

The EU-project “Nanosafe2: safe production and use of nanomaterials” integrates the expertise of the groups Functional Peptides, Nanoparticle Technology and Technical Support NT to achieve the ambitious goal of engineering a biosensor capable of detecting airborne nanoparticles as well as dissolved nanoparticles.

Cross-links:

Functional Peptides

Nanoparticle Technology

In principle peptides that bind to TiO<sub>2</sub> are selected from libraries of 100 million peptide species presented on bacterial flagella. These peptides are immobilized to a biosensor based on quartz crystal microbalance (QCM) or surface plasmon resonance (SPR) technology to detect nanoparticles which are dispersed in a liquid phase passing over the sensor surface.

In detail we investigated the possibility of capturing and detecting nanoparticles immersed in aqueous solvent with techniques based on specifically binding peptides. To this end the group has selected peptides that bind to TiO<sub>2</sub> coated wafers. 31 of these peptides have been sequenced so far. In order to identify characteristic binding motifs, bioinformatics and statistical analyses with these sequences were carried out that indicate possible common features (e.g. in the amino acid composition or consensus sequences) of these peptides. It is intended to compare binders with not binding peptides, and combine the different approaches (i.e. screening of libraries and bioinformatic analysis) to optimally exploit the full strength of each method.

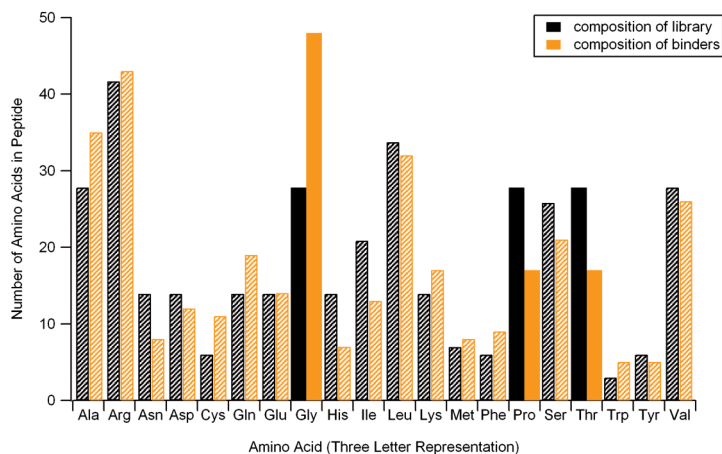


Figure 1: Statistical analysis of peptides that comprised bacterial flagella with the ability to bind to TiO<sub>2</sub>-wafers. Significant deviations between the composition of the library and the binders are represented by bold columns.

We also started to establish “CIS-display”, a novel in vitro selection technique that allows for larger peptide libraries to select peptides from. “CIS-display” is based on a DNA construct that is subsequently bound “in cis” to its own translation product while being processed using a coupled in vitro transcription/translation system. “In cis” describes the special property of the binding process; the protein is fused to the same DNA molecule from which it originated, thereby linking the phenotype with its genotype.

In order to realize the detection of airborne NPs using this liquid based sensor, a device that transfers NPs from air into a solution is being constructed.

We developed two alternative experimental designs for the transfer of particles from air to aqueous solution. According to the priorities set in the first six months, the chosen designs are based on washing flasks and sprayers, respectively. To calibrate the transfer efficiency of this device an instrument that emits a defined amount of NPs was constructed and in collaboration with the external project partner HVBG-BIA tested to assure proper functionality.



Figure 2: Set-up of the air-to-water transfer device with two washing flasks (background). In the foreground a NP-cloud generator where  $\text{TiO}_2$ -NPs (white powder on bottom) are catapulted into the air by vibrations of a loudspeaker.

In an effort to characterize commercially available  $\text{TiO}_2$  nanoparticles used in our experiments, we employed Transmission Electron Microscopy and discovered that the particles form agglomerates of the order of 100 nm to 1  $\mu\text{m}$ . A very fast and inexpensive method for the detection of particles in aqueous solution is UV/VIS-absorption spectroscopy. Hence, we made preliminary spectroscopic measurements of a dilution series of  $\text{TiO}_2$  in water.

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Cooperation:

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CEA, Saclay, France (coordinator)

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