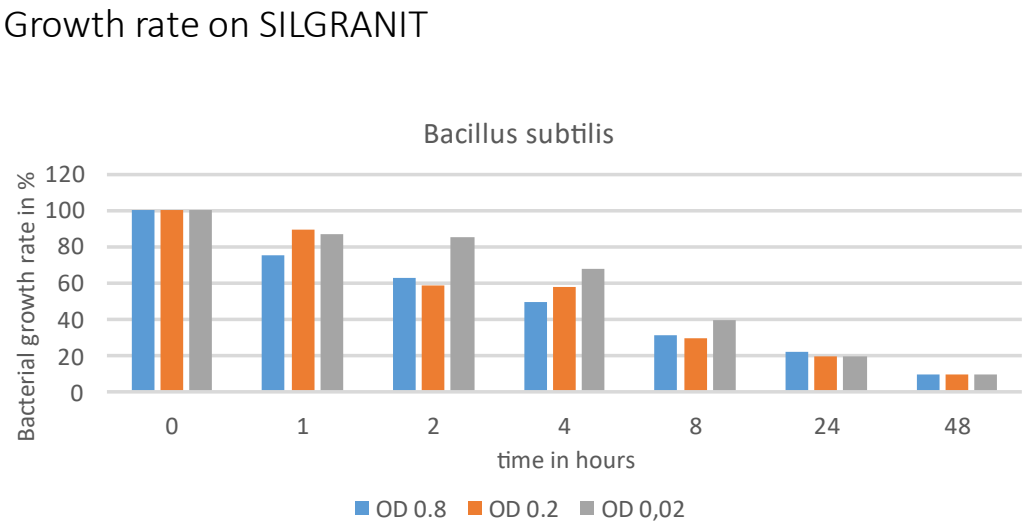


Figure 5: Growth rate of Bacillus subtilis on SILGRANIT. A SILGRANIT surface was contaminated with Bacillus subtilis in various concentrations (OD 600nm 0.8; 0.2; 0.02). After defined time intervals (1, 2, 4, 8, 24, 48 hours), the surviving bacteria were quantitatively determined. The initial concentration is set to 100%. The mean value of three analyses carried out independently of one another is shown.

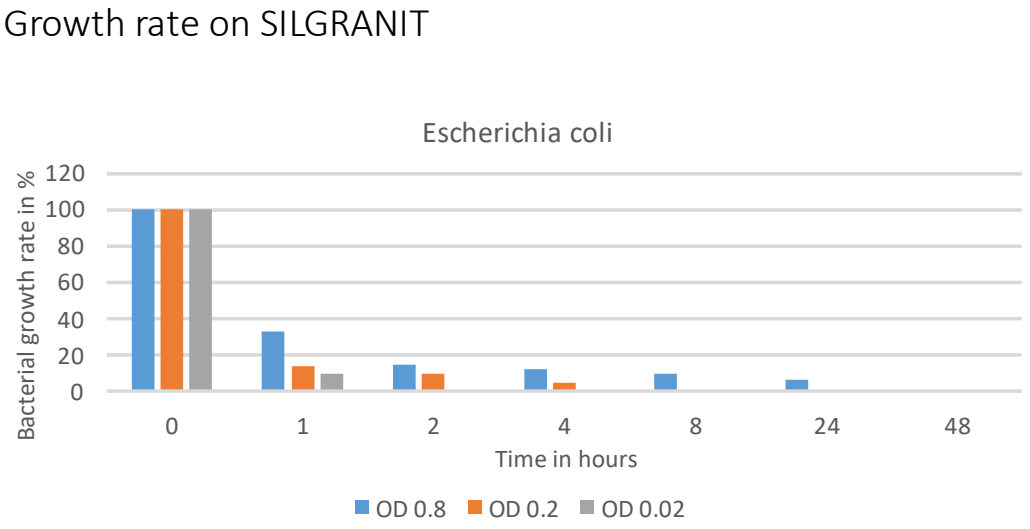


Figure 6: Growth rate of Escherichia coli on SILGRANIT. A SILGRANIT surface was contaminated with Escherichia coli in various concentrations (OD 600nm 0.8; 0.2; 0.02). After defined time intervals (1, 2, 4, 8, 24, 48 hours), the surviving bacteria were quantitatively determined. The initial concentration is set to 100%. The mean value of three analyses carried out independently of one another is shown.

