# Selective Adhesion Behaviour of Genetically Engineered Peptides for Chemical Force Microscopy and Nanoparticle Capturing

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**Abstract**

Genetically engineered peptides for inorganics (GEPIs) are a promising approach to provide highly selective interactions between surfaces or complex structures and target entities, such as molecules or nanoparticles. Bottom-up construction or patterning of surfaces and interfaces can be achieved by employing selectively binding building blocks [1]. Furthermore, in sensing applications the selectivity towards a particular analyte can be increased by functionalising the sensor surface with selectively binding molecules [2]. Thus GEPIs are versatile building blocks for a range of applications, from tissue engineering, to biosensing, nanomaterials, and water remediation.

Atomic force microscopy (AFM) allows for force measurements between a sharp tip and a target surface. Spatial variations in the molecule-surface interaction force can be measured by covalently attaching molecules to the AFM tip and recording force-distance curves over an array of spots on a surface. In the present study, a gold coated AFM tip was functionalised with peptides via the thiol group of their terminal cysteine. Using a sequence from Naik et al. [3], the peptide C-terminal was designed to selectively bind silica.

Gold coated AFM cantilevers with a normal spring constant in the range of 10 - 100 pN, as determined via the thermomechanical method, were used to measure the adhesion force between tailor-made peptides and inorganic surfaces. Prior to the AFM measurements, the cleaned cantilevers were dipped into a 1 mM aqueous solution of the peptides. A Cypher AFM system from Asylum Research was employed. All AFM measurements were taken in $NaH_2PO_4$ solutions adjusted to pH6 which contained trace amounts of the surfactant Tween20. Typically, force maps were measured over an area of 20x20 microns$^2$. Afterwards, the area was scanned in contact mode to image spatial variations in the frictional/lateral force acting between tip and sample surface (lateral force microscopy, LFM) [4].

The substrates for AFM study consisted of thermally oxidised Si chips which were patterned by thermally evaporating Au whilst the surface was partly masked. A metal grid with a pitch size of ~12.7 microns was used as a mask. Evaporation produced periodic square areas of metal on the surface, corresponding to the pattern of the grid (Figure 1a).

A map of the adhesion force measured with a tip that had been functionalised with a silica-binding peptide is shown in Figure 1c. Clearly, a lattice can be seen with a lower adhesion force in the square areas and a higher adhesion force in the areas between the squares. The adhesion force is given by the pull-off force of the force-distance curves measured on an array of 32x32 spots. Approximately, the average adhesion force was ~44 pN on Au coated areas and ~100 pN on silica areas. Also the lateral force was found to be increased on silica (Figure 1b).

As a proof-of-concept application, the binding of silica nanoparticles from a suspension was demonstrated. Prior to exposure to the suspension, a gold surface was functionalised with silica-binding peptides. Figure 2a shows an SEM micrograph of the surface with 100 nm particles bound to it. Control experiments shown in Figures 2b and 2c confirmed that the binding is selective for silica.

In conclusion, it can be stated that the selective binding of tailor-made peptides has been demonstrated by AFM force-distance curve measurements. Also, LFM has shown an increased friction on silica with respect to gold. These findings have been supported by control experiments using alternative tip functionalisations. As a proof-of-concept, the peptides were utilised to selectively bind silica nanoparticles on a gold surface.

**References**

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[5] The authors are grateful to A.W. Booker for preparation of the test samples and to N.C. Bell for the synthesis of silica nanoparticles. They thank the Technology Strategy Board for co-funding through a Feasibility Study for Responsible Development of Nanoscale Technologies. This work has been funded by the National Measurement System of the UK Department for Business, Innovation and Skills through the Chemical and Biological Metrology Programme.

