In situ imaging of targeted delivery of drugs to living cells with AFM

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ABSTRACT

Ambient atomic force microscopy (AFM) performed in a closed fluid cell provides a convenient method to investigate the interaction of (functionalized) nanoparticles and living cells. Such studies are of interest for targeted drug delivery in medicine.
Nanoparticles were characterized regarding their mechanical properties and the quality of the surface modification. Cells were grown in a monolayer and investigated in buffer solution with contact and intermittent contact mode AFM.
Given these promising initial results two lines of research are going to be followed:

- A time series of the uptake of the nanoparticles by the cells shall be recorded.
- Cantilevers functionalized with the surface-modified nanoparticles shall scan the living cell surface to determine any site specific binding forces.
  With this method we might succeed in providing a quantitative analysis of binding strength correlated with the type of functionalization of the respective nanoparticles.

Drug Targeting

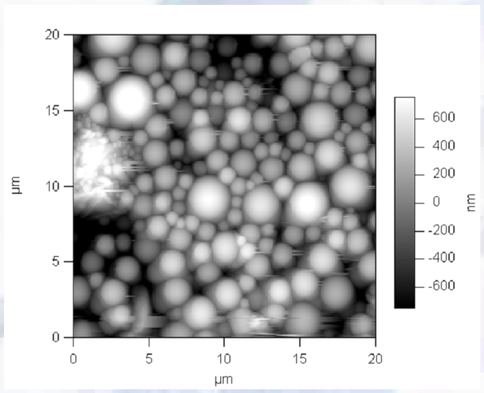
Why?
Many innovative therapies involve the application of highly potent drugs that would cause severe side effects when distributed over the whole body. Only a few percent of the applied dose reach the diseased tissue.

How?
The active component is encapsulated in a biocompatible and biodegradable carrier, which can be coated with a layer of specific recognition molecules (*functionalized*).

Benefits?

- Drugs can be delivered precisely to a localized target.
- Dramatic reduction of the required dose.
- Reduction of adverse effects
- Applicability of unstable biomolecules

Nanoparticles

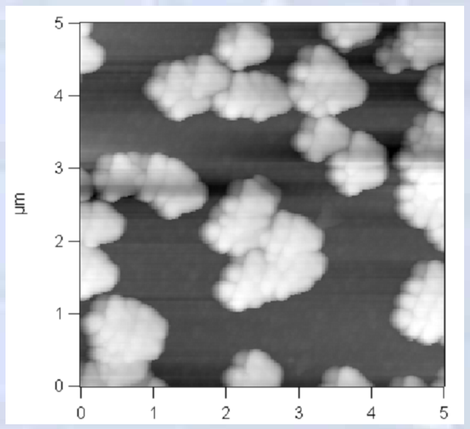


PLGA particles of sizes between 200nm and 2µm, imaged in contact mode AFM.

Nanoparticles made of poly(lactic-co-glycolic) acid (PLGA) are biocompatible and biodegradable. [1] The ratio of lactic acid to glycolic acid determines degradation time and the release profile. Therefor they are promising candidates as carriers for targeted drug delivery. The particle uptake efficiency is reported to significantly depend on the particle size [2]. Therefor precise production and quality control is required.

Applications for AFM

Characterization of nanoparticles



Uncoated nanpoarticles of 200nm diameter form problematic agglomerates.

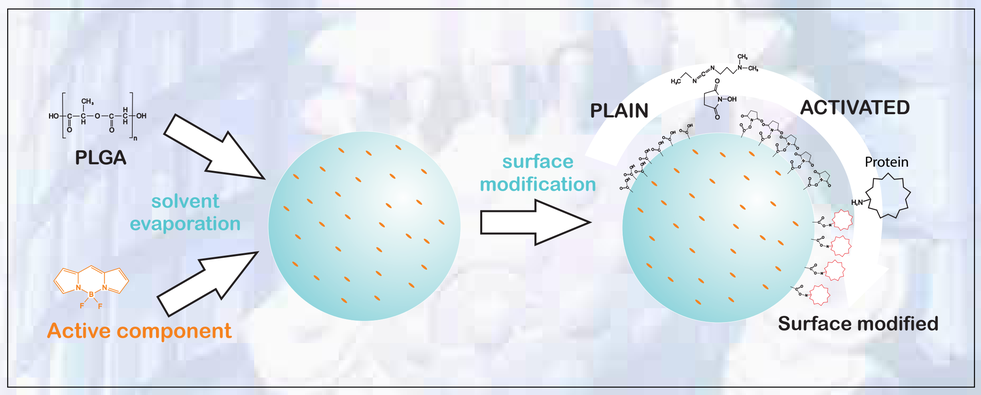
Determine:
Size and shape by contact or intermittent contact mode imaging.

Mechanical properties by force mapping.

Thickness and homogenity of the coating by force mapping and phase imaging.

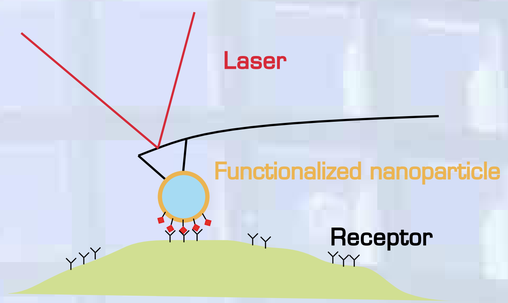
Functionalization

To enable a specific delivery to the site of interest, nanoparticles can be surface modified with different ligands. Different chemical protocols can be applied to obtain a covalent attachment of proteins or other biomolecules. This enables targeting or stealth effects and alters the distribution of the particles within the body.



Functionalization process: The active components are encapsulated in PLGA nanoparticles by solvent evaporation technique. In a further step specific receptor proteins are covalently bound to the surface. With these ligands, the nanoparticle will bind to a certain type of cell only. [3]

Particle binding strength



A functionalized nanoparticle, glued to the tip of the cantilever, is used to probe for site specific surface interactions.

Particles, functionalized with different receptors will be glued to a cantilever.

This probe will be pressed softly onto the cell surface and rectracted after a short timespan for the receptors to bind.

The unbinding forces will be recorded by force mapping over the cell surface. [4]

These issues will be addressed within the context of an interdisciplinary study by physicists and cell biologists.

References

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