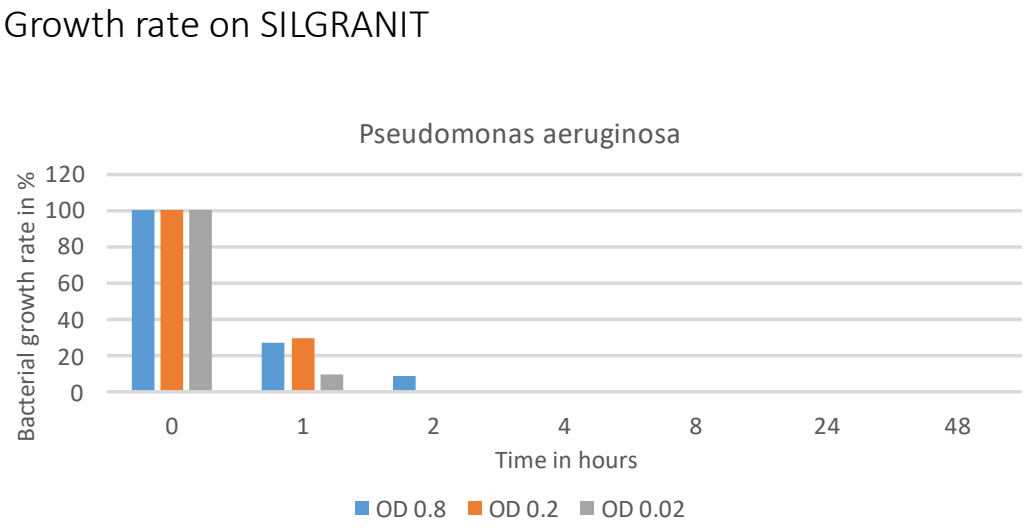


Figure 7: Growth rate of Pseudomonas aeruginosa on SILGRANIT. A SILGRANIT surface was contaminated with Pseudomonas aeruginosa in various concentrations (OD 600nm 0.8; 0.2; 0.02). After defined time intervals (1, 2, 4, 8, 24, 48 hours), the surviving bacteria were quantitatively determined. The initial concentration is set to 100%. The mean value of three analyses carried out independently of one another is shown.

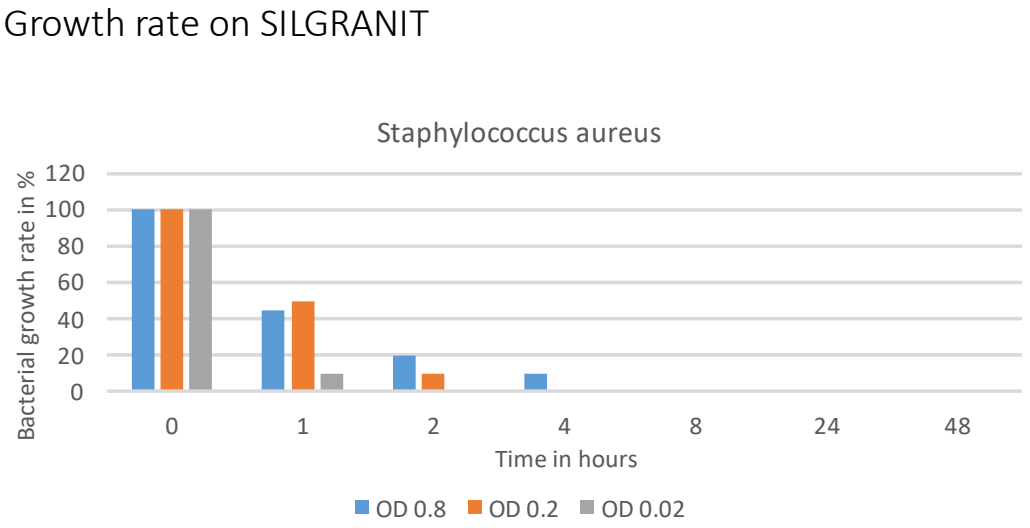


Figure 8: Growth rate of Staphylococcus aureus on SILGRANIT. A SILGRANIT surface was contaminated with Staphylococcus aureus in various concentrations (OD 600nm 0.8; 0.2; 0.02). After defined time intervals (1, 2, 4, 8, 24, 48 hours), the surviving bacteria were quantitatively determined. The initial concentration is set to 100%. The mean value of three analyses carried out independently of one another is shown.

