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A Self-Assembling Peptide with the Potential of Non-Invasive Regeneration of Early Caries Lesions

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Introduction

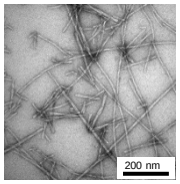


Fig. 1: Transmission Electron Microscope (TEM) image of the self-assembled peptide P11-4

Natural remineralization of tooth material in demineralized small subsurface lesions provides a challenging purpose. Currently early dental caries are mainly treated by mechanical techniques, like dental fillings or tooth extraction, or by applying protective barriers to the tooth surface, like varnished, containing e.g. fluoride.

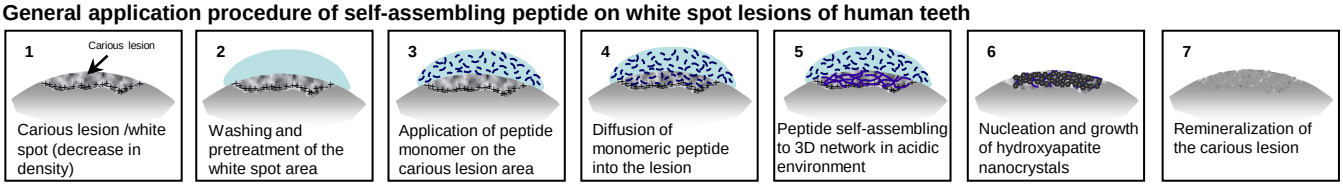
As a result research is concerned with non-invasive regeneration methods of early caries in enamel/ white spot lesions. The scientific breakthrough was achieved by applying a self-assembling peptide (SAP) to the cavities. The fully synthetic peptide forms a 3D supramolecular network *in situ* and is assumed to trigger nucleation of calcium phosphate nanocrystals, resulting in a biomimetic mineralization and therefore a regeneration of demineralized cavities [1].

In this project, the diffusion, assembly and remineralization process of the peptide P11-4 were studied in artificially demineralized cavities of human teeth [2].

Methods

Supramolecular peptide network were detected by transmission electron microscopy (TEM). To stabilize the self-assembled peptide in artificially generated white spot lesions of human teeth, Critical Point Drying (CPD) was performed and subsequently the cavities were visualized by scanning electron microscopy (SEM). Furthermore matrix-assisted laser desorption/ionization (MALDI-TOF) measurement was performed on peptide treated white spot lesions. The remineralization status was detected by micro Computer Tomography (μCT).

General application procedure of self-assembling peptide on white spot lesions of human teeth



Results

TEM pictures represented self-assembled fibrillar protein structures (**Figure 1**-see above). SEM pictures of the CPD process in artificially drilled deepenings of human teeth showed, that the peptide completely diffused into the enamel cavity. In comparison with a non-peptide treated reference (**Figure 2 A**), the presence of the organized peptide was visible by a gelled layer covering the artificially drilled deepening (**Figure 2 B**). Additional experiments with MALDI-TOF showed, that the peptide remained in artificial white spot lesion in a stable and unimpaired state **(Figure 3).** CT measurements represented a successful remineralization status *in vitro* in white spot lesions of human teeth according to the test duration **(Figure 4).**

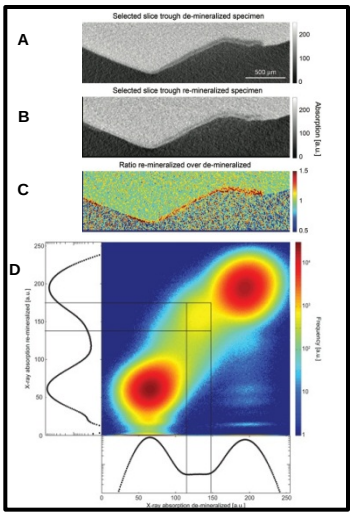


Fig. 4: μCT images of a selected slide through a specimen either after demineralization **(A)** and after peptide treatment and remineralization **(B)**. **(C)** shows the ratio of remineralized over demineralized specimen. **(D)** Data plot of specimen absorption.

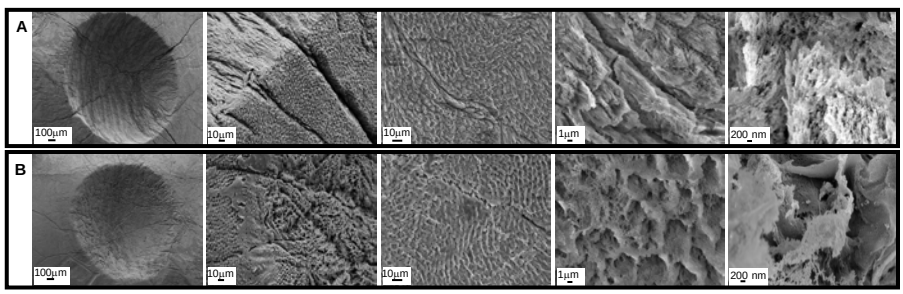


Fig. 2: TEM images of artificially drilled deepening in human enamel: **(A)** without peptide (reference) and **(B)** with peptide under self-assembling conditions after CPD.

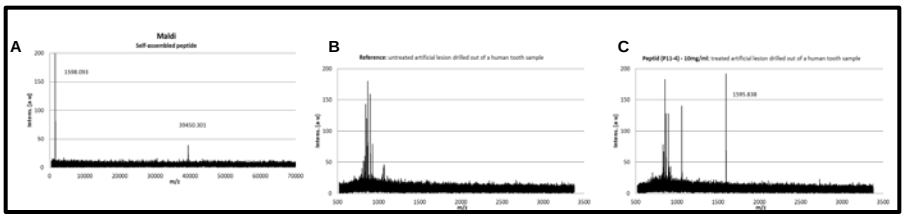


Fig. 3: MALDI-TOF results: **(A)** self-assembled peptide, **(B)** Reference: untreated artificial lesion drilled out of a human tooth sample without peptide treatment, **(C)** Artifical lesion drilled out of a human tooth sample after application of P11-4 (10mg/ml) and incubation for 2 weeks in remineralization buffer .

Conclusion

It was successful shown, that the peptide P11-4 is able to induces biomimetic mineralization and therefore shows a big non-invasive regeneration potential.

Outlook

Further work will be dedicated to the thermodynamic assembly rate by small angle X-ray scattering (SAXS), Raman spectroscopy, Confocal microscopy and enzyme-linked immunosorbent assay.

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