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An innovative, preventive acting "bioinspired" antimicrobial surface based on peptides for space and Earth

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Introduction:

Using antimicrobial surfaces is a well suited approach to prevent and reduce microbial loads in sensitive areas – avoiding by this endangering of human health, health of organisms e.g. in bioregenerative life-support systems and technical equipment.

- In spaceflight being beneficial
 - in confined environments in LEO and during exploration activities.
 - to support breeding activities of e.g. algae in bioreactors and for biological experiments.
 - to meet the COSPAR planetary protection requirements.
- On Earth being beneficial during medical activities/ food handling, in swimming baths, bathrooms, public transport etc.. For its dedicated use, they must be free of any releasing toxic substance (e.g. silver), otherwise higher non-target organisms would be negatively affected. Resistances of bacteria to these toxic substances and rather unspecific acting are further disadvantages of them.

Goal of the project BALS (ESA):

Using antimicrobial peptides (AMPs) from nature (e.g. from frog skin), immobilized on surfaces, are a suited alternative. AMPs are low toxic, acting specifically, and long-term use of AMPs - especially in confined habitats - should not induce resistances in bacteria. Wide range of different AMPs were identified so far.

The goal of the project BALS (<u>B</u>ioinspired <u>a</u>ntimicrobial <u>l</u>acquer for <u>s</u>pace) was the development of an antimicrobial acting lacquer based on peptides, including verification in a series of space relevant tests.

Antimicrobial working of AMP:

• AMPs (Fig. 1) can penetrate bacterial membranes, thus killing bacteria by destroying the electrochemical gradient or leading to an uncontrolled permeation of water out of the bacteria cell (Fig. 2). Binding to periplasmatic receptors/intracellular molecules could lead to a severe disturbance of vital functions.

Lacquer Concept:

- Full bioinspired antimicrobial lacquer was developed as a 2-layer concept: a) Basic lacquer layer, containing amines for linking AMPs
 - b) AMP-laver

* Source: Jenssen, H. et al. Clin. Microbiol. Rev. 2006, 19, 491-511 ** Source: http://www.farmfak.uu.se/farm/farmfyskem-web/

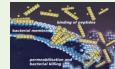


Fig. 1: Exemplary AMP secondary structures*

Fig. 2: AMP acting **

Test set- up and Test Results:

- Antimicrobial activity against S. cohnii and E. coli (after 96 h, compared to a reference lacquer without AMPs) was shown according to a modified ISO 22106 test procedure: E. coli abundance was reduced by 77 %, S. cohnii (see Fig. 3) by 100 %.
- Adhesion strength on space relevant stainless steel and aluminium was demonstrated in a ECSS-Q-70-13A-test series.
- Absence of effects on higher organisms and the environment was shown in a laboratory aquatic biological multispecies test system (AquaHab®) with fish, snail, crustacean and plants (7 day incubation with leachate water (Fig. 4).

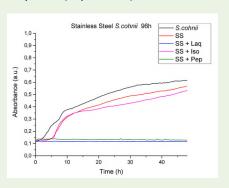


Fig. 3: Absorbance/ abundance of S. cohnii after 96 h incubation on bioinspired antimicrobial lacquer based on peptides (SS+Pep).

Remark w.r.t. lacquer reference samples:
(1)SS+Laq = lacquer sample, incl. amines, thus of course acting antimicrobial.
(2)SS+ISO= "blocked" lacquer samples: amines were blocked by isocyanate = representing the final situation w.r.t. exposure to the real environment of the final lacquer (AMPs are also blocking amines), thus used as the lacquer control group.

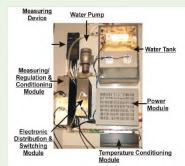


Fig. 4: AquaHab-System;

Effect parameter:
- Photosynthetic +
respiratory activity
- Plant and fish growth
- Fish behaviour
- Lethality of all

 Lethality of a organisms

Outlook:

With demonstrated feasibility and use (TRL 4) by tests, all preconditions are now given for the <u>further development and qualification</u> until a full commercial exploitation, ready to be used in several application fields in space and on Earth.

Acknowledgement:

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