**Figures**

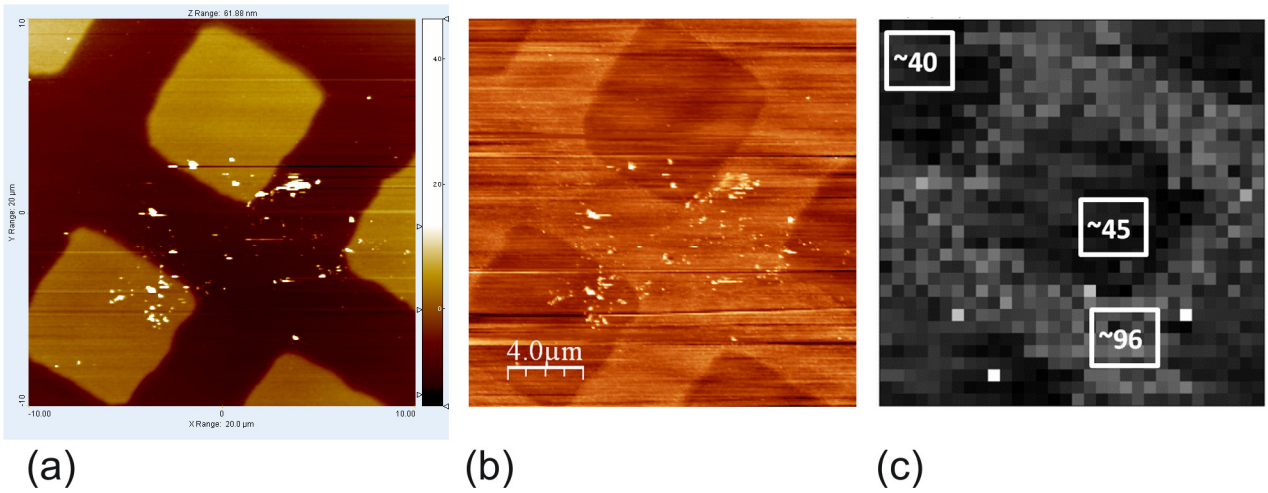


**Figure 1.** Results of AFM measurements using an AFM tip functionalised with silica-binding peptides. (a) Topography image of an array of Au squares on a silica surface. The scan width is 20 microns. (b) Corresponding lateral force image measured when scanning from left to right. (c) Adhesion map of another area of the same surface. The numbers given are in units of pN and are the average values of the marked areas. The pixel resolution of the map is 32x32.

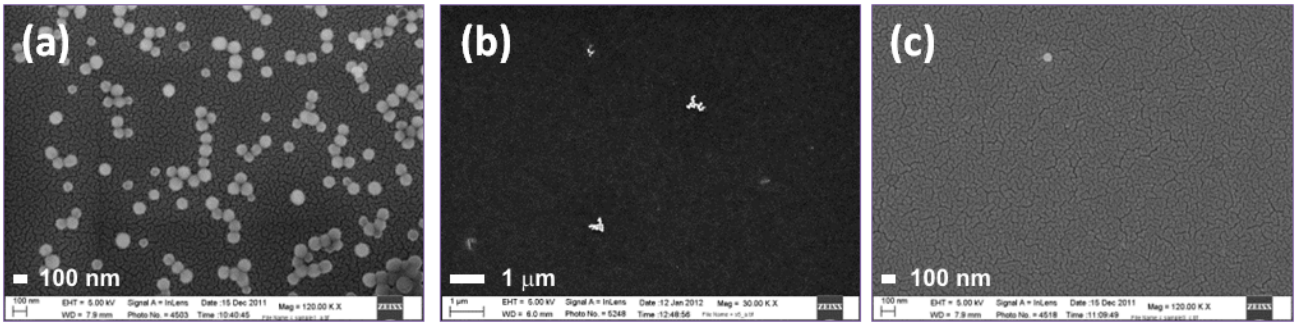


**Figure 2.** Selective binding of silica nanoparticles. SEM micrographs of a polycrystalline gold surface functionalised with (a) silica-binding peptides and incubated in silica nanoparticle solution; (b) silica-binding peptides and incubated in a silver nanoparticle solution and (c) silver-binding peptides and incubated in a silica nanoparticle solution.