Growth rate in complete medium

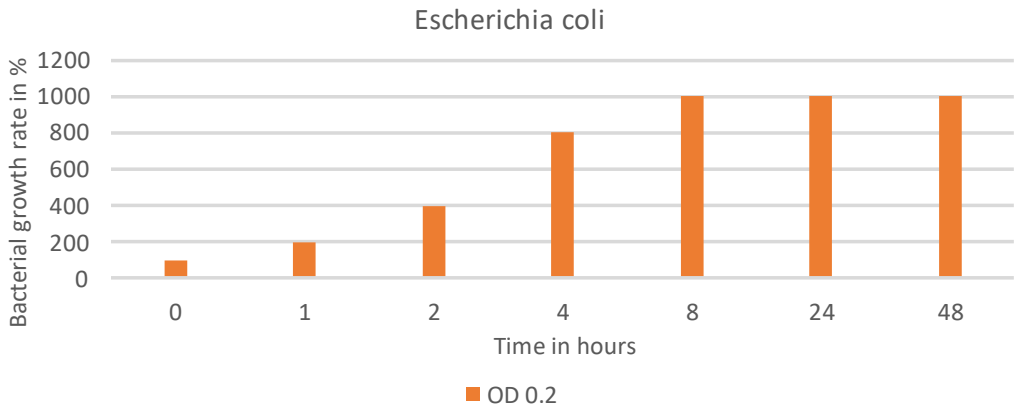


Figure 9: Growth rate of Escherichia coli in complete medium (brain heart infusion). An Escherichia coli starting solution with an OD (600 nm) of 0.2 was selected. After defined time intervals (1, 2, 4, 8, 24, 48 hours), the bacterial concentration was determined quantitatively. The initial concentration is set to 100%. The mean value of three analyses carried out independently of one another is shown.

Growth rate in H_2O

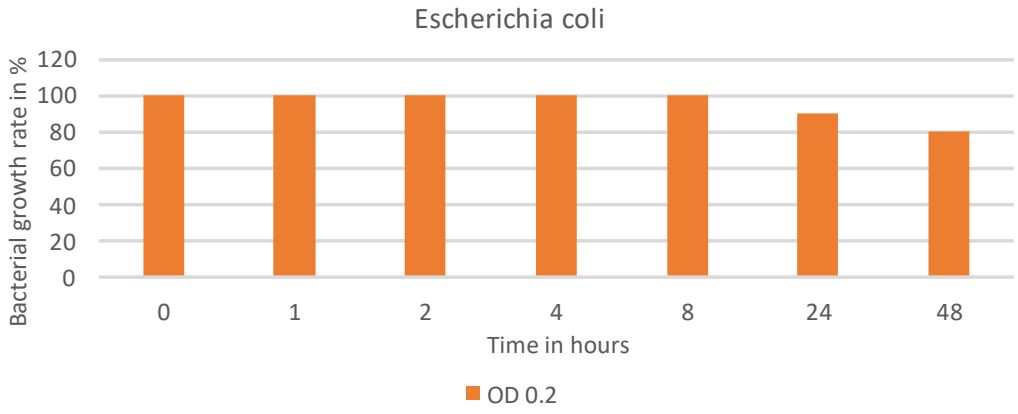


Figure 10: Growth rate of Escherichia coli in H2O. An Escherichia coli starting solution with an OD (600 nm) of 0.2 was selected. After defined time intervals (1, 2, 4, 8, 24, 48 hours), the bacterial concentration was determined quantitatively. The initial concentration is set to 100%. The mean value of three analyses carried out independently of one another is shown.

Summary

After the SILGRANIT material has been disinfected, there is a sufficient reduction in bacteria in the case of medically relevant microorganisms.

The SILGRANIT material inhibits the formation of microbial biofilms.

Microorganisms that are on the SILGRANIT material are inhibited in growth and killed over time #.

The "killing" mechanism is not based on an active killing by the SILGRANIT material, but rather on the fact that the microorganisms on the SILGRANIT material have no basis for survival.